Simultaneous Determination of Caffeine and Chlorogenic Acid in Vietnamese Coffee Products: First-Order Derivative Spectra and HPLC-DAD as a Comparison

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Due to an overlap in the their absorption spectra, the direct simultaneous analysis of caffeine (CFI) and chlorogenic acid (CGA) by UV-Vis is impossible. This study aims to propose an analytical method that employs the principle of first-order derivative spectra for the simultaneous determination of CFI and CGA in coffee. The extraction was conducted at 90 °C in 50 minutes using deionized water as a "green" extraction solvent. The performance of the proposed method was evaluated and the results were not significantly different to those obtained with the HPLC-DAD reference method, i.e., $t_{experimental} < t_{critical}$ (2.23, P = 95%). The working ranges were from 0.50 to 16.5 mg L^{-1} and 2.00 to 28.5 mg L^{-1} for CFI and CGA, respectively, with favourable repeatability (RSD_r of 1.88% and 0.77%) and reproducibility (RSD_R of 2.34% and 1.43%) values. The proposed method was applied to evaluate CFI and CGA concentrations in several ground coffee products of different varieties and roasting degrees, and was found to have the potential to replace the HPLC-DAD for regulating and controlling CFI and CGA as it is a quick, easy, and cheap method.

Keywords: First-order derivative; caffeine; chlorogenic acid; coffee; roasting degrees

Received: July 2022; Accepted: September 2022

The coffee plant belongs to the order Rubiales, family Rubiaceae, genus Coffea. Coffee is one of the most popular non-alcoholic beverages in the world. Billions of cups have been reported to be consumed per day due to its unique flavour and function of evoking mental alertness. Nearly a thousand volatile compounds have been identified in coffee. Most coffees originate from Africa (Ethiopia), followed by Arabia, Constantinople, and Venice [4]. Arabica (*Coffea arabica*) and Robusta (*Coffea canephora var. robusta*) are two popular coffee varieties worldwide, and in Vietnam specifically. Arabica accounts for approximately 75 %, while Robusta coffee is considered more acidic and contributes to 25 % of worldwide production [5, 6]. Most commercial coffee drinks are made from Arabica and Robusta coffee or a mixture of them at various ratios [7]. Based on the colour variation (mainly observed and regulated by the professional coffee producers) [8] and roasting temperature within 10 - 15 minutes, the coffee products can be divided into many types such as light roast (about 170 °C), medium roast (180 - 190 °C), medium-dark roast (220 - 230 °C), and dark roast (240 - 250 °C) [9]. Besides the changes in flavour due to the differences in coffee varieties and roasting degrees, the variation in antioxidant capacity and chemical composition of coffee products, mainly due to roasting temperature and time, have also gained attention [8, 10-14].

Coffee is a major dietary source of purine alkaloids. Caffeine (CFI), or 1,3,7-trimethylxanthine, is a derivative of the purine base methylxanthine, which predominantly causes the bitter taste and could be employed to determine the quality of coffee products [15]. CFI serves as a stimulator of the central nervous system, and helps coffee drinkers to stay awake, feel less fatigued, and be more energetic [16]. However, in cases of excessive consumption, CFI can cause several unpleasant symptoms and induce potential excitement and anxiety in drinkers. Therefore, the recommended daily intake of coffee might vary according to the health of each individual [13, 17]. Furthermore, CFI content differs depending on the coffee variety, i.e., Robusta coffee contains about twice as much as Arabica [13, 17, 18]. Although CFI is relatively stable in solution, the concentration of CFI in coffee products may change with the roasting degree [17, 19].

Chlorogenic acids (CGAs) are a family of esters formed by phenolic acids (trans-cinnamic acids) and quinic acid. CGAs are responsible for producing the pigment, flavour, and aroma of coffee beans, thus determining the quality and acceptability of the

beverage [20]. CGAs are generally considered secondary plant products that protect against environmental stress. Moreover, CGAs are believed to demonstrate certain antioxidant capacities, playing an important role in protecting food, cells, and organs from oxidation [20, 21]. There has been increasing scientific evidence to indicate that CGAs could have health benefits in humans [22, 23], with positive effects on blood pressure control and glucose regulation [24, 25]. As with CFI, the concentration of CGAs are different among coffee varieties and suffer losses during roasting, but coffee is still rich in CGAs [21]. CFI and CGA are considered important substances for determining the quality of coffee, and the ratio of CFI to CGA has been reported as a potential indicator of roasting degree [13, 19].

