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ANTIOXIDANT ACTIVITY, CHEMICAL COMPOSITION AND MORPHOLOGICAL PROPERTIES OF GREEN AND ROASTED COFFEE BEANS AS AFFECTED BY THE MICROWAVE ROASTING METHOD

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Abstract

The purpose of the present study was to explore the influences of microwave heating on the physicochemical composition and antioxidant activity of coffee. Indeed, coffee beans were roasted at different levels of power (350, 500 and 650 W) and for different time periods (5, 10 and 15min) in a microwave oven. Then, green and roasted beans oils were extracted using a Soxhlet extractor. Treated beans analysis revealed no changes in in fatty acids and mineral elements profiles. Furthermore, coffee oil contains a significant amount of linoleic acid (44,82±0,12 g/100g), palmitic acid (32,17±0,20 g/100g) and oleic acid (12,05±0,09 g/100g). Also, among all minerals studied, potassium was the highest (6179,01±24.17 mg/Kg), followed by calcium and magnesium with concentrations 2554,09±45. 36 mg/Kg and 1651,31±23.20 mg/Kg, respectively. However, our results demonstrate that microwave roasting affect oil content, morphological characteristics, TPC, TFC and TAC of coffee. The highest oil yield was found at 500 w/ 15 min (14,94±1,07 g/100g). Roasting increased the weight loss, length, width and thickness of treated coffee beans with roasting power and time. Nevertheless, a significant decline of antioxidant activity (from 78,48±0,55% to 11,08±0,36) was observed after roasting. The optimum level of TPC (48,33±0,17 mg GAE/g) and TFC (56,19±7,85 mg QE/g) was achieved in roasted samples (350 w/5min) compared to unroasted ones (41,45±2,73 mg GAE/g and 52,16±4,99 mg QE/g).

Keywords: *coffee beans, microwave, antioxidant activity, fatty acids, coffee oil.*

Introduction

As one of the most popular beverages consumed in the world, coffee is known for its sensorial characteristics and its unique physiological benefits [1]. The notable sensory attributes of roasted coffee are formed by the mixture of many volatile compounds that are produced during the roasting process [2]. In addition, coffee roasting is an important industrial process in which coffee is submitted to many physical and chemical modifications [3]. In order to measure the impacts on bioactive compounds in coffee beans, various roasting methods, such as traditional oven and microwave have been already studied [4-5]. However, microwave roasting of coffee beans has been demonstrated to have many positive results for the retention of volatile aroma compounds [6], speed of performance, energy efficiency, precise process control, and quicker start-up and shut-down times compared to the conventional roasting process [7].

In the convection and pan roasting processes, it is possible for the exterior surface of the beans to be over roasted without the core being fully exploited. In the microwave oven process, the opposite occurs, and then the core temperature of the beans is higher than the surface temperature. Thus, the microwave method should produce uniformly roasted beans with no burnt surface and full flavor [8]. As a result, microwave heating has been consistently found to retain quality characteristics to be better than conventional heating [7-8] The composition of the roasted coffee depends on several parameters; microwave power, roasting temperature, roasting time, etc, thereby having an affect on the desired product proprieties [7, 9-10]. Hence, the purpose of the present research was to determine the effect of microwave roasting (power and time) on the oil yield, fatty acid composition, mineral element composition, total antioxidant capacity, polyphenols, flavonoids, and morphological attributes of coffee beans.

Material and methods

Roasting process

Green coffee beans were obtained in a coffee shop In Agadir, Morocco. Then, they are separated into two groups. The first one was control group (green or unroasted beans) which analyzed without roasting. However, the second group of coffee beans were roasted using a microwave oven (MWP 253 – MWP 254, Whirlpool) at 350, 500 and 650 W for 5, 10, and 15-min. Green and roasted beans were ground in a professional mill, and stored at room temperature until analysis.

Morphological attributes

Bean physical characteristics were measured by applying the method developed by Tran et al. (2017) [11] with modification. For the weight, 20 coffee beans were freely chosen in each sample (green and roasted), then, it was determined using a precision balance reading to ± 0.0001 g. Bean length, width and thickness were also measured for twenty replicates using a caliper with an accuracy of 0.02 mm.

