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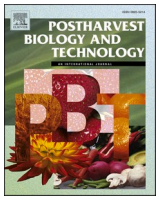

Impact of post-harvest treatments on physicochemical and sensory characteristics of coffee beans in Huila, Colombia

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ABSTRACT

Post-harvest methods applied on coffee cherries processing impact the resulting physicochemical characteristics, and the roasting modulates the beans composition, influencing their sensory analysis. Thus, this work studied the influence of different post-harvest methods (dry, wet and semi-dry) and roasting intensity on the physicochemical characteristics, antioxidant capacity (DPPH, ABTS) and bioactive compounds content of coffee (*C. arabica* L. var. Colombia) samples. Additionally, sensory attributes were determined. The results showed that the degree of roasting modified strongly all the investigated parameters, mainly colour, antioxidant capacity and 5-hydroxymethylfurfural content. Furthermore, the Principal Components Analysis (PCA) explained 87.4 % of the variability of the samples and the results of the antioxidants and hydroxymethylfurfural are related to the roasting degree, while the pH and the chlorogenic acids content allow discriminating the green samples. Finally, the post-harvest methods determined the sensory profile, resulting in coffees with highly differentiated characteristics and classified as "Specialty coffee".

1. Introduction

Coffee is one of the most commonly consumed beverages in the World and one of the most outstanding commodities in international transactions and domestic supply in terms of quantity and value (Vegro and de Almeida, 2019). Coffee production is geographically located in the "coffee bean belt", bounded by the tropics of Capricorn and Cancer, being the three largest producing countries Brazil, Vietnam and Colombia. Furthermore, coffee cultivation provides income and employment for millions of households; however, over 90 % of coffee is exported as green bean form, and value addition in the coffee industry is concentrated in the importing countries (International Coffee Association., 2019).

Quality improvement is a complementary strategy to productivity enhancements that can place a coffee producer on the pathway into high-value market segments, where coffee growers can ensure good practices on-farm and in primary processing, especially in post-harvest treatments. In Colombia, Huila is the region with the largest coffee cultivated area (Agronet, 2021), offering high coffee quantity (979, 327.73 t/year) in addition to outstanding sensory quality. This excellence has allowed Huila to receive Denomination of Origin status in 2013. This recognition leads to the production of specialty coffees as alternative sales channels to producers and associative groups for receiving significantly higher incomes (Gutiérrez-Guzmán et al., 2018). Therefore, understanding the chemical characteristics of post-harvest methods is essential to maintain high-quality production and increase coffee revenues reflected in better livelihoods.

Coffee processing starts immediately after cherry beans harvesting to avoid undesirable fermentations and fruit spoilage (Barrios-Rodríguez et al., 2021). Different post-harvest methods can be applied to obtain "parchment coffee", which is the product that is commercialised on the international market to be transformed into coffee drink. Post-harvest treatments aim to remove the pulp of the cherries, being a key-step to obtain high-quality in the final product. In Huila, the most common post-harvest method is wet processing; however, dry and semi-dry processing are also performed but at a lower rate. In the wet method,

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the cherries are first depulped, followed by a natural fermentation, which duration depends on: i) ripeness of the fruit, ii) coffee variety, iii) amount of beans, and iv) environmental conditions where the fermentation takes. After this step, beans are washed and finally sun-dried. Wet method requires the use of specific equipment to remove the pulp and substantial quantities of water. Moreover, beans treated by this process present better quality and higher cost when compared to beans processed by the dry method. In the dry or "natural" method, the selected cherries (without any previous transformation) are dried until they reach a moisture content of 10–12 %, followed by a mechanical removal of the dry cherry (pod). Finally, the semi-dry method combines both dry and wet procedures, where the cherries are de-pulped and then sun-dried without using the fermentation step for mucilage removal (Duarte et al., 2010). It is well known that post-harvest processing has pronounced effects on the chemical composition of coffee beans, being wet coffee considered to present superior quality because of its higher acidity and outstanding aroma than those obtained by the other methods (Duarte et al., 2010). However, wet processing exhibits some drawbacks, such as it is time-demanding, requires a high water usage generating excessive contents of organic pollutants and uncontrolled fermentation on farms resulting in the lack of coffee quality predictability. Once parchment coffee is obtained, a hulling step is carried out to remove the parchment from the bean. Then, beans are processed for roasting, which is a crucial step where physico-chemical changes occur, and a wide range of volatile compounds are developed, determining the typical flavour and aroma characteristics of the roasted coffee. The roasting step also leads to changes in the functional properties of coffee, causing the loss of phenolic compounds, naturally present in the green beans, and forming other antioxidant compounds because of the Maillard reaction products (Wang et al., 2011). One of these products is 5-hydroxymethylfurfural (5-HMF), which appears as a consequence of sugar degradation and to which specific toxicological effects have been attributed (Alsubot and Aldiab, 2019).

Usually, expert roasters control coffee roasting who determine temperature and time conditions according to their expertise. In general, the endpoint of the roasting is established by a visual criterion depending on the bean colour, and the coffee will be classified according to its roasting degree as medium and dark roasted. As mentioned before, the post-harvest method and roasting strongly influence the coffee beans' chemical, functional, and sensory characteristics, determining their quality. Local producers need to know how these traditional treatments affect the quality of the final product, being these data critical to improve their on-farm treatments and allow them to achieve product differentiation about other commercially available coffee worldwide. Therefore, this work aimed to determine the effect of three post-harvest treatments (dry, wet and semi-dry) and roasting degree on the chemical composition, antioxidant capacity and sensory characteristics of coffee (*Coffea arabica* L. var. Colombia) processed in the Huila region. This gained information can help understand the complexity (acidity and fruit notes) of a coffee resulting from genetic variability and climatic diversity.