Due to the overlap of the molecular absorption spectra of CFI and CGA, direct and simultaneous determination of these two substances in coffee matrices by UV-Vis is impossible. Several attempts have employed preliminary chemical separations before UV-Vis measurement [26-29]. These separation approaches, based on liquid-liquid extraction using organic solvent(s), were effective in solving the spectrophotometric issue but were relatively time-consuming, usually requiring multiple extraction cycles and steps for quantitative analysis and could not be considered "green chemistry". Separation based on liquid chromatography coupled with an ultraviolet (LC-UV) or diode array detector (LC-DAD) [6, 17, 19, 30] and (tandem) mass spectrometer (LC-MS, LC-MS/MS) [13, 31] is another approach for the simultaneous determination of CFI and CGA in coffee samples. Analytical procedures developed on chromatographic principles exhibit good potential to determine the CGA species [11, 21, 30, 32]. However, such analyses require well-trained operators, complicated instruments, and time for the chromatographic separation. The conception of a quick, simple, and inexpensive method to determine both CFI and CGA in coffee products and their related sample matrices has generated great interest, especially among coffee producers, manufacturers and even consumers.

The present study aimed to investigate and evaluate an analytical method based on the first-order derivative spectra to directly and simultaneously determine CFI and CGA in roasted ground coffee products from Vietnam. The proposed method employed deionized water to extract CFI and CGA from the sample matrices, a "green chemistry" approach. Several parameters related to the extraction procedure, including extraction time and temperature, were investigated to obtain optimized operating conditions. Then, the analytical method was applied to several commercial Arabica and Robusta roasted ground coffee products. As scientific data for Vietnamese coffee products are still limited, this study is among the first to determine and evaluate two specific compounds present in these products in terms of variety and roasting degree. Moreover, the proposed method requires no pre-treatment before UV-Vis measurement. It potentially negates the requirement for expensive and complicated equipment in cases where the CFI and CGA values need to be controlled during various phases of production.

EXPERIMENTAL

Chemicals and Materials

Caffeine (CFI, ≥ 99% purity) and chlorogenic acid (CGA, ≥ 95% purity) were purchased from Merck (Germany) and used to prepare the single stock solutions of 1000 mg L^{-1} in methanol (HPLC grade, Merck, Germany). The intermediate standard solutions of 100 mg L^{-1} CFI and CGA were prepared by diluting the stock solutions with deionized water (DIW, Millipore, USA). The working standard solutions for CFI and CGA had concentrations of 0.50 - 16.5 mg L^{-1} and 2.00 - 28.5 mg L^{-1}, respectively. The working standard solutions for the UV-Vis spectrophotometry and in the initial mobile phase of 25% solvent B (methanol) and 75% solvent A (0.1% v/v trifluoroacetic acid in DIW) of the chromatographic analysis reference method were prepared daily by appropriate dilution of the intermediate standard solutions in DIW.

Fifteen ground coffee products (3 Arabica coffee samples with 5% butter addition) of the Arabica (ara) and Robusta (ro) varieties of medium (medi) and dark roast were purchased in the local supermarkets in Ho Chi Minh City, Vietnam. The roasted ground coffee products, which originated from Lam Dong Province, Vietnam, were packed in non-permeable polypropylene/aluminium/polyethylene bags, hermetically sealed under vacuum and stored at a temperature of 25 °C and humidity of 70 % until use.

Extraction of Caffeine and Chlorogenic Acid

The extraction procedure of CFI and CGA from the roasted ground coffee products was conducted based on ISO 20481 (2008) [33] with some modifications. Briefly, 0.2 (±0.001) g of ground coffee was weighed into a glass reaction tube. Then, 10.00 mL of DIW was added to the tube, and the mixture was vortexed for two minutes. After that, the tubes were transferred into a water bath set at 90 °C, and the extraction was performed within 60 minutes. The tubes were centrifuged at 4500 rpm for 3 minutes before collecting the supernatant. The extracts were filtered through a 0.45-µm PTFE membrane prior to further spectrophotometric and chromatographic analyses.

The effects of extraction temperature (50, 60, 70, 80 and 90 °C) and extraction time (10, 20, 30, 40, 50 and 60 minutes) were evaluated using two representative roasted ground coffee samples, namely arabica-dark without butter (ara-dark), and with 5% w/w butter addition (ara-dark-5-butter). Two surveyed samples with and without butter were used to evaluate

the effects of sample matrices containing butter on the CFI and CGA values, in order to broaden the application of the proposed analytical method. The CFI and CGA concentrations for each condition were obtained in order to determine the most favourable extraction procedure.