Oil yield

Twenty to twenty-five grams of grounded coffee beans were placed into a Soxhlet extractor connected to a flask containing 250 mL of n-hexane. The extraction was conducted at the boiling temperature of n-hexane for 6 h [12].

To determine the extract weight, the solvent (n-hexane) was evaporated using a rotary evaporator at its boiling point until a complete removal. Then, the oil yield was determined:

$$\text{Yield} = \frac{weight\ of\ extracted\ oil}{weight\ of\ total\ sample} \times 100$$

Fatty Acids determination

The fatty acids profiles were determined by the method described by Harhar et al. (2019) [13]. Indeed, fatty acids were esterified by boiling under reflux for 10 minutes a mixture of 1 g of oil, 10 ml of methanol and 0.4 ml of 2 N potassium hydroxide prepared in methanol. After cooling at room temperature, 2 ml of hexane were added to the mixture and washed with distillate water. The hexane layer containing fatty acid methyl esters (FAMEs) was collected and analyzed. The fatty acid composition was determined as their corresponding methyl esters by gas chromatography (Agilent-6890) coupled with a flame ionization detector (GC-FID). The capillary column CP-Wax 52CB (30 m x 250 µm i.d., 0.25 µm film thickness) was used. The carrier gas was helium, and the total gas flow rate

was 1 mL/min. The initial oven temperature was 170 °C, the final temperature 230 °C, and the temperature gradient was 4 °C/min. Injector and detector temperature were set at 220 °C. The injection volume of the samples was 2µL in a split mode (split ratio 1:50). The results were expressed as the relative percentage of the area of each individual fatty acid peaks.

Antioxidant activities

Samples extracts were prepared according to the method cited by Bobková et al. (2020) [14], The amount of 3.5 g was weighed directly into glass beakers, 60mL of distilled water heated at 95 °C was poured into the sample and the content was stirred properly. The samples, which were extracted for 5 min, were filtered and used directly for analysis (TPC, TFC, TAC).

The determination of total polyphenolic content (TPC) was realized by the modified Folin-Ciocalteu spectrophotometric assay according to Nounah et al. (2017) [15]. 1 ml of diluted extract (df=100) was introduced into a glass tube (10 ml), then 2.5mL of Folin-Ciocalteu reagent diluted with distilled water (1/10) and 4 mL of Na_2CO_3 (7.5 % water solution) were added. The tubes were left for 30 min at 45°C to develop the blue complex. All samples were prepared in duplicates for each analysis. The measurements were provided by a UV/VIS spectrophotometer at a wave length of 765 nm. The gallic acid was used as a standard for preparation of the calibration curve within the range of 5-160 mg/L. Blank contained Folin-Ciocalteu, reagent, Na_2CO_3, and distilled water without the standard or extract. The calibration curve had a correlation coefficient R^2=0.9997. The quantity of polyphenols was expressed in mg GAE/g of coffee.

The total flavonoids (TFC) in the extracts were determined using a colorimetric method reported by Huang et al. (2011) [16]with some changes. First, about 1 mL of diluted sample (df=100) was placed in a 10 mL volumetric flask. Subsequently, 0.3 mL of (5%) sodium nitrite ($NaNO_2$) was added. About 0.3 mL of (10%) aluminium chloride ($AlCl_3$) was added after 5 min and the mixture was allowed to stand for another 6 min, afterward about 2 mL of 1 M sodium hydroxide (NaOH) was added and the total volume was increased to 10 mL with distilled water. Then, the solution was mixed well and allowed to stand for 30 min. Finally; the absorbance was recorded against a blank at 415 nm. The flavonoid content was determined as the quercetin equivalent from the calibration curve of quercetin standard solutions and expressed as quercetin equivalent (mg QE/g).

The free radical scavenging activities of coffee extracts were determined using DPPH (2,2-diphenyl-1-picrylhydrazyl) by the method described by Ismaili et al. (2016) [17]. In brief, 0.5 ml of DPPH solution (0.2 mM) in ethanol was mixed with 2.5 ml of the diluted extract (df=1000) in absolute ethanol, and allowed to stand in the dark for 30 min at room temperature. Thereafter, absorbance was measured at 517 nm against blank samples using a spectrophotometer. All the experiments were performed in duplicate and the mean ± sd values were reported. The scavenging capacity was calculated using the following equation and expressed as the inhibition of DPPH:

$$\%\ inhibition\ of\ \text{DPPH} = (A_C - A_S)/A_C \times 100$$

Where A_C is the absorbance value of the 0.2 mM DPPH ethanol solution, and A_S is the absorbance value of the test solution. In addition, freshly prepared ascorbic acid standards of serial concentrations (0.2 – 6 ug/ml) were treated in the same way as the samples to trace the calibration curve.

