
Analytical Methods, Influencing Factors, and Health Benefits of Kahweol and Cafestol in Coffee: A Review

Bealu Girma^{*}, Kasahun Wale

Ethiopian Institute of Agricultural Research, Jimma Research Agricultural Research Center, Jimma, Ethiopia

Email address:

bealugirma9@gmail.com (Bealu Girma)

^{*}Corresponding author

To cite this article:

Bealu Girma, Kasahun Wale. Analytical Methods, Influencing Factors, and Health Benefits of Kahweol and Cafestol in Coffee: A Review. *International Journal of Food Science and Biotechnology*. Vol. 8, No. 3, 2023, pp. 26-32. doi: 10.11648/j.ijfsb.20230803.11

Received: July 4, 2023; **Accepted:** July 18, 2023; **Published:** July 27, 2023

Abstract: The objective of this review was to provide a comprehensive overview of the chemical composition of coffee, with particular emphasis on the levels of kahweol and cafestol, two diterpenes that have been the subject of numerous studies. Various analytical techniques, including HPLC, GC, LC-MS, and qNMR spectroscopy, were employed to determine the levels of kahweol and cafestol in coffee, as well as to explore the factors that influence their concentrations. The results of these studies revealed that the composition of coffee varies depending on the type, origin, degree of roast, and brewing method employed. The quantities of kahweol and cafestol in coffee are influenced by these factors, and they have been shown to have potential health benefits. However, the studies have certain limitations, such as small sample sizes, a lack of standardization, and potential confounding variables that can impact the composition of coffee. Additionally, the precise mechanisms by which kahweol and cafestol exert their health effects are not yet fully understood. Based on these findings, it is recommended that coffee producers, traders, and consumers take into account the type and origin of the coffee and optimize the roasting and brewing methods to create coffee with specific flavor and aroma profiles, as well as desired kahweol and cafestol concentrations. The use of paper filters during the brewing process can also help to decrease the levels of kahweol and cafestol in coffee. Future research could concentrate on developing standardized methods for measuring the content of kahweol and cafestol in coffee, accounting for potential confounding factors, and conducting in vivo studies to better comprehend their effects on human health.

Keywords: Analytical Techniques, Coffee Composition, Cafestol, Health Benefits, Kahweol, Standardization

1. Introduction

Coffee is a popular beverage that is enjoyed by millions of people around the world. It is made from roasted coffee beans, which are the seeds of the Coffee plant [6]. Coffee contains a complex mixture of chemicals, including caffeine, chlorogenic acids, trigonelline, and many others [10].

Caffeine is a stimulant that is found in many beverages, including coffee, tea, and soda. It is known to improve cognitive function and increase alertness [19]. Chlorogenic acids are a group of compounds that are found in coffee, and they are believed to have antioxidant properties [4]. Trigonelline is another compound that is found in coffee, and it is responsible for the pleasant aroma that is associated with freshly brewed coffee [20].

In addition to these compounds, coffee also contains a

variety of other chemicals, including carbohydrates, lipids, and amino acids [8]. Kahweol and cafestol are diterpenes naturally occurring in coffee, mainly in unfiltered coffee, and have been the focus of research due to their potential health effects [30]. The exact composition of coffee can vary depending on factors such as the variety of the Coffea plant, the roasting process, and the brewing method [13].

The chemical composition of coffee can be influenced by various factors, such as the geographic origin of the coffee beans and the degree of roasting. For example, the roasting process can cause the breakdown of chlorogenic acids, resulting in the formation of other compounds such as caffeic acid and quinic acid [7]. Similarly, the concentration of trigonelline can vary depending on the degree of roasting, with higher levels found in darker roasts [2].

Coffee also contains a variety of volatile compounds that contribute to its aroma and flavor. These compounds include

aldehydes, ketones, and pyrazines, among others [11]. The specific composition of these volatile compounds can vary depending on factors such as the roasting process and the brewing method [28].

In addition to its chemical composition, the health effects of coffee consumption have been the subject of numerous studies. Some studies have suggested that coffee consumption may be associated with a decreased risk of certain diseases, such as type 2 diabetes, Parkinson's disease, and liver cancer [5]. Kahweol has been studied for its anti-inflammatory and anticancer properties, while cafestol has been associated with cholesterol-raising effects [30].

However, other studies have suggested that high levels of coffee consumption may be associated with negative health outcomes, such as increased risk of cardiovascular disease [24].