First-Order Derivative Spectra

The ultraviolet-visible (UV-Vis) absorption spectra of CFI and CGA were measured by a UV-Vis 1800 instrument (Shimadzu, Japan), operated with UVProbe 2.34 software. The UV-Vis operation parameters were: scanning range 200 - 400 nm, intervals of 0.5 nm, intermediate scanning speed, and "zero-crossing" for detecting the quantification wavelengths for both CFI and CGA during the proposed first derivative spectra. A quartz cuvette with a light path of 1 cm was utilized.

Reference Method

High-performance liquid chromatography coupled with a diode array detector (HPLC-DAD) system (Agilent 1200, USA) was used as the reference method to compare with the proposed analytical method employing first-order derivative spectra. The HPLC-DAD used a C18 column (Supelco C18, 5 µm, 250 x 4.6 mm). All chromatographic analyses were conducted using ratios of 0.1% v/v trifluoroacetic acid in DIW (solvent A) and methanol (solvent B). The gradient mode was initially set at 25% solvent B. The eluent was changed to 35% solvent B within 20 minutes, and subsequently to 100% solvent B in the next 5 minutes. It was finally returned to the initial condition of 25% solvent B to prepare for the next run. The injection volume and flow rate were 20 µL and 1.0 mL min^{-1}, respectively. The retention times of CFI and CGA were 9.8 and 15.6 min, respectively. The quantification of CFI and CGA by HPLC-DAD was performed by establishing calibration curves (plotting the analyte concentrations and their corresponding peak areas) for both CFI and CGA, at 272 nm and 326 nm, respectively.

Evaluation of Proposed Analytical Method

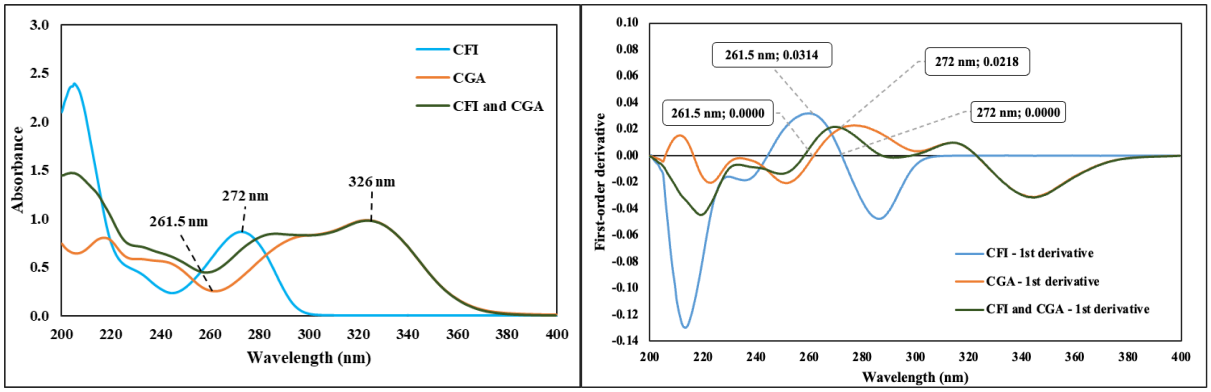
The proposed analytical method for determining CFI and CGA in coffee products was evaluated according to the guidance provided in Appendix F of AOAC (2016) [34]. The analyte quantification was carried out based on calibration curves, which were established based on the linear relationship ($y = ax + b$) between the concentrations of each analyte (mg L^{-1}) and their first-order derivative spectra, recorded at 261.5 nm and 272.0 nm for CFI and CGA, respectively. The limits of detection (LOD) and quantification (LOQ) were calculated by simultaneously analysing 11 blank samples containing only the extraction solvent. The estimated average concentration ($\bar{x}$) and standard deviation (SD) of each analyte were applied in the following relationships: $LOD = \bar{x} + 3SD$ and $LOQ = 3LOD$ [35]. The repeatability (intra-day precision) and reproducibility (inter-day precision) values were assessed through the calculation of the relative standard deviation values for six replicates within one day (RSD_r) and for three separate days (RSD_R), respectively. The accuracy of the proposed analytical method was evaluated by a t-test, using HPLC-DAD as the reference method [35].

The proposed analytical methods were then used to determine CFI and CGA in several Vietnamese roasted ground coffee products (expressed as % w/w dry weight, briefly written as % w/w in this study). All analytical data were analysed with Microsoft Office Excel 2016, and expressed as a mean value ± standard deviation (SD) for each roasted ground coffee sample. The statistical analyses in this study were carried out at a significance level of 0.05.