2. Materials and methods

2.1. Experimental design

Arabica coffee cherries (*Coffea arabica* L.) var. Colombia was used for the experiments. Three post-harvest treatments (dry, semi-dry and wet) were applied to the cherry beans. A sensory analysis was performed in coffee samples resulting from the three post-harvest methods. Analytical determinations were conducted on ground coffee at different roasting degrees: no roasting (green beans), medium roast coffee and dark roast coffee of the three different post-harvest treatments; at least six independent samples of each *post-harvest treatment x roasting degree* combination were used to perform the analytical determination.

2.2. Chemicals

The reagents employed in the antioxidant activity determinations were: 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), 2,2′-azinobis-(3-ethylbenzthiazoline-6-sulfonate) (ABTS), 2,2-diphenyl-1-picrylhydrazyl (DPPH)) and the standards of the caffeine, 5-(Hydroxymethyl) furfural (5-HMF), and chlorogenic acid (CGAs) were purchased from Sigma-Aldrich (Madrid, Spain). Folin-Ciocalteu phenol reagent and gallic acid employed for measuring the total phenolics content (TPC), Carrez reagents, and methanol and acetonitrile employed for HPLC analyses were purchased from Scharlab S. L. (Barcelona, Spain). Ultrapure water was used throughout the experiments (Mili Q-System, Millipore, Milford, USA).

2.3. Sampling, post-harvest processing and roasting

Coffee cherries were manually collected at the optimum mature stage in a farm at 1700−1800 m above sea level in the Huila region. The coffee beans were selected to eliminate defective beans and impurities and then divided into three batches. Each batch was processed following a different post-harvest treatment: dry (D), wet (W) and semi-dry (SD) (Barrios-Rodríguez et al., 2021). The schedule of the procedure employed for each treatment is shown in Fig. 1. Depending on the post-harvest treatment, the initial moisture content was different, thus the drying time for each treatment was also different. Sun-dried conditions were ambient temperature 28.6 ± 2 °C, RH 62.4 ± 9 % and average wind speed 0.24 m s^{-1}. The moisture content achieved after sun-drying was between 10 and 12 % for all the treatments. After post-harvest, each batch was divided into three new groups, two of these groups underwent the roasting step while a third group was kept as green beans (GB), later used for analytical determinations and sensory analysis. The roasting step was carried out on green beans (150 g per roasting type) processed in a TC 150R rotary drum roaster (Quantik SAS, Bogotá, Colombia) at 190 ± 2.5 °C for a variable time according to the roasting degree. The roasting intensity was visually established by an expert coffee roaster as medium roasted (MR) 8.24 ± 0.17 min and dark roasted (DR) 9.12 ± 0.11 min. Roasting for the sensory analysis was performed following the Specialty Coffee Association (SCA) protocol (Specialty Coffee Association., 2018). The whole procedure was repeated with the three different post-harvest methods.

2.3.1. Physico-chemical characterisation

Coffee samples from the different treatments were analysed to determine moisture content, water activity (a_w), pH, colour parameters and browning index (BI).

The moisture content of the samples was determined using an Agricultural Grain and Seed Moisture Meter PM-450 (KETT, Villa Park, USA) based on the measurement of the capacitance of the sample by comparing the dielectric constant of the solid sample with water. Water activity (a_w) was determined using a water activity meter (Decagon Devices, Pullman, WA, USA). The pH was determined following a method described by Mazzafera (1999), where 2.25 *g* of ground coffee was mixed with 50 mL of deionised water at 80 °C, cooled to room temperature, and the pH value was measured using a Starter 5000 pH meter (Ohaus Instruments, Shanghai, China). Non-enzymatic BI of the samples was determined according to Contreras-Calderón et al. (2016). For this purpose, 7 mL of deionised water was added to 1.5 *g* of ground coffee, mixed, and then centrifuged (2604 *g* / 10 min / room temperature). The supernatant was collected and the process was repeated. The supernatants were mixed and subjected to a clarification process with 1.5 mL of Carrez I (15 % potassium ferrocyanide (w/v)) and 1.5 mL of Carrez II (30 % zinc acetate (w/v)) and deionised water was added until a final volume of 25 mL. Finally, the extract was filtered and, then the absorption at 420 nm was measured with a Thermo Scientific Helios Zeta UV–vis spectrophotometer (Thermo Fisher Scientific Inc., Waltham, USA). Colour was determined on ground coffee (8 *g*), placed

