

DISCRIMINATION OF COFFEE SPECIES USING KAHWEOL AND CAFESTOL: EFFECTS OF ROASTING AND OF DEFECTS

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ABSTRACT: The two most commercialized coffee species worldwide are: *Coffea arabica* L. (arabica) and *Coffea canephora* Pierre ex A. Froehner (robusta). Since these coffees differ in their commercial value and acceptability, adulteration and mislabeling are major concerns. The diterpenes kahweol and cafestol are considered potential indicators of conilon coffee addition, as they are present in different contents in the species. The degree of roasting and the presence of defective beans may affect the theor of several coffee constituents. The aim of this work was to evaluate the possibility of discriminating the coffee species arabica and robusta through their kahweol and cafestol contents. Samples of arabica, robusta, and of their blends, with different amounts of defects and degrees of roasting (light, medium and dark) were studied. After direct saponification and extraction with *tert*-butyl methyl ether, the samples were analyzed by reverse-phase HPLC with UV detection. The kahweol content varied between 661 and 923 mg/100 g in the arabica coffee, and its presence was not observed in the conilon. Cafestol ranged from 360 to 478 mg in arabica, and from 163 to 275 mg/100 g in conilon coffee. The addition of conilon coffee reduced diterpene contents, but this effect varied according to the amount of defects and roasting degrees. A higher intensity roast did not affect diterpene degradation. No differences in the kahweol and cafestol levels, comparing defective or regular beans, were observed. In the analysis of coffee samples with different degrees of roasting and defects, the parameters kahweol and cafestol showed potential for discriminating between the species.

Key words: *Coffea arabica*, *Coffea canephora*, diterpenes, HPLC.

DISCRIMINAÇÃO DE ESPÉCIES DE CAFÉ POR CAVEOL E CAFESTOL: INFLUÊNCIA DA TORRA E DOS DEFEITOS

RESUMO: As principais espécies comerciais de café no mundo são *Coffea arabica* L. (arábica) e *Coffea canephora* Pierre ex A. Froehner (conilon). Uma vez que esses cafés diferem em valor comercial e aceitabilidade, adulteração e erros na rotulagem são a maior preocupação. Os diterpenos caveol e o cafestol são relatados como potenciais indicadores da adição de café conilon, por estarem presentes em diferentes concentrações nas espécies. O grau de torra e a presença de grãos defeituosos alteram os teores de diversos constituintes do café. Objetivou-se neste trabalho avaliar a possibilidade de discriminar cafés arábica e conilon pelos seus teores de diterpenos. Para alcançar tais objetivos, utilizaram-se amostras de café arábica, conilon e misturas com diferentes graus de torração (clara, média e escura) e proporções de defeitos. Após saponificação direta e extração com *tert*-butil metil éter, as amostras foram analisadas empregando-se CLAE de fase reversa e detecção no UV. O teor de caveol variou de 661 a 923 mg/100 g nos cafés arábica, e não foi observada sua presença nos conilon. O cafestol variou de 360 a 478 mg/100 g no arábica e de 163 a 275 mg/100 g no conilon. A adição de conilon reduziu o teor de diterpenos, mas o efeito foi diferenciado, dependendo do grau de torra e defeitos. O aumento na intensidade de torra não implicou maior degradação dos diterpenos. Verificou-se que não houve diferença nos teores de caveol e cafestol, comparando-se grãos defeituosos ou não. Os parâmetros caveol e cafestol mostraram-se promissores como discriminadores das espécies na avaliação de amostras com diferentes graus de torra e defeitos.

Palavras-chave: *Coffea arabica*, *Coffea canephora*, diterpenos, CLAE.

1 INTRODUCTION

Coffee, recognized by its characteristic flavor and aroma, is one of the most consumed beverages worldwide. Brazil is the crop's main producer and exporter and constitutes its second biggest consumer market (ABIC, 2009). The most cultivated species are *Coffea arabica*

L. and *Coffea canephora* Pierre ex A. Froehner (robusta) (CONAB, 2009). In Brazil, robusta coffee's most common cultivar is conilon (BRAGANÇA et al., 2001). Arabica coffee produces a beverage of higher sensorial quality while conilon, with a lower commercial value, presents a higher content of soluble compounds (MENDES, 1999; SOUZA et al., 2004).