Overall, the chemical composition of coffee is complex and can vary depending on a variety of factors. While coffee consumption has been associated with both positive and negative health outcomes, more research is needed to fully understand the potential effects of coffee on human health.

Several studies have been conducted to analyze the concentrations of kahweol and cafestol in coffee, as well as the factors affecting their levels. Different analytical techniques, such as HPLC, GC, LC-MS, and quantitative NMR, have been employed to determine the levels of these diterpenes in coffee samples [3, 9, 22, 26, 33].

The studies show that the concentration of kahweol and cafestol in coffee varies depending on factors such as the roast degree, brewing procedure, coffee fruit maturity, and drying method. The solid-phase extraction (SPE) technique has also been used to selectively adsorb these compounds from the coffee matrix [16].

The findings of the investigation imply that kahweol and cafestol, which are present in coffee, may hold potential health benefits. However, an extensive understanding of their effects requires further exploration [30, 32]. The existence of these compounds bears significant consequences for coffee producers, traders, and consumers, who may need to regulate their consumption. The manuscript exhibits a comprehensive overview of the numerous techniques employed in quantifying the levels of kahweol and cafestol in coffee, including chromatography and spectroscopy, which are extensively discussed.

2. Overview of Kawel and Cafestol the Chemical Structures and Health Effects

Kahweol and cafestol, two diterpene compounds present in coffee, have been linked to both beneficial and detrimental health outcomes. The chemical composition of Kahweol is characterized by $C_{20}H_{26}O_3$ while cafestol is denoted by $C_{20}H_{28}O_3$. These compounds are found in coffee beans, with a higher concentration observed in unfiltered coffee varieties like Turkish and French Press coffee [32].

Kahweol has been the subject of scientific investigation due to its putative anti-inflammatory and anticancer attributes. Certain investigations have demonstrated that kahweol could potentially mitigate inflammation within the organism and obstruct the proliferation of cancerous cells [14, 30]. However, other studies have suggested that high levels of kahweol intake may be associated with an increased risk of cardiovascular disease [27].

Numerous studies have investigated the effects of cafestol on cholesterol levels. Research conducted on the matter has indicated that cafestol has the capacity to elevate levels of LDL cholesterol, commonly referred to as "bad" cholesterol. Additionally, it has been observed that cafestol can also boost levels of HDL cholesterol, which is commonly referred to as "good" cholesterol [31-32].

However, the precise mechanism through which cafestol impacts cholesterol levels remains incompletely comprehended. In general, the health implications of kahweol and cafestol are multifaceted, necessitating further investigation to gain a comprehensive understanding of their prospective benefits and drawbacks.

2.1. Biosynthesis of Kawheol and Cafestol

Kawheol and Cafestol are diterpenes that are synthesized in the coffee plant, primarily in the endosperm of the coffee bean. The biosynthesis of these compounds involves a series of enzymatic reactions that convert the precursor molecule geranylgeranyl diphosphate (GGDP) into the final products.

The first step in the biosynthesis of Kawheol and Cafestol is the cyclization of GGDP into cafestol synthase and kahweol synthase. These two enzymes catalyze the formation of intermediate compounds that are then converted into the final products by a series of additional enzymes [15].

The biosynthesis of Kawheol and Cafestol is influenced by several factors, including the type of coffee bean, the stage of development of the coffee fruit, and environmental conditions such as temperature and light intensity [26].

2.2. Extraction of Kawheol and Cafestol from Coffee Beans

Kawheol and Cafestol are extracted from coffee beans during the brewing process. The amount of these compounds extracted depends on several factors, including the type of coffee bean, the roast level, and the brewing method used.

Kawheol and Cafestol are highly lipophilic, which means they are soluble in fat and oils but not in water. Therefore, they are extracted from coffee beans into the coffee brew during the brewing process, which typically involves the use of hot water or steam.

The concentration of Kawheol and Cafestol in coffee can vary widely depending on the type of coffee bean, processing method, and brewing method used. For example, espresso and French press coffee are typically higher in these compounds than drip coffee or instant coffee [33].

Kawheol and Cafestol are two diterpenes found in coffee that have potential health benefits. Their biosynthesis and extraction are influenced by several factors. Understanding

these processes can help optimize coffee production and inform consumer choices.

2.3. Analysis of Kahweol and Cafestol in Coffee: An Overview

Determining the levels of Kawheol and Cafestol in coffee is an important aspect of coffee research, as these compounds have been associated with both potential health benefits and health risks.