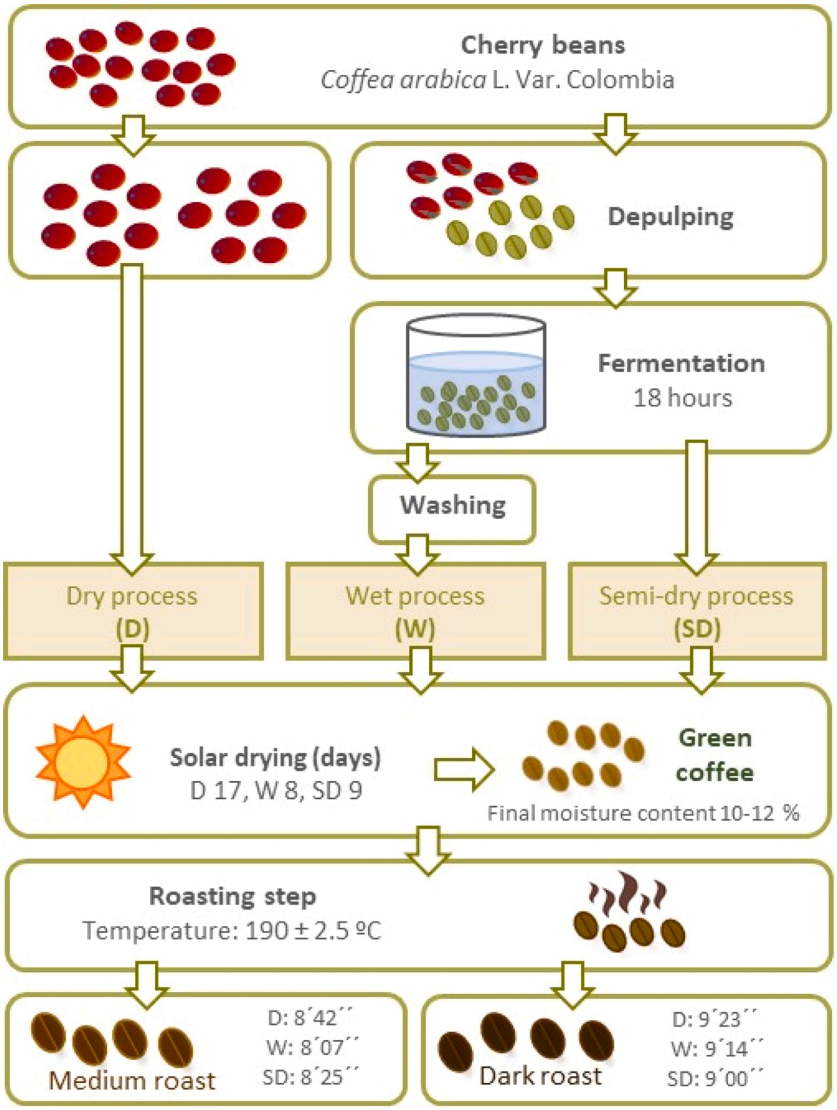

Fig. 1. Scheme of the post-harvest conditions of the cherry beans produced in the region of Huila, Colombia.

homogeneously in a layer at the bottom of a Petri dish (75 mm diameter), covered with a low reflectance optical glass CR-A5/1829−752 M. Measurements were taken at ten different sample points using a CM-700d colorimeter (Konica Minolta, Ramsey, USA). The colour was recorded using the CIEL*a*b* system (D65 illuminant and 10° of visual angle).

2.4. Antioxidant capacity and total phenolics content

2.4.1. Extract preparation

Antioxidant capacity, TPC and CGAs content were determined from the hydroalcoholic extract prepared according to DIN 10,767 procedure (DIN, 2005). For the sample extraction, 1 g of ground coffee was weighed, placed in an Erlenmeyer flask and 50 mL of methanol-water (50/50, v/v) was added. The mixture was kept under constant stirring for 40 min and finally filtered using Whatman No. 1 filter paper (Sigma-Aldrich, Spain). The preparation of the extracts was performed in triplicate.

2.4.2. Antioxidant capacity

The antioxidant capacity was determined using the ABTS and the DPPH assays, following the methodology described by Jeszka-Skowron et al. (2016) with some modifications.

For the ABTS assay, 20 µL of the extract diluted in methanol (1:4 v/v) was mixed with 2 mL of the ABTS solution (7 mM of ABTS and 2.45 mM of potassium persulfate) and the mixture was left in darkness at room temperature for 6 min. The absorbance of the sample was measured at 734 nm. Trolox was used for calibration and the results were expressed as the Trolox Equivalent Antioxidant Capacity (TEAC) in micromoles of Trolox Equivalents (TE) per kg on a dry weight basis (mmol kg^{-1}).

For the determination of antioxidant activity by the DPPH method, 300 µL of the extract diluted in methanol (1:50, v/v) was taken and 2.7 mL of the DPPH solution was added. The mixture was left in the dark at room temperature for 60 min. The absorbance of the samples against a reagent blank was measured at 517 nm. The results were expressed as the TEAC (mmol kg^{-1}).

2.4.3. Determination of total phenolics content

The TPC was determined using the Follin-Ciocalteu assay. For this, 300 µL of the diluted extract (1:50 v/v) with the methanol-water mixture (50/50 v/v), 5 mL of Folin-Ciocalteu reagent and 4 mL of sodium carbonate (10 % w/v) were spun in a vortex mixer and incubated in the dark at room temperature for 60 min. The absorbance of the sample was measured at 765 nm. For the preparation of the calibration curve, solutions of gallic acid (between 50–750 mg L^{-1}) were used. The results were expressed as g of gallic acid equivalent per kg of sample on a

dry weight basis (g kg^{-1}).

2.4.4. Determination of chlorogenic acids

The CGAs analysis was performed using the same HPLC equipment and C_{18} analytical column (Scharlab, Barcelona, Spain). The elution was set at a constant flow of 1 mL $min^{-1,}$ and a mixture of methanol and water (0.2 % acetic acid (v/v)) (80:20) was used as starting eluent conditions. The initial elution condition was maintained for 5 min, then the proportion of methanol increased to 44 % in 15 min, and finally reduced to 20 % until 24 min was completed. The signal monitoring was performed at 324 nm.

2.5. Determination of caffeine

Caffeine determination was carried out following the procedure described by Naegele (2016). 0.5 g of ground coffee and 0.5 g of MgO were combined in 25 mL of double distilled water and stirred for 20 min at 90 °C in a water bath. Then, the solution was cooled down to room temperature and filtered. The coffee extract was diluted with ultrapure water (2:10 v/v) and filtered through a 0.45 μm syringe filter of regenerated cellulose. The samples were analysed with a Hitachi LaChrom Elite HPLC system (Hitachi Ltd., Tokyo, Japan) equipped with an autosampler (model L-2200) and UV detector (model L-2400). The separation was carried out with a C_{18} analytical column (Scharlab, Barcelona, Spain), using isocratic elution mode methanol:water (25:75 v/v) for 10 min at a constant flow rate of 1 mL min^{-1}. The signal monitoring was performed at 280 nm.