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Coffee's lipid fraction contains, in its composition, diterpenes, kahweol and cafestol, which have well known effects on human health, such as hypercholesterolemic, anti-carcinogenic and antioxidant action (KURZROCK & SPEER, 2001a; LAGO, 2001; SPEER & KÖLLING-SPEER, 2006; URGERT et al., 1995). Arabica coffee has higher diterpene contents than conilon; kahweol appears in higher concentrations in arabica coffee beans, while only traces of it are detected in the conilon. Cafestol is found in both species (KURZROCK & SPEER, 2001b; RUBAYIZA & MEURENS, 2005).

Conilon coffee may be added to arabica in commercial blends for standardizing flavor and decreasing costs (DIAS, 2005; MENDES, 1999; SOUZA et al., 2004). In raw beans, the species can be distinguished visually, as their coloring and shape are different. After roasting and grinding, this distinction is no longer possible and the species must be discriminated in other ways (KEMSLEY et al., 1995; LAGO, 2001).

Brazilian roasted commercial coffees, in terms of quality, flavor, prices, defects and species, are divided into three product categories: Traditional, Superior and *Gourmet*. To categorize these coffees, analysis of the different conilon concentrations in the blends, maximum percentage of defects and minimum score in the sensorial analysis is required (ABIC, 2009).

Because they are present in different concentrations, diterpenes can be used to identify coffee species (FREGA et al., 1994; KURZROCK & SPEER, 2001a,b; LAGO, 2001; RUBAYIZA & MEURENS, 2005; URGERT et al., 1995). The literature also shows that diterpenes have relatively high thermal stability, which favors their use as discriminators between coffees subjected to different degrees of roasting (DIAS, 2005; LERCKER et al., 1996).

There are still few studies on the composition of defective beans, although they represent 20% of the total coffee bean production (OLIVEIRA et al., 2006). Moisture, lipids, protein, caffeine, saccharose and acidity contents have been studied (FRANÇA et al., 2005; MAZZAFERA, 1999; MORAIS et al., 2007; OLIVEIRA et al., 2006), but no data on diterpenes is available.

The objective of this work was to determine the diterpene composition of *C. arabica* and *C.*

canephora coffee roasted in different conditions and with different amounts of defects, to evaluate the potential of the diterpenes to discriminate between species of roasted coffee.

2 MATERIAL AND METHODS

To evaluate the stability of the diterpenes in the roasting process, the species *C. arabica* and *C. canephora* (conilon variety), respectively arabica 1 and 2 and conilon 1 and 2, provided by the Café Iguaçu company (Cornélio Procópio, Paraná state, Brazil), were used. The coffee beans had different quality levels (number of defective beans) and different geographic origins. Arabica coffee 1 (A1) was constituted by beans from Minas Gerais state and had fewer defects, while arabica coffee 2 (A2) came from Paraná state and presented a higher number of defective beans. As to the conilon coffees, respectively from Rondônia and Espírito Santo states, the C2 presented a higher amount of defects. The four samples were subjected to light, medium and dark roasting, resulting in 12 samples which were ground in a 0.84 mm particle size (ABNT 20 sieve). Although the parameters for classifying roasting degree and number of defects are specific to the company, the samples presented defect levels corresponding to beans between type 4 and 8, and the roasting degree was characterized through color measurements. From these 12 pure samples, blends of arabica (A) were made with 20, 30 and 50% of conilon coffee (C) for the different roasts (light, medium and dark) and levels of defects in the beans (1 and 2).