RESULTS AND DISCUSSION

UV-Vis Absorption and First-Order Derivative Spectra of CFI and CGA

The absorption spectra of CFI and CGA in methanol (UV cutoff of 205 nm) were scanned from 200 - 400 nm at ambient temperatures (Figure 1). The maximum wavelengths of CFI and CGA recorded by the UV-Vis spectrophotometer were 272.0 nm and 326.0 nm, respectively. Figure 1(a) indicated that the spectrum of CFI was characterized by its absorbance in the region of 240 - 300 nm, with a maximum at approximately 272 nm due to the C=O group in the CFI molecule [36]. In comparison, the absorbance spectrum of CGA had two maximum values. The first band was at about 217 nm (with a shoulder at nearly 240 nm), while the second band was localized at approximately 326 nm (with a shoulder at about 298 nm). The maximum absorbance of CGA at 326 nm was due to the occurrence of a HOMO/LUMO transition of π to π^* [37]. Due to the overlap in the CFI and CGA absorption spectra, the direct and simultaneous determination of these two substances by UV-Vis is impossible. Several analytical approaches using UV-Vis have been reported, but these require pre-treatment before spectrophotometric measurement, such as liquid-liquid extraction using ethyl acetate, chloroform, carbon tetrachloride or a mixture of them as the extraction solvent to separate CFI and CGA from the sample matrices [28, 29, 38]. However, such approaches are not considered "green chemistry" due to the utilization of organic solvents. They are also usually time-consuming because of the multiple steps involved in the sample preparation. The proposed analytical method in this study employed the first derivative spectra of CFI and CGA to directly and simultaneously quantify these compounds in roasted ground coffee products. The described method



(a) (b)

Figure 1. (a) The molecular absorption spectra and (b) first-order derivative spectra of CFI, CGA, and a mixture of CFI-CGA

minimizes sample preparation and allows the user to choose a quantification wavelength for each analyte that is not influenced by the signal of the other (Figure 1(b)).

In the first derivative spectra, the background was smoother, which reduces the potential of matrix effects affecting accuracy [39]. In the current study, "zero-crossing" was used to detect the quantification wavelengths for each analyte [40]. At a wavelength of 272.0 nm, the recorded first derivative of CFI was zero, so 272.0 nm (a zero-crossing point of CFI) was employed to determine CGA, while the first derivative of CGA at 261.5 nm was zero; hence 261.5 nm (a zero-crossing point of CGA) was utilized to quantify CFI (Figure 1(b)).

Effect of Extraction Conditions on CFI and CGA Values

Before using the UV-Vis instrument, CFI and CGA must be quantitatively extracted from the coffee matrix. The current study employed DIW as the extraction solvent without adding other organic solvents to ensure a "green chemistry" approach. Single extraction was applied, and the influences of extraction temperature (Figure 2(a)) and extraction time (Figure 2(b)) on the analyte extraction yields were investigated to determine the optimal conditions. The results in Figure 2(a) indicated that the CFI and CGA values obtained increased with extraction temperature from 50 to 90 °C. Typical values obtained with ara-dark were (0.84 ± 0.03) % - (1.02 ± 0.02) % w/w for CFI and (0.45 ± 0.01) % - (0.62 ± 0.01) % w/w for CGA; while values for ara-dark-5-butter were (0.81 ± 0.03) % - (0.97 ± 0.01) % w/w for CFI and (0.39 ± 0.01) % - (0.55 ± 0.01) % w/w for CGA. Hence, it could be observed that higher temperatures increased the extraction efficiency of bioactive compounds [41, 42]. In this study, extraction was not performed at 100 °C due to the boiling point of water, which caused difficulties in pipetting, thus leading to lower repeatability between analyses. From the obtained results, 90 °C was used as the extraction temperature for further investigation. Additionally, the results in Figure 2(b) indicated that a prolonged extraction duration (10 - 60 min) also resulted in higher extraction efficiencies for CFI and CGA. Particularly, the concentrations of CFI and CGA increased between 10 and 50 min, i.e., for ara-dark: (0.93 ± 0.03) % - (1.10 ± 0.01) % w/w for CFI and (0.42 ± 0.01) % - (0.61 ± 0.01) % w/w for CGA; for ara-dark-5-butter: (0.78 ± 0.03) % - (0.99 ± 0.02) % w/w for CFI and (0.43 ± 0.01) % - (0.60 ± 0.01) % for CGA. The highest extraction yields for both CFI and CGA were recorded at 50 min and decreased slightly in the next 10 min (i.e., ara-dark: (1.10 ± 0.01) % *vs.* (1.07 ± 0.02) % w/w for CFI and (0.61 ± 0.01) % w/w *vs.* (0.60 ± 0.01) % w/w for CGA; ara-dark-5-butter: (0.99 ± 0.02) % *vs.* (0.93 ± 0.02) % w/w for CFI and (0.60 ± 0.01) % w/w *vs.* (0.58 ± 0.01) % w/w for CGA). This might be explained by the thermal decomposition of bioactive compounds, particularly CGA in coffee matrices [11, 19, 43]. Therefore, to ensure accurate and quantitative results, 50 min was chosen as the extraction time at 90 °C. As seen in Figure 2, the behaviour of both investigated samples (with and without 5% butter addition) was similar in terms of extraction temperature and time.