2.6. Determination of 5-hydroxymethylfurfural

For the determination of 5-HMF, 0.5 *g* of ground sample was weighed in a 10 mL centrifuge tube and 5 mL of 0.1 % formic acid solution (v/v) was added. The tube was stirred for 1 min and centrifuged (4500 *g* / 4 °C / 10 min) and, then the supernatant was collected. Two additional extractions were performed using 2 mL of the formic solution (0.1 % v/v). All the supernatants were pooled and dissolved in 10 mL of double-distilled water. Then, 5 mL of this extract was clarified with 250 μL of Carrez I and 250 μL of Carrez II. The resulting mixture was centrifuged again, and the supernatant obtained was filtered with a 0.45 μm nylon syringe filter. The 5-HMF determination was conducted with the HPLC equipment and C_{18} column described above, using as mobile phase a mixture of acetonitrile and 0.1 % formic acid (5:95 v/v) in isocratic mode at a constant flow of 1 mL min^{-1}, and monitoring the signal at 280 nm.

2.7. Sensory evaluation

Sensory analysis was performed on 160 g of roasted coffee from each of the evaluated post-harvest methods. A four-judge trained panel evaluated the samples following the cupping protocol of the Specialty Coffee Association (Specialty Coffee Association., 2018), using the parameters established by Colombian technical standards NTC 2758 and NTC 3566, adapting recommendations proposed by other authors (Ladino-Garzón et al., 2016). The evaluation considered 10 attributes rated on a 16-point scale, representing the levels of quality in quarter-point increments between numeric values from 6 to 9. The overall score was calculated as the sum of all the attributes subtracting the defects. A final score greater than 80 indicates a speciality coffee. This procedure was carried out at Centro Surcolombiano de Investigación en Café (CESURCAFÉ, Neiva, Colombia),

2.8. Statistical analysis

The multifactorial ANOVA analysis, followed by the Tukey multiple comparison test, was used for the statistical comparisons. Differences were considered significant at $p < 0.05$. Principal Component Analysis (PCA) was employed to quantitatively investigate relationships among the coffee treatments concerning the parameters evaluated (except for colour parameters, the browning index and sensory evaluation). Statistical data processing was performed with Statgraphics Centurión XVII (Manugistics Inc., Rockville, USA).

3. Results and discussion

3.1. Physicochemical characterisation

The results obtained after physico-chemical characterisation of green and roasted coffee samples processed by different post-harvest methods are summarised in Table 1.

The moisture content of green beans and roasted coffee samples were in the range reported in other studies, such as 8–12 % for green beans and below 5 % for roasted coffee (Tripetch and Borompichaichartkul, 2019). The roasting step led to a moisture reduction compared with the moisture content of green beans, being this reduction higher as roasting increased. Moisture reduction varied within 73–78 % for medium roast samples and within 85–87 % for dark roast samples, independently of the post-harvest method employed. Furthermore, the differences in the moisture content found between post-harvest methods for the same roasting degree could be attributed to the adjustment of the traditional roasting, which occurs visually according to the criterion of the expert roaster. In this sense, the roasting time required to achieve the target of perceived colour was different depending on the post-harvest method employed, leading to the moisture differences mentioned above.

The pH values of samples submitted to the dry treatment were similar to those reported by other authors (Ferreria et al., 2013), being values lower than those obtained for the wet and semi-dry samples at a given roasting degree. The dry post-harvest method leads to a low pH, probably to the absorption of acids produced during beans fermentation throughout the drying periods (Ferreira et al., 2013). The microbial activity contributes to the acidification during fermentation, leading to the observed pH decrease. In the case of wet processing, the pH reduction could be related to the accumulation of lactic acid and acetic acid generated by lactic acid bacteria; however, in the case of the dry processing, the incidence of yeast, mainly *Saccharomyces*, *Candida* or *Pichia* species, lead to the production of acetic, propionic, malic and citric acids as metabolites, with a subsequent potential metabolites migration into the seed contributing the acidity of the final product (De Bruyn et al., 2017). The pH of the coffee samples was also affected by the roasting process. Previous studies have reported higher pH values of green beans compared to those observed in roasted coffee (Lee et al., 2017). Rao et al. (2020) also found that the pH of the brew increased with the roasting degree.

Colour differences were observed among the post-harvest methods and roasting degree (Table 1). Colour of coffee provides essential information on roasting degree and sensory properties, as well as on functional properties, chemical characteristics, and acceptance of green and roasted beans. Colour of green beans is highly influenced by variety and origin, and it has been reported that Arabica green beans are greener than Robusta, which presents a yellower colour. Arabica variety shows lower a* values while Robusta variety is characterised by higher b* values (Santos et al., 2016). The results obtained in the present study were similar to those reported in other works (Santos et al., 2016; Sacchetti et al., 2009) for Arabica variety. Tripetch and Borompichaichartkul (2019) evidenced that colour differences, which can appear in green coffee during storage or post-harvest treatments, are a strong indication of oxidative processes and natural enzymatic biochemical transformations. In our study, the green coffee samples processed by the different treatments showed very similar colour values, indicating that the post-harvest method did not affect enzymatic transformator oxidation processes. Roasted coffee samples showed lower luminosity values, b* (yellow), chroma and tone than green coffee samples for all post-harvest treatments and roasting degrees. One of the

Table 1
Moisture content, water activity (a_w), pH, colour parameters and browning index (BI) obtained by dry (D), wet (W), and semi-dry (SD) for green beans (GB), medium roast coffee (MR), and dark roast coffee (DR). Results are expressed as mean ± standard deviations (n = 6).