A third batch of arabica coffee, provided by the Instituto Agrônômico do Paraná – IAPAR (Londrina, Paraná state, Brazil), was used to evaluate the influence of defects on diterpene contents. The defective beans were removed from this batch to obtain samples A3 (defective bean-free arabica coffee) and A4 (100% defective beans from the A3 sample). The A4 sample was constituted by 37% of sour beans, 16% black, 15% immature, 22% insect-damaged and 10% broken beans. The samples were subjected to light roasting and ground in the granulometry above, after which ratios of 10, 20 and 30% of defective beans (A4) were added to the A3 sample.

The samples were stored in a cold chamber (0°C) until analysis. Moisture was determined, in

duplicate, using infra-red equipment (OHAUS-MB200, USA) at 105 °C, for 7 minutes (DIAS, 2005). Color was determined, in triplicate, using a Byk Gardner colorimeter (USA) with illuminant D65 and 45/0 geometry, and by evaluating the lightness (L^*) and hue ($H^* = \arctan(b^*/a^*)$) parameters, where b^* is the blue/yellow component and a^* is the red/green one.

The extraction of diterpenes was carried out by direct saponification with potassium hydroxide (KOH, Synth), followed by extraction with methyl tert-butyl methyl ether and clean-up with water, according to the flow chart in Figure 1 (DIAS et al., 2010).

The chromatography analysis was carried out in a Shimadzu liquid chromatograph (Kyoto, Japan), composed of a quaternary system of solvent pumping (model LC10ATvp), DGU-14 Avp online degasser, a Rheodyne injection valve with a 20 μ L fixed loop, a CTO-10 Asvp column oven and a UV/VIS diode array spectrophotometry detector (SPD-M10 Avp). A reverse phase column (Spherisorb ODS1, 250 x 4.6 mm, 5 μ m) and oven temperature of 25 °C, and an acetonitrile/water (55:45) isocratic elution in a 0.9 mL/min flow rate and detection at 230 and 290 nm were also used, for cafestol and kahweol, respectively (DIAS et al., 2010). The analysis was done in duplicate.

The water used to prepare standards and solutions was obtained through Milli-Q (Millipore) purification and filtering. The mobile phases were filtered in a Millipore vacuum filtration system through

a 0.45 μ m membrane (Millipore) and degassed for 2 minutes. HPLC grade acetonitrile (Carlos Erba) was used.

The identification of the compounds was based on retention time comparison and coelution with the authentic standards (kahweol and cafestol, Axxora, San Diego). Quantification was carried out by external standardization, generating calibration curves with concentrations between 50 to 1000 mg/100 g of coffee.

To analyze the effects of roasting degree and species on diterpene contents (samples A1, A2, C1, C2 and blends A1/C1, A2/C2) in a randomized split-plot design, the results were subjected to analysis of variance and Tukey test ($p \leq 0.05$) using the SISVAR. (2009) statistical package. Species represented the main plot and roasting degree the subplot. A significant interaction ($p \leq 0.05$) between species (pure or blends) and roasting degree indicated that the species are affected differently by the roasting process, and this effect was then studied separately in each species. If the main x subplot interaction is not significant, the species present similar behavior regarding the roasting process, and comparisons were made between the global means of each species in each roast and the global roasting means in each species.

The mean test (Tukey, $p \leq 0.05$) (STATSOFT, 2006) and ANOVA were used to compare the diterpene contents in arabica coffee (A3) and in blends with different percentages of defective coffee beans (0, 10, 20, 30 e 100 % of A4).