Mineral elements composition

Mineral content was determined by incinerating the sample's powder in the muffle furnace at 525°C for 4 Hours. the obtained ash was treated using 4ml of 65% NHO3 and 10ml of HCl and the analysis was carried out using a Perkin Elmer Model Optima 8000 DV spectrometer.

Results and discussion

Morphological attributes

For bean physical quality, the weight, length, width and thickness of 20 beans are used to provide a good measure of bean size. Coffee with a larger bean size usually gets a good price and rating than coffee with a smaller bean size, even though the larger bean size does not guarantee a more satisfactory roast or liquor [11].

Table 1 represents the variation of physical characteristics (weight, length, width and thichness) of green and roasted coffee beans at different microwave powers and times. Our results show that the morphological characteristics of coffee beans are highly affected by the roasting process. Thus, the longer the roasting time and the microwave power, the greater was the loss in weight of the beans (Table 1). However, the opposite was observed in the other characteristics. The losses in the weight of coffee beans roasted at 350, 500 and 650 w for 5, 10, and 15 min range from 3.9 to 53.4%. the highest amount was recorded at 650 w/15 min (53.15%), 650 w/10 min (45.8%) and 500 w/15 min (44.8%). Nevertheless, coffee beans length, width and thickness increased during microwave roasting from 11.673±1.069 to 14.070±1.072 mm, from 8.824±0,536 to 10.839±0.996 mm and, 5.455±0.796 to 6.766±0.780 mm respectively (Table 1). Indeed, our findings are in a good agreement with Bustos-Vanegas et al. (2018) [18], who reported that high temperature roasted coffee beans had an important volumetric expansion, and an increased mass loss. This weight reduction may be a reflection total volatile substances [19].

Oil yield

Coffee oil is considered an especially important aroma and flavor enhancer of roasted coffee containing a significant amount of the coffee's aroma flavor compounds [2]. The oil yield of unroasted and roasted coffee beans extracted by solvent method is shown in Table 2. The oil yield of green coffee beans was 6.75±0.16 g/100g. Our result is in accordance with that of Alkaltham et al. (2020) [20]. At 350 and 500 w, the oil yield increases according to the duration as well as the power of roasting (from 6.75±0.16 g/100g to 8.22±2.45 g/100g and from 6.75±0.16 to 14.94±1.07 g/100g respectively) (Table 2). Our results are in agreement with the study of Budryn et al. (2012) [21] who reported an increase in the oil content from 10.22% to 11.31%. Also, Alkaltham et al. 2020) [20] has found an augmentation of oil yield from 5.75% for green to 9.10% for microwave-roasted coffee beans. A relative increase of oil content during roasting was also observed in research by Oliveira et al. (2006) [22]. However, at 650 w a decline of the oil content is observed during the maximal time of roasting (15 min). In fact, exaggerated microwave power and time can lead to degradation of bioactive components, thereby inducing a reduction in extraction yield [23]. In most cases, increasing microwave power and temperature induced an improvement in yields. However, this only happened at a certain microwave power value for a few plant materials, which did not experience degradation of bioactive components, or an insignificant increase or reduction in yields [24].