Post-harvest	Roasting degree	Moisture (g kg^{-1})	a_w	pH	L*	a*	b*	BI
	GB	100 ± 1aA	0.618 ± 0.096aA	5.79 ± 0.04aA	54 ± 4aA	4 ± 2aA	21 ± 2aA	0.053 ± 0.001aA
D	MR	27 ± 1bA	0.479 ± 0.053bA	4.82 ± 0.05bA	32 ± 1bA	10 ± 1bA	17 ± 1aA	1.233 ± 0.003bA
	DR	15 ± 2cA	0.402 ± 0.021cA	5.07 ± 0.02cA	25 ± 1cA	9 ± 1cA	8 ± 1aA	1.260 ± 0.002cA
	GB	113 ± 5aB	0.632 ± 0.002aA	5.97 ± 0.05aB	53 ± 2aB	2 ± 1aB	18 ± 2bB	0.052 ± 0.002aB
W	MR	24 ± 1bB	0.449 ± 0.028bA	4.86 ± 0.02bB	29 ± 1bB	12 ± 1bB	19 ± 1bB	1.287 ± 0.002bB
	DR	15 ± 1cB	0.436 ± 0.047cA	5.09 ± 0.02cB	24 ± 1cB	10 ± 1cB	9 ± 2bB	1.322 ± 0.003cB
	GB	108 ± 1aB	0.609 ± 0.003aA	5.83 ± 0.05aB	55 ± 1aA	3 ± 1aA	19 ± 2cC	0.040 ± 0.005aB
SD	MR	25 ± 2bB	0.515 ± 0.013bA	4.90 ± 0.03bB	30 ± 1bA	12 ± 1bA	13 ± 1cC	1.254 ± 0.001bB
	DR	16 ± 1cB	0.452 ± 0.043cA	5.10 ± 0.03cB	25 ± 2cA	10 ± 1cA	10 ± 1cC	1.380 ± 0.010cB
F-ratio$^{p-value}$								
Post-harvest (P)		13.60***	1.63ns	15.51***	5.51***	7.71***	38.74***	159.68***
Roasting degree (R)		10,862***	89.67***	3818.72***	3291.53***	1386.83***	447.13***	151748.13***
P x R		27.84***	2.08ns	16.85***	1.74ns	26.65***	70.28***	131.39***

Different lowercase letters indicate significant differences for the roasting degree. Different capital letters indicate significant differences for the post-harvest method. p-value: ns: no significant; *** $p < 0.001$.

most important modifications occurring during coffee roasting is colour change due to non-enzymatic browning reactions such as Maillard and caramelisation. The roasting degrees, medium and dark, were established on an empirical basis by experienced personnel. As the same roasting conditions can lead to different sensory properties depending on the multiple factors, the expert coffee roasters can determine the end of the roasting process by visual inspection of the external colour of the beans. In our study, coffee samples roasted at the same level showed similar values of the colour parameters, independently of the post-harvest method employed, which explains that the endpoint of the roasting is established by visual evaluation. Santos et al. (2016) reported that coffee beans become darker as the roasting process proceeds and the yellowish character decreases markedly (b* decreases). The colour variation from the initial green-yellow to red-orange and, finally, to brown is typical in matrices where Maillard's and caramelisation reactions take place. This fact explains the reduction in luminosity (L*), a* and b* values, as roasting degree increased. The values of the CIEL*a*b* parameters of roasted coffee samples agreed with the values given by other authors for coffee samples subjected to different roasting degree (Contreras-Calderón et al., 2016; Ludwig et al., 2013; Schots et al., 2020). In general, the same effect of roasting on coffee colour has been observed in other studies carried out on coffees of Arabica, Robusta species and mixtures of these (Contreras-Calderón et al., 2016; Ludwig et al., 2013). Particularly, Sacchetti et al. (2009) employed L* values to classified coffee samples according to their roasting degree, using this parameter as a time-temperature indicator of the total thermal effect.

As it was expected, green coffee samples had the lowest BI values, regardless of the type of post-harvest method used, being BI higher in the coffee samples subjected to more intense roasting (Table 1). Contreras-Calderón et al. (2016) observed a wide range of BI on ground commercial coffees (0.33–1.81) based on the roasting degree. Likewise, these authors found differences on the post-harvest methods reporting that roasted coffees processed by wet and semi-dry fermented treatments showed greater browning than the coffees processed by the dry treatment.

3.2. Antioxidant capacity

The antioxidant capacity values from ABTS and DPPH assays for coffee samples are shown in Fig. 2. Values obtained by both analytical methods were higher in roasted coffee samples than in green beans, regardless of the post-harvest method used.