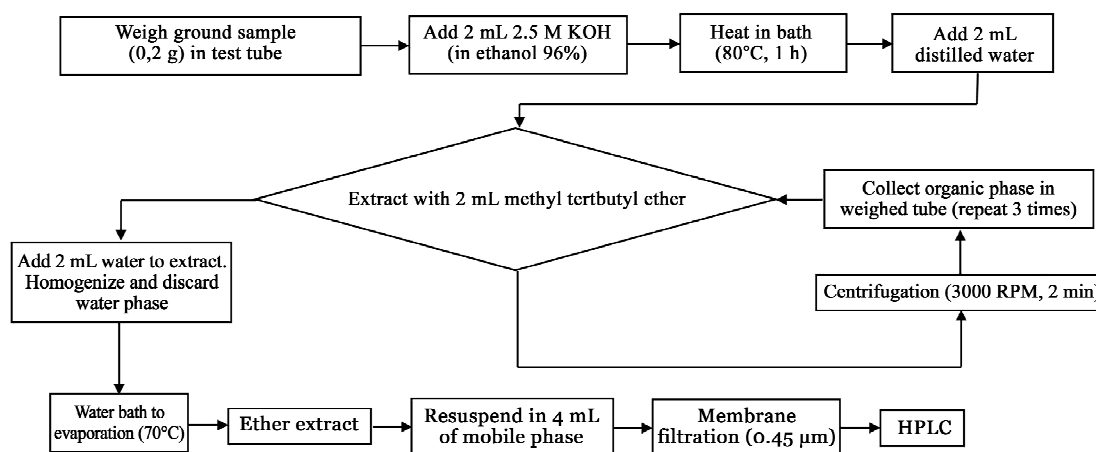


Figure 1 – Flow chart of the extraction of the diterpenes kahweol and cafestol (DIAS et al., 2010).

3 RESULTS AND DISCUSSION

The coffees presented moisture between 1.2 and 4.0 g/100 g of product, considering the different species (arabica and conilon) and degrees of roasting. These values were below the maximum moisture allowed (5.0 %) (BRASIL, 2008), and similar to those described by Nicolau-Souza et al. (2010) for 38 samples of commercial coffees (2.7 to 3.8 g/100 g of product).

In color analysis, it was observed that L^* and H^* decreased with a higher degree of roasting. In the light roast, the L^* values varied between 29 and 40 and the H^* values observed were above 63. In the medium and dark roasts, the lightness values varied from 20 to 29 and from 14 to 20, respectively. In the medium roast, values between 58 and 65 were found for H^* , while in the dark roast they were below 58, reaching even 50. The samples that underwent less intense thermal processes were lighter and more yellow, while the more severe treatments made the samples redder and darker (Table 1).

In general, the L^* and H^* values were similar to those described in the literature. Dias (2005) described L^* ranging between 18 and 38, and H^* between 41 and 63 for arabica and conilon coffees with light, medium and dark roasting. Nicolau-Souza et al. (2010) reported a mean L^* value of 20 and H^* value of 56 for samples of different brands of roasted commercial coffees.

In a typical chromatogram of medium roasted arabica and conilon coffees, the retention times of the diterpenes were close to 16 (kahweol) and 17 minutes (cafestol) (Figure 2). The sum of kahweol and cafestol (mg/100 g of dry base sample), which corresponds almost to the total diterpenes, varied from 1021 to 1344 in the arabica coffee, and from 163 to 275 in the conilon, regarding the different degrees of roasting (light, medium and dark) (Tables 2 and 3).

These results are aligned with the literature, which establishes that the diterpenes would correspond to between 10 and 20% of the lipid fraction in roasted coffees (GROSS et al., 1997; SPEER & KÖLLING-SPEER, 2006). Diterpene contents of 1.3% in arabica coffee and 0.2% in conilon have been reported (LAGO, 2001; URGERT et al., 1995).

The kahweol content varied from 661 to 866 mg/100g in arabica coffee samples subjected to different roasting processes, while in the conilon coffee these compounds were not observed (Table 2). In general, it was observed that adding conilon coffee to the arabica significantly reduced the kahweol contents in the light, medium and dark roasting, but the behavior varied according to the intensity of the roast and the type of coffee sample (1 or 2) used.

Comparing the cafestol contents (Table 3), in the arabica coffee values between 360 and 478 mg/100 g were found, while for the conilon they varied from 163 to 275 mg/100 g in the different roasting degrees. In general, adding conilon to arabica coffee significantly reduced the cafestol concentration.

In the literature, a great variation in the concentration of the compounds can be found, as well as controversies regarding the presence or absence of the compound kahweol in conilon coffee.