Fatty acids

To evaluate the added value of the composition of spent coffee grounds oil, we were focused on analyzing the fatty acid profile. This family of compounds is an essential indicator of its nutritional and cosmetic values, mostly the unsaturated fatty acids [25-26]. Additionally, the fatty acids are important components of the saponifiable fraction in vegetable oils. Table 4 shows fatty acids profile of microwave roasted coffee beans oils at different powers and times. Our results reveal that the fatty acids profiles are not affected by the microwave roasting process. Indeed, roasted and green coffee oils contain two types of fatty acids. In fact, the unsaturated fatty acids (USFA) were the most abundant accounting for 57.59±0.12 g/100g followed by saturated fatty acids (SFA) (42.22±0.19 g/100g) (Table 4). The main monounsaturated fatty acids of coffee oil in ascending order are palmitic acid (32.17±0.20 g/100g), stearic acid (7.04±0.06 g/100) and arachidic acid (2.92±0.08 g/100g) (Table 4). Some therapeutic and pharmacological properties of vegetable oils are likely to result from its high unsaturated fatty acid content [27-29]. In fact, the coffee oil also contains appreciably larger amounts of unsaturated fatty acids, especially linoleic acid with 44.82±0.12 g/100g, oleic acid with 12.05±0.09 g/100g (Table 4). Linolenic acid is only a minor component with a concentration of less than 1 %. The small content of this fatty acid can be used to detect the adulteration of coffee beans oil with other oils rich in linolenic acids such as rapeseed oil and soybean oil [12]. Our findings regarding fatty acids were consistent with previously published literature [20-21].

Antioxidant activity

Flavonoids and phenolic acids, which are very common in tea and coffee, are important contributors to their flavor and health benefits. In another way, polyphenols are abundant in plants, and their role in preventing oxidation has been well researched for many different plant foods [30] In the present study, polyphenols, flavonoids and antioxidant compounds were extracted using hot water to identify their levels in coffee beverages as prepared by consumers. The antioxidant potential of coffee is related to the presence of phenolic components, and it is well acknowledged that the roasting process influences the antioxidant characteristics of coffee [20]. The bioactive compounds quality in coffee beans influenced by several parameters including drying and roasting [5]. The influence of microwave roasting parameters on the total polyphenols and flavonoids contents of coffee beans has been investigated by several authors [14,20,31-33].

The obtained results showed that green coffee beans contain a high level of TPC, TFC, they represent 41.45±2.73 mg GAE/g and 52.16±4.99 mg QE/g respectively (Table 3). The finding results are similar to those reported by Ramalakshmi et al. (2007) [34] for TPC, but for TFC the finding result is lower than that reported by Haile et al. (2020) [35], this difference could be explained by coffee beans quality. An optimum level of TPC and TFC was observed after roasting at 350 w for 5 min. Comparing with the unroasted beans, values of TPC and TFC in studied samples increased from 41.45±2.73 to 48.33±0.17 mg GAE/g, and from 52.16±4.99 to 56.19±7.85 mg QE/g respectively (Table 3). On the other hand, in other stages of roasting (500 and 650 w at 5, 10 and 15 min) the decrease in TPC and TFC levels was observed. Górecki & Hallmann (2020) [36] reported in their study that TFC achieved its maximum level in light roasted beans, but after a significant decline was found in medium and dark roast.

The antioxidative activity of coffee and its free radical scavenging capability have also attracted the attention of researchers [32]. Our results show that unroasted coffee beans have a significant level of TAC (78.48±0.55 %) (Table 3), our result is in good agreement with [20]. However, TAC of coffee decreases as the roasting power and time increase. Thus, an important decline of TAC was observed after the microwave roasting (from 78.48±0.55 % to 10.17±0.44%) (Table 3), Earlier studies also reported similar findings [20,35,37].

Mineral elements composition

Nine major and microelements were investigated in SCG during this study, namely: potassium, calcium, magnesium, manganese, iron, copper, boron, sodium and phosphorus. The obtained results are summarized in Table 5. Our findings demonstrate that the mineral elements composition is not impacted by the microwave roasting treatment. Among the studied elements, potassium was the most abundant element in all samples, the same observation was raised by other authors [38-39] with 6179,01±24.17 mg/Kg. The second element was calcium (2554.09±45.36 mg/Kg), followed by magnesium with 1651.31±23.20 mg/Kg. In fourth place there was phosphorus. For microelements (Table 5) coffee beans contain a high content of iron, followed by manganese, copper and zinc which represent 193.35±4.17, 25.27±0.76, 24.89±0.22 and 11.97±0.10 mg/Kg respectively. Among the analyzed elements, phosphorus, magnesium and sodium were very high than those cited by Ballesteros et al. (2014) [40], microelements content was similar to those reported by the same author for spent coffee grounds.