These results agree with those given in other studies, where it has been shown that coffee antioxidant capacity increased after roasting (Daglia et al., 2000; Del Castillo et al., 2002; Sacchetti et al., 2009). The increase in antioxidant activity after roasting has been described either to the release of high-active low molecular weight polyphenols or to the formation of brown compounds that show antioxidant activity. However, there is some controversy about the effect that roasting has on the antioxidant activity of coffee. In contrast to the results found in this

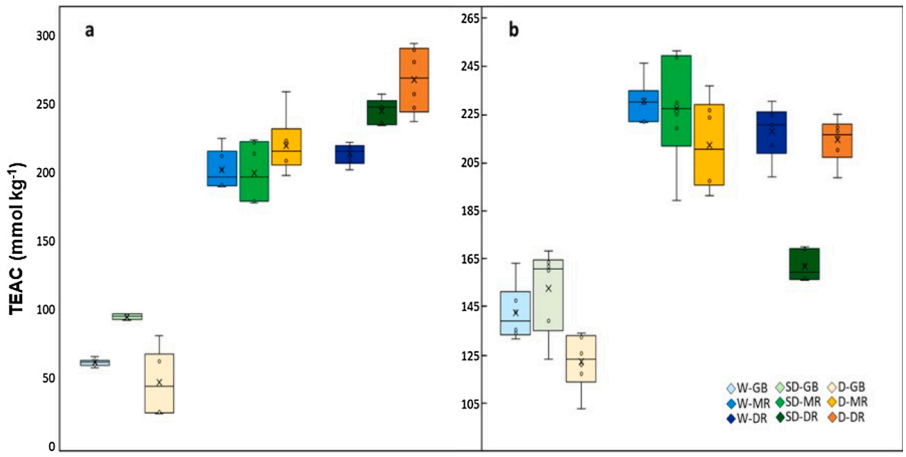

Fig. 2. ABTS (a) and DPPH (b) radical scavenging capacity from coffee extracts: green beans (GB), medium roast (MR) and dark roast (DR) of the post-harvest processing methods: semi-dry (SD), wet (W) and dry (D). Results were calculated on the basis of dry weight. Vertical bars indicate standard deviations (n = 6) (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

work, Nebesny and Budryn (2003) reported that green beans had an antioxidant activity higher than the same coffee subjected to roasting, and they also indicated that the intensity and duration of heat treatment had a negative effect on the antioxidant capacity of coffee. Some authors have found that light roasting improved antioxidant capacity concerning green and dark roasted coffees (Hečimović et al., 2011). These higher values in light and medium roasts could be attributed to their higher content in phenolic compounds (Vignoli et al., 2014). However, other studies have established no correlation between the degree of roasting and the antioxidant activity value of coffee (Bedoya-Ramírez et al., 2017; Contreras-Calderón et al., 2016). Kocadağlı and Gökmen (2016) stated that the variability among different studies could be attributed to differences in the extraction procedures and the analytical methods used. Also, the selected trials could be the main causes of the disparity in the results consulted. In addition, the relationship between the degree of roasting and the antioxidant activity could depend on other factors such as the coffee species (Arabica or Robusta), characteristics of the beans or other roasting parameters (Del Castillo et al., 2002).

3.3. Total phenolics, chlorogenic acids, caffeine and 5-hydroxymethylfurfural

The content of total phenolics, chlorogenic acids, caffeine and 5-HMF in the different coffee samples are shown in Table 2.

Values of TPC in green beans was within the range of values given by other authors (Jeszka-Skowron et al., 2016; Tripetch and Borompichaichartkul, 2019; Hečimović et al., 2011; Pérez-Hernández et al., 2013). There is great variability in the TPC of green coffee beans in the literature since the value of these compounds is affected by multiple factors such as species, variety, origin, and beans characteristics. In the study carried out by Contreras-Calderón et al. (2016), the phenolic content of 41 different roasted coffee samples was analysed, ranging between 125−491 mg kg^{-1} coffee. The TPC obtained in roasted samples was higher than in green beans for all the post-harvest methods evaluated. Hečimovič et al. (2011) found similar results to those obtained in this study by analysing coffees from different varieties, in green beans and at the roasted degree (light, medium and dark), where the highest TPC were observed in light and medium roasted coffees.

The term "chlorogenic acids" (CGAs) refers to a family of polyphenol esters that include hydroxycinnamic acids (caffeic acid, ferulic acid and p-coumaric acid) with quinic acid. CGAs are the compounds that contribute most to the antioxidant properties attributed to green beans (Babova et al., 2016; Duarte et al., 2010). The obtained CGAs content in the green beans coincides with the results given in other studies for this same variety (Macheiner et al., 2019). Thus, the differences can be attributed to the origins for the same coffee variety (Babova et al., 2016). In the study conducted by Babova et al. (2016), where coffee samples from different species and different origin were analysed, the relationship between the antioxidant capacity of green beans and the content of CGAs was demonstrated. This correlation has also been observed in the present study, where the green beans samples processed by the dry treatment had a lower CGAs content and a lower antioxidant activity than those processed by the wet and semi-dry method. CGAs were reduced because of the roasting process, which decreased higher as the roasting degree increased. In this sense, in roasted coffee samples, the antioxidant activity could also be attributed to other compounds, such as Maillard reaction products produced during roasting, mainly melanoidins, and which have demonstrated antioxidant capacity (Nebesny and Budryn, 2003; Pérez-Hernández et al., 2013).

The caffeine content in coffee is one of its main quality parameters. Caffeine content ranged from 10.8–13.4 g kg^{-1}, being these values similar to those found in other studies for coffees from different species (Babova et al., 2016; Duarte et al., 2010; Hečimović et al., 2011; Macheiner et al., 2019; Pérez-Hernández et al., 2013). It is interesting to note that the caffeine content is closely related to the coffee variety, Robusta species can contain twice as much caffeine as the Arabica species (Babova et al., 2016). Moreover, it should be pointed out that the coffee samples processed by the dry post-harvest method presented higher caffeine values. Green samples showed lower caffeine values than roasted coffee for all post-harvest methods, which is in accordance with other studies (Hečimović et al., 2011); however, other authors have established that the change of caffeine content during the roasting process can be neglected (Macheiner et al., 2019). It is necessary to highlight that the roasting process has a huge impact on the coffee beans physical structure, especially the so-called “first crack” that increases the overall volume, inside porosity and pore volume of the coffee bean, enlarging the surface area for the caffeine the extraction. Regarding the effect of roasting degree, the caffeine content in medium roasted coffee samples was higher than in those subjected to more intense roasting for all post-harvest methods. These data agree with the results obtained in other studies (Hečimović et al., 2011). The lower caffeine content in dark roast samples could be attributed to the release of caffeine in the gas phase during roasting processes as a consequence of coffee bean cracking (Novaes et al., 2019).