In arabica coffee, kahweol contents have been observed in a wide range, from 100 mg to over 736 mg/100g of coffee (FREGA et al., 1994; KURZROCK & SPEER, 2001a; LAGO, 2001; RUBAYIZA & MEURENS, 2005; URGERT et al., 1995). Nicolau-Souza et al. (2010) observed, for commercial coffees, probably mostly arabica and conilon blends, kahweol contents between 100 and 800 mg/100g of coffee. Some authors have highlighted the absence of kahweol in

Table 1 – Lightness (L^*) and hue (H^*) values of the arabica (A1 and A2) and conilon (C1 and C2) coffees subjected to different degrees of roasting.

Species	Lightness			Hue		
	Light	Medium	Dark	Light	Medium	Dark
A1	29.0 ± 0.5	20.1 ± 0.1	14.7 ± 0.5	63.8 ± 0.8	58.7 ± 0.3	50.8 ± 0.4
A2	31.9 ± 0.4	21.1 ± 0.2	16.6 ± 1.0	63.0 ± 0.4	62.7 ± 0.8	50.0 ± 0.8
C1	39.3 ± 1.2	28.9 ± 0.5	19.9 ± 0.1	66.9 ± 0.9	64.4 ± 0.5	57.7 ± 3.1
C2	32.7 ± 0.4	21.7 ± 0.3	13.6 ± 0.7	67.6 ± 0.4	62.1 ± 0.1	55.2 ± 2.0

*Mean ± standard-deviation of the values in triplicate.

conilon coffee (DIAS, 2005; NACKUNSTZ & MAIER, 1987); others have found concentrations of the compound below 13 mg/100 g of coffee (FREGA et al., 1994) and/or only traces of it (KURZROCK & SPEER, 2001a; RUBAYIZA & MEURENS, 2005).

The cafestol results in the literature vary from 76 to 300 mg/100 g of conilon coffee and 100 to 700 mg/100 g of arabica coffee (FREGA et al., 1994; KURZROCK & SPEER, 2001a; RUBAYIZA & MEURENS, 2005). Nicolau-Souza et al. (2010) described, for commercial coffees, values between 250 and 550 mg/100g of coffee.

The diterpene values found in this work were similar to those described in the literature. High kahweol concentrations were observed in arabica coffee, which coincided with the highest results in the literature; however, the cafestol results of this work coincided with the intermediate values described.

It is important to consider that there are many differences between the extraction procedures used in the different methods of diterpene quantification. In many works, preliminary extraction (cold extraction with solvents or Soxhlet) is followed by saponification, leading to a higher possibility of oxidation (RUBAYIZA & MEURENS, 2005; SPEER & KÖLLING-SPEER, 2006). Direct saponification has been described as an efficient alternative to extract other unsaponifiable compounds, such as cholesterol and its oxides, which avoids the formation of oxidation products (SALDANHA et al., 2006). As kahweol presents one double bond more than cafestol in its structure, which makes it more sensitive to oxidation, alterations in the method have a greater effect on kahweol values. Dias et al. (2010) emphasized the need for special care in sample preparation, as a greater oxidation of kahweol was observed in extractions in different coffee matrices.