Analytical Method Performance for Determining CFI and CGA in Coffee Products

The CFI and CGA calibration curves were established based on the linear relationships between analyte concentrations and their corresponding first derivative spectra at 261.5 nm and 272 nm, respectively. The blank sample used to correct the measured spectra of the analytes was DIW. Figure 3 shows the calculated derivative spectra for different mixed standard concentrations (0.50 - 8.00 mg L^{-1} and 1.50 - 24.0 mg L^{-1} for CFI and CGA, respectively). The established working ranges for CFI and CGA were 0.50 - 16.5 mg L^{-1} and 2.00 - 28.5 mg L^{-1}, respectively, with their corresponding R^2 values of 0.9997 and 0.9992 exhibiting goodness of linearity according to

Appendix F of AOAC (2016) [34]. Compared to HPLC-DAD, the proposed analytical method had similar working ranges for both analytes.

The operating ranges obtained with this method were wide and compatible with various coffee products with the same dilution factors. As seen in Table 1,

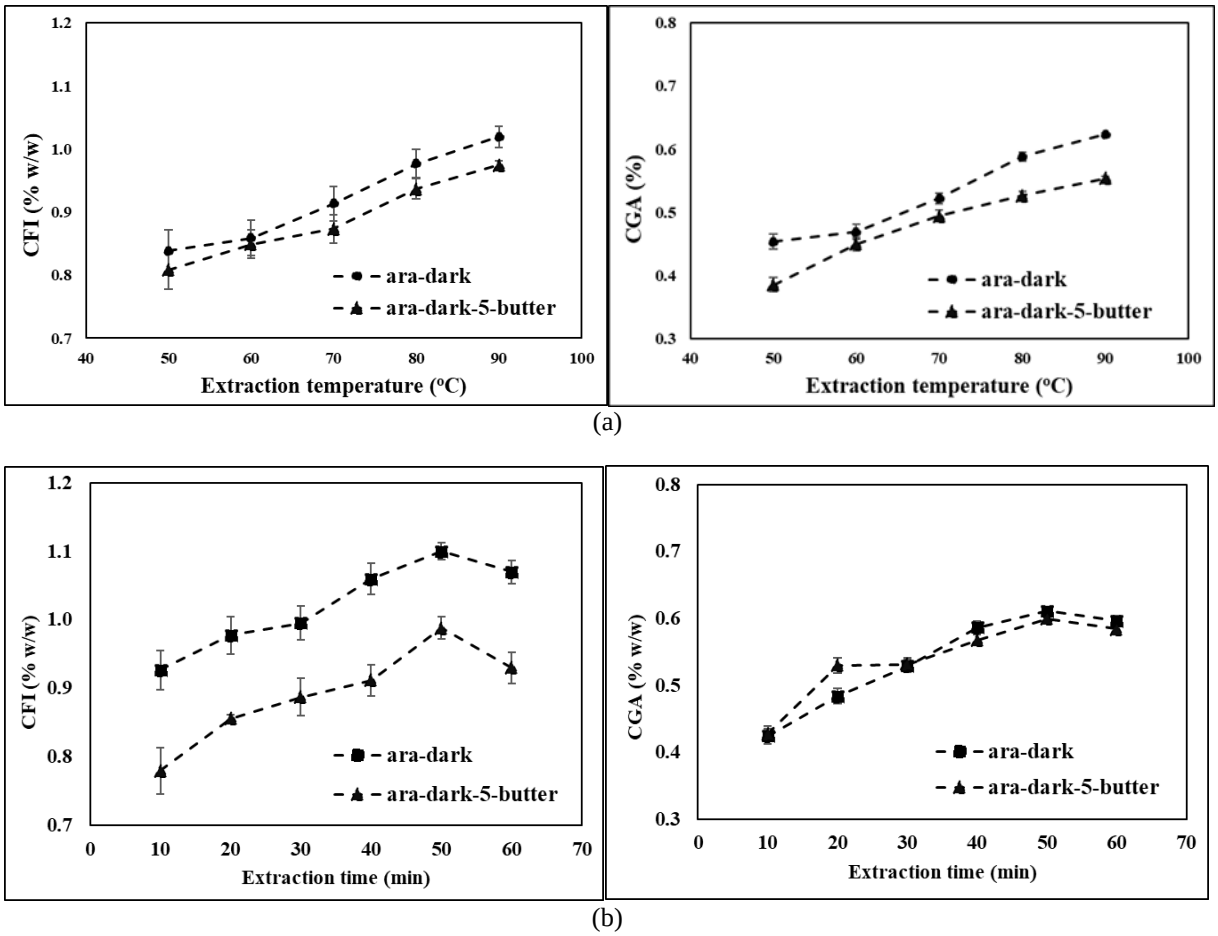


Figure 2. Effects of (a) extraction temperature and (b) extraction duration on CFI and CGA in coffee products

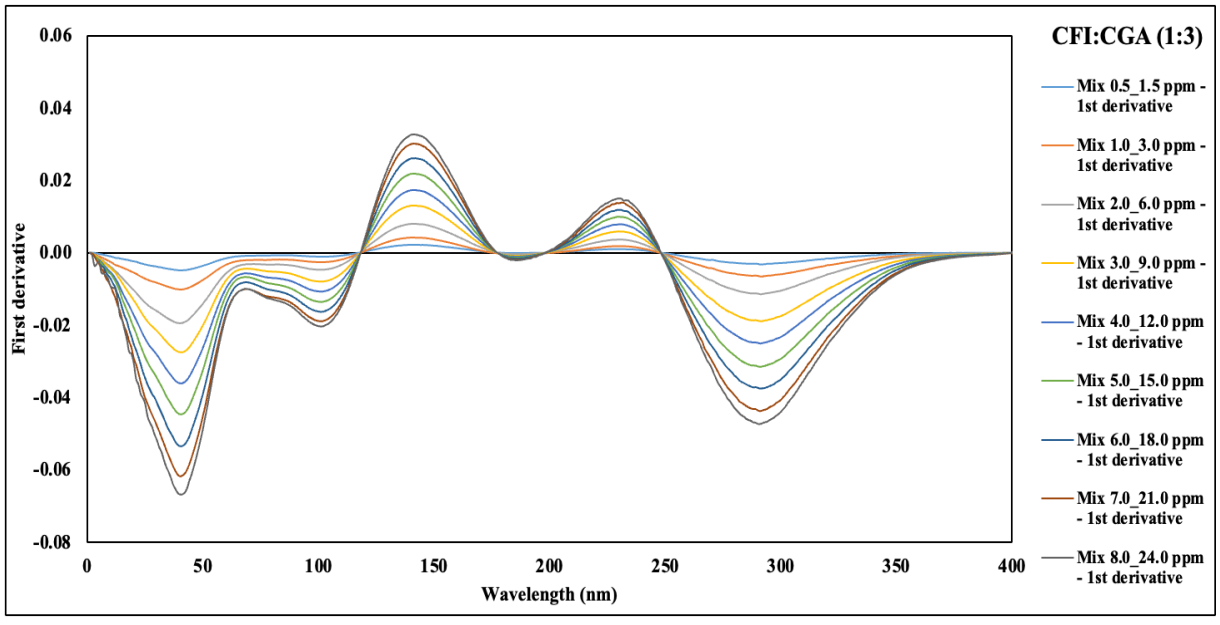


Figure 3. First derivative spectra of CFI and CGA mixtures with different concentrations

Table 1. Comparison of method performance between the proposed method and HPLC-DAD

Criteria	Caffeine		Chlorogenic acid	
	Proposed method	**HPLC-DAD**	**Proposed method**	**HPLC-DAD**
Working range	0.50 - 16.5	0.50 - 16.5	2.00 - 28.5	2.00- 28.5
LOD-LOQ (mg L^{-1})	0.10 - 0.30	0.10 - 0.30	0.42 - 1.25	0.33 - 1.00
Slope (a)	0.00188	69.61	0.00104	51.46
Intercept (b)	–0.00003	–6.23	0.00016	–21.99
R^2	0.9997	0.9997	0.9991	0.9959
Repeatability (%RSD_r)	1.88	0.77	0.77	0.76
Reproducibility (%RSD_R)	2.34	1.34	1.43	1.45
Experimental t-value*	0.693		0.346	
Critical t-value	2.23		2.23	