Values of 5-HMF were in the range of those reported in other works with similar coffee varieties and post-harvest methods (Park et al., 2021). However, the concentration of 5-HMF on coffee is very variable and depends on different factors, such as beans chemical composition, initial humidity of the sample, and roasting procedure that includes time

Table 2
Total Phenolics content (TPC), chlorogenic acids (CGAs), caffeine, and 5-hydroxymethylfurfural (5-HMF) of coffee obtained by dry (D), wet (W), and semi-dry (SD) for green beans (GB), medium roast coffee (MR), and dark roast coffee (DR). Concentrations were calculated on the basis of dry weight. Results are expressed as mean ± standard deviations (n = 6).

Post-harvest method	Roasting degree	TPC (g kg^{-1})	CGAs (g kg^{-1})	Caffeine (g kg^{-1})	5-HMF (mg kg^{-1})
	GB	23 ± 5aA	19.7 ± 0.6aA	11.3 ± 0.9aA	ND
D	MR	49 ± 8bA	17.9 ± 1.4bA	13.4 ± 0.5bA	348 ± 1.5aA
	DR	49 ± 1bA	11.1 ± 0.3cA	12.4 ± 0.5bA	475 ± 6.6bA
	GB	24 ± 6aA	24.2 ± 2.1aB	10.8 ± 0.9aB	ND
W	MR	51 ± 7bA	18.3 ± 0.2bB	12.1 ± 0.3bB	398 ± 3.4aA
	DR	47 ± 5bA	10.0 ± 0.3cB	11.7 ± 0.4bB	455 ± 3.1bA
	GB	26 ± 3aA	25.0 ± 1.70aC	10.3 ± 0.7aB	ND
SD	MR	50 ± 7bA	18.6 ± 0.4bC	12.3 ± 0.3bB	425 ± 15aB
	DR	45 ± 7bA	11.8 ± 0.2cC	12.7 ± 0.6bB	520 ± 55bB
F-ratio$^{p-value}$					
Post-harvest (P)		1.3^{ns}	15.51***	6.43***	6.96*
Roasting degree (R)		135.71***	3818.72***	31.05***	1002.96***
P x R		0.8^{ns}	16.85***	2.21^{ns}	3.4^{ns}

Different lowercase letters indicate significant differences for the roasting degree. Different capital letters indicate significant differences for the post-harvest method. p-value: ns: no significant; *p < 0.05 *** p < 0.001. ND: no detected.

and temperature. The highest content of 5-HMF found in our study was observed in the semi-dry treatment, which could be related to the presence of the mucilage, which was conserved during fermentation and drying steps. This causes an increase in the concentration of sugars in the beans, where sucrose is the main disaccharide in coffee and its principal degradation product is the 5- HMF (Hu et al., 2021). In contrast, the wet and dry processing removed some of these sugars, as the mucilage was washed after fermentation (Hameed et al., 2018) or eliminated by the hulling step after sun-drying. Regarding the degree of roasting, 5-HFM content was higher in samples subjected to dark than medium roasting. In general, it is assumed that 5-HMF is intensively formed during the first stage of the roasting process until a maximum level is reached, as necessary time to achieve this maximum dependent on the roasting temperature (Hamzalıoğlu and Gökmen, 2020). Temperatures and roasting times in our experiment were lower than those used in other works, which did not allow us to reach the same levels of 5-HMF observed in the literature (Bedoya-Ramírez et al., 2017; Contreras-Calderón et al., 2016). Although the amount of 5-HMF decreases with the roasting intensity, beans subjected to light or very intense roasting degree are considered to have the lowest 5-HMF values since this compound degrades at very high processing temperatures (Vignoli et al., 2014).

In an attempt to further discriminate the nine conditions (obtained by the combination of post-harvest-treatment and roasting intensities) based on their composition and physico-chemical properties, all the parameters analysed (except for colour attributes) were studied through Principal Component Analysis (PCA). Colour parameters were not used for PCA since they were only related to the roasting process and post-harvest treatment had no effect. The PCA score plot is shown in Fig. 3 and is based on the first two principal components, PC1 (79.83 %) and PC2 (7.21 %), which were responsible for a cumulative variance of 87.04 %. The first principal component was positively correlated to moisture, a_w, pH, and chlorogenic acids. The PC1 clearly distinguished all green coffee samples from roasted coffee samples, as is expected by the important effect of roasting on moisture and a_w. However, the PC1 also differentiated the samples with higher CGAs content (green beans) from those that exhibited higher antioxidant activity and higher TPC and HMF content (roasted coffee); although, their content in CGAs is lower. This fact would demonstrate that even CGAs are known to have an important antioxidant activity, other compounds formed during roasting play an essential role in the antioxidant capacity of coffee. The second principal component (PC2) promoted a distinction between roasting intensities. Thus, the negative values of PC2 showed grouping in the high degree of roasting, being the samples with the lowest TPC and higher HMF content. According to our results, it was evident that roasting had an important impact on the composition of coffee samples, but post-harvest processing did not affect the analysed parameters.