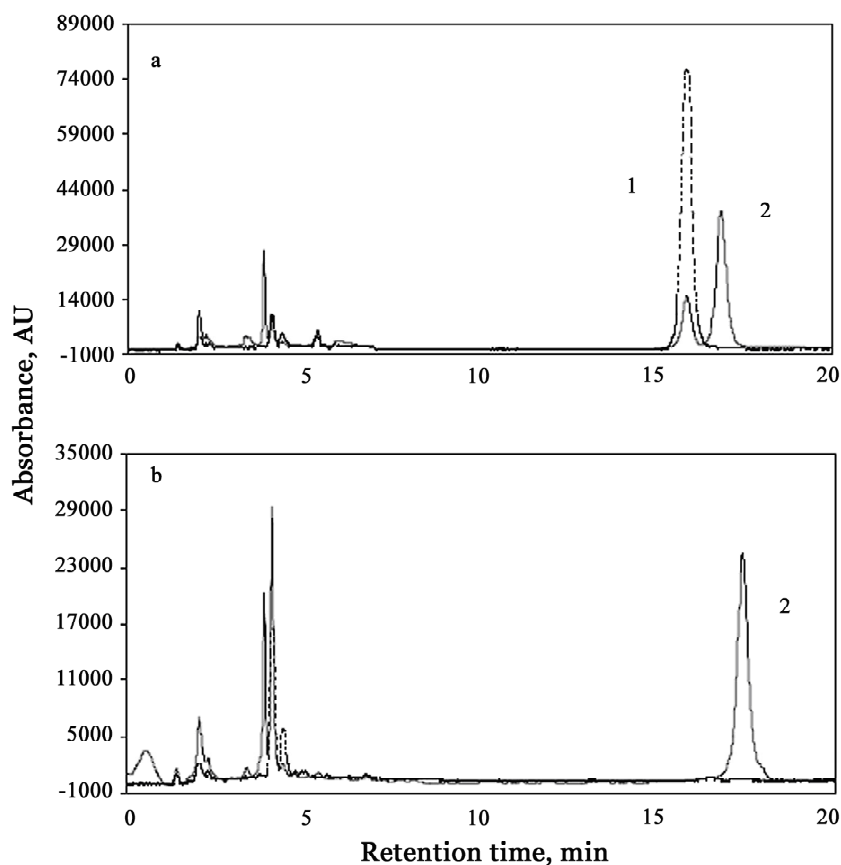


Figure 2 – Typical diterpene chromatograms of medium roasted coffees: arabica (a) and conilon (b). Detection at 230 (—) and 290 nm (---). Kahweol (1) and cafestol (2) peaks.

Even if we do not take into account the possible differences in efficiency of the analytical methodologies, there is still a variation of the raw material used. Initially, the geographic origin and the varieties used could be considered the factors responsible for the greatest variations in diterpene concentrations. Kurzrock & Speer (2001a), in their review, observed significant variations in the kahweol and cafestol esters of green coffee beans of different origins (Asia, Africa and South America).

Frega et al. (1994) assessed the contents of various diterpene alcohols in raw and roasted arabica (12 different origins) and conilon (8 origins) coffee beans, as well as in the blends of the two species. The authors observed kahweol variations between 414.8 and 672.7 mg and cafestol variations between 299.4 and 583.6 mg/100g in different samples of roasted arabica coffee. For the conilon varieties, the kahweol contents remained between 3.6 and 12.5 mg and the cafestol, between 76.4 and 190.1 mg in 100g of coffee.

Another factor that could be considered responsible for these variations is the roasting process, as there is no consensus in the literature on the stability of these compounds. Some authors have observed that diterpenes present good stability in high temperatures, but they can form dehydroderivates (dehydrocafestol and dehydrokahweol) in small amounts when roasting temperatures are raised (LAGO 2001; SPEER & KÖLLING-SPEER, 2006). Speer & Köllig-Speer (2006) also highlighted the presence of other decomposition products, such as kahweal, cafestal, isokahweol and dehydroisokahweol, as well as alterations in the kahweol and cafestol esters, in a more severe roasting process. Urgert et al. (1995) assessed the behavior of kahweol and cafestol in arabica coffee subjected to a high degree of roasting (26.5% of weight loss), and concluded that roasting did not reduce the concentrations of these compounds.

In Tables 2 and 3, a significant interaction ($p \leq 0.05$) was observed between species and roasting in the kahweol concentrations in samples 2 (pure species and blends) and the cafestol concentrations in samples 1 (pure species and blends). However, in samples 1 (A1, C1 and A1/C1 blends) and 2 (A2, C2 and A2/C2 blends), no significant interactions ($p = 0.13$ and $p = 0.12$) in their respective kahweol and cafestol

concentrations were observed, indicating that they depended only on the proportions of the coffee species, that is, they were not affected by different roasting parameters. Therefore, comparisons were made between the overall means of the species in each roasting degree and the overall means of the roasting degree for each species. The dark roast samples presented a lower cafestol content than the light and medium roast ones. This shows that these samples were affected differently by roasting.