(*) max value

the estimated LOD and LOQ values were low compared to the actual concentrations of CFI and CGA in the coffee products (in the percentage range). The calculation of %RSD_r, %RSD_R and recovery tests were used for evaluating the repeatability (intra-day precision), reproducibility (inter-day precision), and trueness of the proposed analytical method. The results fulfilled the requirements in Appendix F of AOAC (2016) for analytical method performance evaluation [34]. The CFI and CGA values were also obtained using the reference method. The results obtained using the proposed analytical method did not exhibit a significant difference, according to the t-test at 95% confidence level, where the highest experimental t-values obtained were lower than the critical t-value (2.23), indicating the proposed method provided accurate data with favourable precision (%RSDs calculated from Table 2 were 0.43 % - 3.7 % and 0.43 % - 4.0 % for the proposed method and HPLC-DAD, respectively). However, compared to the HPLC-DAD method, using first-order derivative spectra reduced the analysis duration (i.e., direct measurement and derivative calculation by the available software *vs.* 20 - 30 minutes for chromatographic separation per injection) and used simple instrumentation [19, 43]. Moreover, the proposed method did not require any preliminary chemical separation and only employed DIW as the extraction solvent to quantitatively extract the analytes from the sample matrices, thus an environmentally friendly and "green chemistry" approach. The proposed method also has an important advantage: it was developed based on a UV-Vis spectrophotometer, which is currently available in almost all laboratories and is also a quick, easy, inexpensive, and direct analytical approach for the simultaneous determination of CFI and CGA in roasted ground coffee products and other similar coffee-related matrices.

Application of the Proposed First-Order Derivative Spectra Method for Coffee Products

The evaluated analytical method based on first-derivative spectra was used to determine the concentrations of CFI and CGA in different roasted ground coffee products collected in Vietnam. The present study investigated the effects of coffee varieties and roasting degrees on the concentrations of CFI and CGA in roasted ground coffee products. For other matrices such as milk coffee or food products containing coffee, matrix effects should be investigated first before applying the first-order derivative spectra measurement to ensure reliable and accurate results.

The results in Table 2 showed that the ro-medi-1 and ara-dark-5-butter-1 samples exhibited the highest (2.12 ± 0.04% w/w) and lowest CFI concentrations (0.94 ± 0.02% w/w), respectively. As a general trend, the Robusta coffee products had higher CFI values than Arabica, which has also been previously observed [13, 17, 18]. The variation in CFI values may be explained by the differences between coffee species or varieties [44], leading to differences in CFI accumulation potential, decrease, and stabilization at each stage of growth and development [45]. Moreover, the roasting conditions, typically due to changes in temperature and time depending on the roasting degree [8, 17, 46], also affect the CFI levels in coffee products. It is clear from Table 2 that the content of CFI decreased with the change from medium to dark roasting modes for both species. The higher temperature and longer roasting time applied in the dark mode compared to the medium mode potentially led to the partial decomposition of CFI. Similar observations have been noted regarding the variability of CFI in coffee products with different roasting degrees [47, 48].

Table 2. Concentrations of CFI and CGA in coffee products obtained from the proposed method and HPLC-DAD.