3.4. Sensory evaluation

The values obtained in the sensory analysis of the coffee beverage obtained by the three post-harvest treatments are shown in Fig. 4.

According to the SCA, all coffee brews showed a positive final evaluation higher than 80 points, which allows classifying these coffees as Specialty coffee (Fig. 4). Furthermore. The final scores given to coffee brews processed by the semi-dry and wet treatments were higher than those obtained by the dry coffee brew. The coffee sensory descriptions showed differences in the sensory profile among treatments. The brewed coffee obtained by the dry method was qualified by the assessors as “medium fruity body” and “fresh medium acidity” and associated with “chocolate” and “caramel” flavours. The wet and semi-dry treatments produced coffee brews associated with “citrus” and “fruity” descriptors. Finally, the brewed coffee produced from the wet method was described as “medium-high body” and “medium-high acidity” compared to dry and semi-dry coffees, which were described as “medium body” and “medium acidity”. These results agree with those reported in other studies that established that coffees processed by the wet method present higher acidity and more aroma than dry-processed coffees (De Melo Pereira et al., 2019). Fermentation during wet and semi-dry processes highly influences the sensory profile of coffee brews since the microbial activity generates a wide range of end-metabolites that strongly affect the chemical composition of processed coffees (Haile and Kang, 2019). The microbial-derived metabolites can diffuse into seeds and remain after the roasting process playing an important role in the quality of the coffee brew. Microorganisms associated with coffee fermentation are different yeast species and lactic acid bacteria, but microbiota present in coffee processing may vary according to several factors, such as regional characteristics, coffee cherry composition and type of fermentation. In this sense, a similar sensory profile detected in wet and semi-dry coffee samples, both submitted to spontaneous fermentation, could be related to the characteristic microbiota present in coffee processing of the Huila region. Despite the differences found among post-harvest methods, the coffee brews evaluated had similar sensory properties. This similarity could be related to the specific and unique combinations of aspects such as geographical area, climatic characteristics, altitude, and practices adopted by each coffee farm.

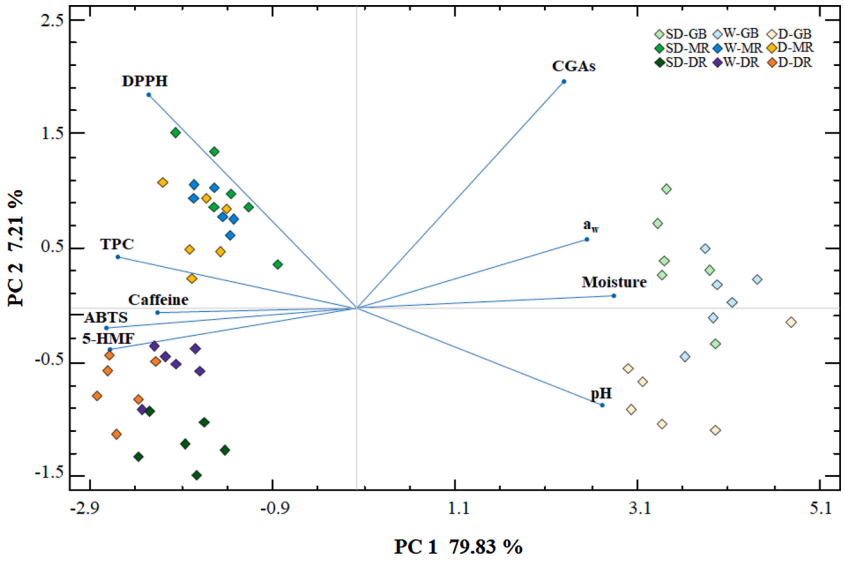

Fig. 3. Principal Component Analysis (Biplot) based on the composition and physico-chemical parameters of coffee samples.

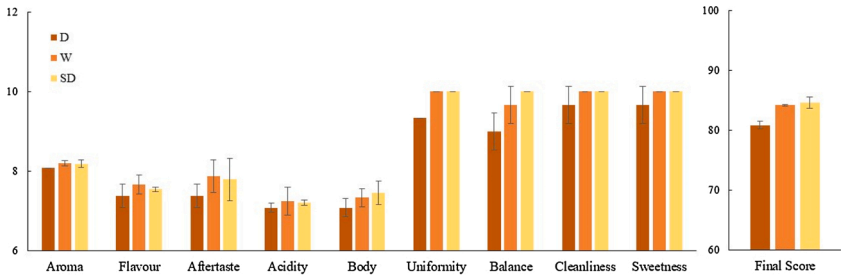

Fig. 4. Evaluations of coffee beverages produced by post-harvest treatments: Dry (D), Wet (W) and semi-dry with fermentation (SD). Vertical bars indicate standard deviations (n = 4).

4. Conclusions

This study provides information that clarifies the effect of post-harvest treatments and roasting degree on the chemical composition, antioxidant capacity and sensory features of coffee processed in the Huila region. The obtained results indicate that the roasting process produces important changes in the physico-chemical characteristics, colour, antioxidant capacity and composition of coffee; however, the traditional post-harvest methods employed at the Huila region, which differ among processing conditions, do not modify main coffee characteristics. The influence of the concentration of chlorogenic acids on the antioxidant activity is maintained in green coffee samples, while in roasted coffee the antioxidant capacity could be mainly correlated to other compounds generated during heat treatment but not affected by post-harvest methods. The evaluated coffee obtained a quality score that allows them to be classified within the range assigned to "specialty coffees". Finally, the post-harvest treatments modify the sensory profile of coffee, resulting in coffees with some sensory differences, but the observed similarities indicate that the unique characteristics of the region of Huila also contribute to define the sensory profile of these coffee brews.

5. Authors statement

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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