Therefore, although in all the cases the addition of conilon coffee reduced diterpene contents, the effect was different according to roasting degree and sample. It was not observed, however, that the roasting process led necessarily to the degradation of the diterpenes; the contents in the medium and dark roasted coffees were not systematically lower than in the light roasted ones and, in some cases, they were even higher.

It is interesting to observe that the total lipid concentration is also influenced by roasting, even if dry base data are used. Lipid concentration can increase proportionally during the process due, mainly, to the destruction of carbohydrates, as roasting breaks down the matrix of the cell wall, leading to the solubilization of polysaccharides and decrease of its molecular weight (LAGO, 2001; OOSTERVELD et al., 2003). REDGWELL et al. (2002) reported that, depending on the type of processing, between 12% and 40% of coffee bean polysaccharides are degraded. In arabica and conilon coffee, an increase in the total lipid concentration, from 11.4% to 15.4% and 6.1% to 9.6%, respectively, (LAGO, 2001) has been observed.

In this work, the samples were obtained by batch processing, in which color was the criteria for determining the degree of roasting. Therefore, small alterations in time and temperature could lead to a difference in carbohydrate degradation, with a consequent variation of the lipid content, which partly justifies the variations of the diterpenes in some of the results in Tables 2 and 3.

Brazilian legislation allows for the presence of defects in coffee; however, it does establish limits to their quantity and type (BRAZIL, 2009). ABIC (2009) recommendations stipulate that coffee be constituted by type 8 beans or above, with a maximum of 20% in weight of defective beans. As the commercial coffees

will present a certain degree of defects, and this percentage can vary, it is important to verify if the presence of defective beans affects diterpene concentrations.

Throughout this work the occurrence of defects has been considered, as the arabica (A1 and A2) and conilon (C1 and C2) coffees also presented, apart from different origins and varieties, differences in defect levels. To assess in more detail their influence, a non-defective arabica sample (A3) and a sample made up exclusively of defective arabica beans (A4) were used. To the A3 sample, different percentages of defective beans (10, 20 and 30%) were added, after which the diterpenes kahweol and cafestol were determined (Table 4).

In these study conditions, the amount of defects did not influence the diterpene concentrations (Table 4), a fact also observed by Moraes et al. (2007) who found similar lipid concentrations in defective and non-defective beans. As coffee can be commercialized from type 2 to type 8, it is important to certify that their kahweol and cafestol concentrations are not heavily influenced by defective beans. It is also important

to highlight that the diterpene concentrations of each specific class of defective bean were not checked. Another, more detailed study, assessing black, immature, sour and other defects would allow for a broader assessment of the issue. However, as samples with different compositions of coffee defects (A1 and A2, C1 and C2) were assessed, and defective and non-defective samples (A3 and A4) were compared, a first important indicator was here obtained that diterpene content is less affected by the amount of defective beans than other compounds previously described as discriminators between the coffee species.

As kahweol and cafestol presented stability in the roasting process, and there were different diterpene concentrations between the species, these compounds are potential discriminators of roasted arabica and conilon coffees.

It is important to continue the study of diterpene composition in roasted coffee. A broad database, including a greater number of brazilian arabica and conilon coffee varieties, could be the basis for establishing criteria and models, based on diterpene contents, to assess conilon concentrations in arabica coffee blends.

Table 2 - Kahweol content (mg/100 g bs.)* in arabica (A1 e A2) and conilon (C1 e C2) coffees and their blends, subjected to different roasting degrees (light, medium, dark).