Coffee products	Proposed method		HPLC-DAD		t-values	
	CFI (%)	CGA (%)	CFI (%)	CGA (%)	CFI	CGA
ara-medi-1	1.29 ± 0.01	2.18 ± 0.01	1.33 ± 0.01	2.22 ± 0.01	0.231	0.231
ara-medi-2	1.30 ± 0.01	2.25 ± 0.01	1.35 ± 0.01	2.31 ± 0.01	0.289	0.346
ara-medi-3	1.30 ± 0.01	2.30 ± 0.01	1.36 ± 0.01	2.33 ± 0.01	0.346	0.173
ara-dark-1	1.04 ± 0.02	0.55 ± 0.01	1.10 ± 0.02	0.58 ± 0.01	0.173	0.173
ara-dark-2	1.05 ± 0.01	0.62 ± 0.01	1.17 ± 0.01	0.65 ± 0.01	0.693	0.173
ara-dark-3	1.04 ± 0.01	0.60 ± 0.02	1.05 ± 0.02	0.64 ± 0.01	0.037	0.146
ro-medi-1	2.12 ± 0.04	1.95 ± 0.03	2.20 ± 0.04	1.92 ± 0.03	0.115	0.058
ro-medi-2	2.10 ± 0.02	2.13 ± 0.03	2.07 ± 0.02	2.14 ± 0.03	0.087	0.019
ro-medi-3	2.08 ± 0.02	2.12 ± 0.03	2.09 ± 0.02	2.11 ± 0.02	0.029	0.023
ro-dark-1	1.78 ± 0.03	1.78 ± 0.02	1.76 ± 0.03	1.75 ± 0.02	0.038	0.087
ro-dark-2	1.66 ± 0.02	1.20 ± 0.01	1.65 ± 0.02	1.22 ± 0.01	0.029	0.115
ro-dark-3	1.71 ± 0.02	1.60 ± 0.03	1.73 ± 0.02	1.65 ± 0.02	0.058	0.113
ara-dark-5-butter-1	0.94 ± 0.02	0.55 ± 0.01	1.05 ± 0.02	0.59 ± 0.02	0.318	0.146
ara-dark-5-butter-2	0.95 ± 0.01	0.52 ± 0.01	0.99 ± 0.01	0.50 ± 0.02	0.231	0.073
ara-dark-5-butter-3	0.97 ± 0.01	0.50 ± 0.02	1.06 ± 0.02	0.57 ± 0.01	0.329	0.110

With CGA, the ara-medi-2 sample showed the highest concentration (2.25 ± 0.01% w/w) while the ara-dark-5-butter-2 sample had the lowest (0.52 ± 0.01% w/w). The results in Table 2 indicate that both Robusta and Arabica varieties showed similar results under medium roasting mode. The differences in the CGA values of these two coffee varieties became greater when the roasting mode was changed from medium to dark. As with caffeine, the dark roasting mode resulted in a decrease in CGA content compared to the medium roasting mode. This result is supported by Belay and Gholap (2009), demonstrating the decrease in CGA concentration and its decomposition (up to 52% of CGA for dark roasting mode) with higher roasting temperature [49]. When the roasting mode was changed from medium to dark, the CGA concentrations decreased more than those of CFI (Table 2). This could be explained because CFI is more stable than CGA during roasting [13, 19]. From the results of this study, the decreasing rate of CGA concentrations (from medium to dark roast) varied between the two coffee varieties: the CGA content decreased sharply by approximately 73.8% for Arabica coffee products, but only dropped by about 27.2% for Robusta products. Ludwig et al. (2014) also reported a nearly 90 %, 76 %, and 54 % loss of CGA for dark, medium, and light roasting degrees, respectively (compared to the CGA content in green coffee beans) [13]. Therefore, it could be concluded that CGA as a bioactive compound was affected not only by the temperature during roasting but was also dependent on the coffee variety [11, 17, 19, 50, 51] This is due to the structural dissimilarity of the coffee beans themselves, leading to differences in the decomposition rate of CGA under the same roasting mode. The roasting activity caused the gradual decomposition and/or transformation of CGA with approximately 8 – 10 % lost for every 1 % of dried matter loss. The isomerization of CGA, along with partial hydrolysis, may create quinic acid and other cinnamic acids [52]. The cinnamic acid released from the breakdown of CGA could participate in further chemical reactions to form different flavour components of coffee products. Hence, the decomposition state of CGA can be used as an indicator of roasting degree [53].

CONCLUSION

This analytical method, based on first-order derivative spectra from a UV-Vis instrument, was investigated and evaluated for the simultaneous determination of CFI and CGA in roasted ground coffee products. The method performance was in accordance with the requirements in Appendix F of AOAC (2016). There was no statistically significant difference in the results obtained from the proposed method compared to the HPLC-DAD reference method. The present method is a quick, easy and inexpensive analytical approach. It could allow coffee producers to perform regular monitoring of CFI and CGA concentrations during processing without requiring the use of expensive and complicated instruments. Additionally, the proposed method demonstrates potential for the design of a

portable device (separate devices for CFI and CGA or a combined device for simultaneous measurement) that could quickly determine the concentrations of CFI and CGA in coffee extracts. Such a low-cost and portable tool could be used by small businesses and consumers to easily increase their knowledge of coffee and/or coffee-related products.

ACKNOWLEDGEMENTS

The authors would like to express their grateful thanks and appreciation to Nguyen Tat Thanh University and the University of Science, Vietnam National University Ho Chi Minh City, for their assistance and support during this study.

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