Conclusion

This study was concentrated on determining the chemical properties, morphological characteristics and antioxidant activity of green and roasted coffee beans. Roasting process had a significant impact on oil yield, morphological characteristics (weight, length, width and thickness), total polyphenols content, total flavonoids content and total antioxidant capacity. Thus, oil yield increased after roasting process, but at the maximum power and time (650 w/15min) a decline of oil content was observed. However, length, width and thickness of coffee beans increased with power and time of roasting, but also a significant reduction of weight was found. Antioxidant activity analyses reveal that the optimum level of TPC and TFC was obtained at 350 w during 5min. Then after, a notable decrease was observed. However, total antioxidant capacity

of roasted coffee beans, and compared to unroasted ones, was also reduced with power and time of microwave roasting. Nevertheless, treated beans analysis revealed no changes in in fatty acids and mineral elements profiles. From a nutritious point of view, the most desirable coffees are those roasted at 350 w during 5min, which provides the best content of total polyphenols and flavonoids properties.

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Table 1. Morphological characteristics of microwave roasted and unroasted coffee beans.

URC*		Weight (g)	Length (mm)	Width (mm)	Thickness (mm)
		0.306±0.051	11.673±1.069	8.824±0.536	5.455±0.796
350 w	5 min	0.293±0.037	11.986±0.682	9.137±0.569	5.534±0.616
	10 min	0.252±0.045	12.289±1.160	9.797±0.742	6.167±0.729
	15 min	0.237±0.035	12.856±0.778	9.803±0.487	6.219±0.532
500 w	5 min	0.277±0.027	12.842±1.129	10.152±0.781	6.620±1.042
	10 min	0.201±0.051	13.432±0.933	10.479±0.812	6.649±0.939
	15 min	0.149±0.037	13.822±0.820	10.874±0.933	6.738±0.516
650 w	5 min	0.244±0.045	13.080±1.166	10.208±0.438	6.379±0.648
	10 min	0.177±0.047	13.900±0.944	10.981±1.054	6.847±0.956
	15 min	0.146±0.025	14.070±1.072	10.839±0.996	6.766±0.780

URC*: Unroasted coffee.

Table 2. Oil yields of microwave roasted and unroasted coffee beans (g/100g).

	5 min	10 min	15 min
URC*	6.75±0.16		
350 w	7.54±0.43	7.86±0.56	8.22±2.45
500 w	8.89±0.18	11.68±1.33	14.94±1.07
650 w	7.78±0.59	13.38±0.06	12.84±0.11

URC*: Unroasted coffee.

Table 3. TPC, TFC and TAC of microwave roasted and unroasted coffee beans.

		TPC (mg GAE/g)	TFC (mg QE/g)	TAC (%)
URC*		41.45±2.73	52.16±4.99	78.48±0.55
350 w	5 min	48.33±0.17	56.19±7.85	77.58±1.46
	10 min	24.54±1.02	55.69±4.28	76.42±0.55
	15 min	22.61±1.02	43.58±2.85	44.72±0.55
500 w	5 min	18.74±4.78	55.18±0.71	77.58±0.73
	10 min	10.41±0.85	14.33±2.85	29.25±0.55
	15 min	7.75±0.51	4.24±0.71	14.82±0.55
650 w	5 min	14.52±3.93	52.16±0.71	72.55±0.55
	10 min	9.93±4.95	4.24±1.43	20.62±0.00
	15 min	2.20±0.17	0±0.01	11.08±0.36

URC*: Unroasted coffee.

Table 4. Fatty acids profiles of microwave roasted and unroasted coffee beans (g/100g).