A1/C1 Proportion (%)	Roasting degree		
	Light	Medium	Dark
100 / 0	829 ^{Aa} ± 16	744 ^{Ba} ± 8	800 ^{Aba} ± 28
80 / 20	530 ^{Ab} ± 2	507 ^{Ab} ± 59	516 ^{Ab} ± 18
70 / 30	410 ^{Ac} ± 12	428 ^{Ab} ± 1	478 ^{Ab} ± 26
50 / 50	327 ^{Ac} ± 33	308 ^{Ac} ± 18	294 ^{Ac} ± 20
0 / 100	0 ^{Ad} ± 0	0 ^{Ad} ± 0	0 ^{Ad} ± 0

A2 / C2 Proportion (%)	Roasting degree		
	Light	Medium	Dark
100 / 0	699 ^{Ba} ± 8	866 ^{Aa} ± 21	661 ^{Ba} ± 16
80 / 20	622 ^{Bab} ± 34	744 ^{Ab} ± 17	544 ^{Cb} ± 45
70 / 30	557 ^{Bb} ± 28	717 ^{Ab} ± 3	391 ^{Cc} ± 29
50 / 50	462 ^{Ac} ± 56	393 ^{Ac} ± 3	267 ^{Bd} ± 34
0 / 100	0 ^{Ad} ± 0	0 ^{Ad} ± 0	0 ^{Ae} ± 0

*Mean standard-deviation of the samples quantified in duplicate. ** Different letters, lower case in the columns and high case in lines, indicate a difference ($p \leq 0,05$).

Table 3 - Cafestol content (mg/100 g bs.)* in arabica (A1 e A2) and conilon (C1 e C2) coffees and their blends, subjected to different roasting degrees (light, medium, dark).

A1/C1 Proportion (%)	Roasting degree		
	Light	Medium	Dark
100 / 0	463 ^{Aa} ± 4	398 ^{Ba} ± 46	420 ^{ABa} ± 4
80 / 20	368 ^{Ab} ± 10	340 ^{Aab} ± 0	314 ^{Abc} ± 8
70 / 30	336 ^{Abc} ± 1	307 ^{Abc} ± 6	315 ^{Ab} ± 16
50 / 50	295 ^{Ac} ± 38	286 ^{Abc} ± 4	281 ^{Abc} ± 30
0 / 100	163 ^{Bd} ± 24	250 ^{Ac} ± 8	242 ^{Ac} ± 6

A2/ C2 Proportion (%)	Roasting degree			
	Light	Medium	Dark	Global Mean
100 / 0	455 ± 1	478 ± 6	360 ± 3	431 ^a ± 63
80 / 20	369 ± 28	346 ± 11	309 ± 32	341 ^b ± 30
70 / 30	351 ± 5	327 ± 17	283 ± 31	320 ^b ± 34
50 / 50	304 ± 32	259 ± 1	247 ± 17	270 ^c ± 30
0 / 100	275 ± 10	235 ± 31	188 ± 6	233 ^d ± 44
Global Mean	351 ^A ± 69	329 ^A ± 95	277 ^B ± 65	

*Mean ± standard-deviation of the samples quantified in duplicate. ** Different letters, lower case in the columns and high case in the lines, indicate a difference ($p \leq 0,05$).

Table 4. Kahweol and cafestol concentrations* (mg/100 g bs) in non-defective (A3) and defective samples (A4) of arabica coffee and in blends of non-defective and defective beans.

A3/A4 Proportion (%)	Kahweol	Cafestol
100 / 0	923 ^a ± 12	459 ^a ± 3
90 / 10	895 ^a ± 3	449 ^a ± 7
80 / 20	913 ^a ± 24	462 ^a ± 10
70 / 30	863 ^a ± 17	430 ^a ± 6
0 / 100	932 ^a ± 55	434 ^a ± 15

*Mean ± standard-deviation of the values analyzed in duplicate. ** Different letters, in the same column, indicate a difference ($p \leq 0,05$).

4 CONCLUSION

The diterpenes kahweol and cafestol are potential discriminators of the species *Coffea arabica* L. and *Coffea canephora* Pierre ex A. Froehner in commercial roasted coffee blends. Adding over 20% of conilon coffee to arabica reduced the kahweol and cafestol contents in coffees subjected to different roastings and amount of defects. As the diterpenes present relative stability in the roasting process and the presence of defective beans did not affect kahweol and

cafestol concentrations, a diterpene-based model is robust enough to be applied to commercial coffees with different degrees of roasting and defects.

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