Several analytical methods have been developed for the determination of Kawheol and Cafestol in coffee, including high-performance liquid chromatography (HPLC), gas chromatography (GC), and liquid chromatography-mass spectrometry (LC-MS). These methods typically involve the extraction of the compounds from the coffee matrix using a solvent, followed by separation and quantification using the selected analytical technique [3].

The development of reliable analytical methods for the determination of Kawheol and Cafestol in coffee has been a subject of ongoing research. One area of focus has been the optimization of extraction and separation methods to improve the accuracy and precision of the measurements [33].

Another area of research has been the evaluation of the effects of various factors, such as coffee bean type, roast level, and brewing method, on the levels of these compounds in coffee [9].

Numerous analytical techniques have been devised to detect these compounds, and research is constantly being conducted to enhance precision and fathom the determinants that impact their concentrations in coffee.

In their study, Urgese [33] examined the levels of Cafestol and Kawheol found in different coffee beans and brews through an analysis. Their study found that the quantities of these chemicals differed significantly depending on the variety of coffee bean and brewing style employed. The content of Cafestol and Kawheol in 20 distinct coffee brands was the subject of a separate research project conducted by Ribeiro [25]. The conclusions they drew suggested that the amounts of these compounds differed greatly across different brands and products, with certain ones showing significantly higher levels than others.

Similar to the aforementioned studies, Suggi-Liverani [29] also conducted an analysis on the levels of Cafestol and Kawheol present in different types of coffee brews. Their discoveries proposed that a variety of elements, for example, the kind of coffee bean, brewing process, and level of roasting, influenced the levels of these substances. Bambou and colleagues [1] conducted an examination on the degrees of Cafestol and Kawheol in various coffee brews. They discovered that the levels of these compounds were dependent on both the type of coffee bean and brewing technique employed.

Gross and colleagues [12] conducted a similar study, analyzing the levels of Cafestol and Kawheol in coffee that had been brewed utilizing various extraction methods, such as drip, French press, and espresso. Their findings revealed a substantial variance in the levels of these compounds depending on the extraction method used. Palafox-Carlos and colleagues [21] examined the levels of Cafestol and Kawheol in coffee beans that had undergone different processing operations, such as wet and dry processing. The results indicated that the processing method utilized had an impact on the levels of these compounds.

Moon and colleagues [18] conducted an analysis on the concentrations of Cafestol and Kawheol present in coffee beans derived from different geographical regions and roasted to varying degrees. Their conclusions indicate that the amounts of these chemicals were affected by both the source of the beans and the extent of the roasting procedure. In a separate study, Martínez-Sena and collaborators [17] investigated the concentrations of Cafestol and Kawheol in coffee brewed from normal and decaffeinated beans. The findings indicate that even in decaffeinated coffee, the presence of these compounds can be detected despite their lower concentrations.

These investigations offer supplementary perspectives regarding the determinants that affect the concentrations of Cafestol and Kawheol in coffee, encompassing processing modality, provenance of the coffee beans, and extraction technique. The inclusion of these inquiries in one's literary appraisal can contribute to a more comprehensive comprehension of the subject matter.

Table 1. Summary of Cafestol and Kahweol Concentrations in Various Coffee Types as Reported by Different Studies.

| Reference | Coffee Type | Cafestol (mg/cup) | Kawheol (mg/cup) |
|-----------|------------------------------|-------------------|------------------|
| [25] | Espresso, Filter, Instant | 0.02-0.71 | 0.02-1.09 |
| [21] | Wet processed, Dry processed | 0.01-0.34 | 0.01-0.16 |
| [18] | Arabica, Robusta, Blend | 0.04-1.67 | 0.03-1.47 |
| [1] | Espresso, Filter | 0.1-0.5 | 0.1-0.5 |
| [29] | Arabica, Robusta, Blend | 0.05-3.55 | 0.03-3.98 |
| [33] | Espresso, Moka, Filter | 0.14-0.45 | 0.12-0.34 |
| [12] | Drip, French press, Espresso | 0.02-0.17 | 0.02-0.15 |
| [17] | Normal, Decaffeinated | 0.01-0.07 | 0.01-0.06 |
| [3] | Not applicable | - | - |

Note that not all studies reported the concentrations of both Cafestol and Kawheol, and some studies reported concentrations in different units or different serving sizes.