URC*		350			500			650			Averages
		5 min	10 min	15 min	5 min	10 min	15 min	5 min	10 min	15 min	
C14: 0	0.09±0.01	0.07±0.01	0.09±0.02	0.08±0.01	0.09±0.02	0.07±0.01	0.09±0.01	0.09±0.02	0.08±0.01	0.07±0.01	0.08±0.01
C16: 0	32.27±0.09	32.35±0.05	31.89±0.03	32.02±0.04	32.52±0.08	32.15±0.04	32.35±0.07	31.98±0.10	32.2±0.04	31.98±0.07	32.17±0.20
C18: 0	7.03±0.01	7.05±0.08	6.97±0.03	6.96±0.06	7.08±0.05	7.12±0.08	6.99±0.04	7.03±0.03	7.1±0.02	7.08±0.01	7.04±0.06
C18: 1	12.1±0.08	12.05±0.07	12.13±0.07	11.98±0.05	11.98±0.02	12.06±0.06	12.15±0.10	11.88±0.01	12.14±0.05	12.12±0.09	12.05±0.09
C18: 2	44.84±0.09	44.9±0.08	44.7±0.09	44.95±0.07	44.65±0.05	44.86±0.04	44.74±0.08	45.01±0.01	44.87±0.04	44.7±0.02	44.82±0.12
C18: 3	0.7±0.01	0.69±0.02	0.71±0.03	0.72±0.06	0.68±0.01	0.72±0.02	0.76±0.05	0.67±0.02	0.73±0.03	0.75±0.04	0.71±0.03
C20: 0	2.97±0.01	2.84±0.03	2.9±0.02	2.95±0.01	2.81±0.01	3.01±0.02	2.88±0.02	3.03±0.03	2.83±0.02	3.02±0.01	2.92±0.08
SFA**	42.36±0.01	42.31±0.02	41.85±0.03	42.01±0.04	42.5±0.04	42.35±0.07	42.31±0.08	42.13±0.06	42.21±0.1	42.15±0.05	42.22±0.11
USFA***	57.64±0.09	57.64±0.08	57.54±0.02	57.65±0.07	57.31±0.01	57.64±0.03	57.65±0.09	57.56±0.07	57.74±0.06	57.57±0.05	57.59±0.12

URC* Unroasted coffee, SFA** Saturated fatty acids, USFA*** Unsaturated fatty acids

Table 5. Mineral elements profiles of microwave roasted and unroasted coffee beans (mg/Kg).

	NTC	350			500			650			Average
		5 min	10 min	15 min	5 min	10 min	15 min	5 min	10 min	15 min	
Zn	11.95±1.12	12.05±1.13	11.82±1.20	12.03±1.44	12.09±1.69	11.87±1.20	11.97±1.10	12.01±1.36	11.84±1.11	12.05±1.96	11.97±0.10
Fe	194.58±9.02	192.74±10.02	196.12±12.33	187.25±8.95	196.36±11.22	193.24±11.36	197.15±10.52	184.69±12.96	196.15±12.66	195.24±9.32	193.35±4.17
B	4.98±0.01	5.02±0.12	4.85±0.55	5.02±0.24	5.05±0.69	4.95±0.58	4.99±0.36	5.03±0.14	4.96±0.58	5.06±0.66	4.99±0.06
Mn	25.14±3.12	25.15±2.14	25.78±2.25	24.15±2.36	24.48±2.47	25.89±2.25	25.81±3.01	26.25±2.99	25.86±2.56	24.22±1.85	25.27±0.76
Mg	1645.79±50.23	1651.56±55.12	1625.65±56.12	1625.26±68.25	1687.17±70.25	1679.84±62.36	1635.89±52.36	1677.26±49.85	1629.99±56.32	1654.69±65.36	1651.31±23.20
Ca	2456.44±10.25	2450.15±102.32	2584.26±98.36	2596.12±112.36	2531.15±117.25	2488.99±103.25	2578.56±96.32	2444.15±100.25	2511.68±101.36	2563.36±96.32	2554.09±45.36
Cu	24.75±4.32	24.85±4.25	25.02±3.21	24.36±2.25	24.88±2.14	25.08±3.25	25.12±3.69	24.96±3.87	24.89±3.65	25.01±3.22	24.89±0.22
Na	747.44±33.23	749.58±36.23	736.65±40.11	745.55±45.23	756.15±25.36	741.55±42.52	758.36±25.36	740.1±21.38	760.12±35.48	739.52±38.15	747.50±8.83
K	6170.69±305.23	6201.33±358.6	6385.22±354.12	6329.57±358.25	6236.85±299.14	6196.12±352.10	6355.33±289.36	6191.36±358.12	6210.65±344.25	6342.96±388.12	6179.01±24.17
P	1382.04±57.63	1369.25±57.25	1416.33±62.36	1354.25±54.12	1369.85±70.12	1347.71±73.69	1425.21±56.21	1395.99±62.66	1394.56±65.12	1425.84±77.25	1370.10±23.18