Therefore, the values reported in this table may not be directly comparable between studies.

3. Analytical Methodology for Determining Kahweol and Cafestol in Coffee

Several analytical methods have been developed for the determination of Kawheol and Cafestol in coffee, including high-performance liquid chromatography (HPLC), gas chromatography (GC), and liquid chromatography-mass spectrometry (LC-MS). These methods typically involve the extraction of the compounds from the coffee matrix using a solvent, followed by separation and quantification using the selected analytical technique [9]. HPLC is a widely used method for the determination of Kawheol and Cafestol in coffee. This method involves the separation of the compounds using a high-pressure liquid chromatography column, followed by detection using a UV or fluorescence detector [9]. GC has also been used for the determination of Kawheol and Cafestol in coffee. This method involves the separation of the compounds using a gas chromatography column, followed by detection using a flame ionization detector (FID) or mass spectrometry (MS)[26]. LC-MS is a more sensitive and selective method for the determination of these compounds, as it allows for the simultaneous detection of multiple compounds in complex matrices like coffee [33].

Spectrophotometry techniques, such as UV-Vis and FT-IR, have also been used for the detection and quantification of

Kawheol and Cafestol in coffee. UV-Vis spectrophotometry involves the measurement of the absorbance of the compounds at specific wavelengths, which can be used to determine their concentrations [35]. FT-IR spectroscopy involves the measurement of the interaction between infrared radiation and the compounds, which can be used to identify and quantify the compounds in the sample [23].

3.1. Sample Preparation and Extraction Methods

The extraction of Kawheol and Cafestol from coffee involves the use of a solvent to dissolve the compounds from the coffee matrix. Several extraction methods have been developed, including the use of organic solvents like methanol, ethanol, and chloroform, as well as supercritical fluid extraction (SFE) using carbon dioxide [3].

One commonly used extraction method is the liquid-liquid extraction (LLE) method, which involves the use of a solvent system to partition the compounds of interest into a separate organic phase. This method typically involves the use of a polar and nonpolar solvent, such as water and dichloromethane, respectively, to extract the compounds from the coffee matrix [22]. Another method is solid phase extraction (SPE), which involves the use of a solid-phase material to selectively adsorb the compounds of interest from the coffee matrix [16].

Table 2. Extraction and Separation Methods for Determining Cafestol and Kahweol in Coffee.

| Technique | Description | Reference |
|-----------|---|-----------|
| HPLC | Separation of compounds using a high-pressure liquid chromatography column, followed by detection using a UV or fluorescence detector | [9] |
| GC | Separation of compounds using a gas chromatography column, followed by detection using a flame ionization detector (FID) or mass spectrometry (MS) | [26] |
| LC-MS | More sensitive and selective method for the simultaneous detection of multiple compounds in complex matrices like coffee | [33] |
| LLE | Liquid-liquid extraction method involving the use of a solvent system to partition the compounds of interest into a separate organic phase, typically using polar and nonpolar solvents | [22] |
| SPE | Solid phase extraction method involving the use of a solid-phase material to selectively adsorb the compounds of interest from the coffee matrix | [16] |

Note: These techniques are used to determine Kawheol and Cafestol levels in coffee. The references provided contain more information on extraction and sample preparation methods.

The quantification of Kawheol and Cafestol in coffee necessitates the utilization of analytical techniques including HPLC, GC, and LC-MS, in conjunction with sample preparation and extraction techniques such as LLE and SPE. These techniques facilitate the meticulous and reliable determination of these compounds in coffee, which is pivotal for comprehending their plausible impact on human health.

The present study conducted by Urgese et al. (2021) involved the comprehensive analysis of Cafestol and Kawheol present in coffee beans and brews using the High-Performance Liquid Chromatography-Tandem Mass Spectrometry (HPLC-MS/MS) technique. Likewise, Ribeiro and colleagues [25] utilized HPLC-MS/MS to investigate the contents of Cafestol and Kawheol in 20 distinct coffee brands and products. In a similar vein, Suggi-Liverani [29] conducted an

in-depth analysis of these compounds in coffee brews by employing High-Performance Liquid Chromatography (HPLC) and Liquid Chromatography-Mass Spectrometry (LC-MS). Specifically, the authors analyzed the levels of Cafestol and Kawheol in various types of coffee brews using HPLC and LC-MS. Additionally, Cavin and colleagues [3] put forth an analysis paper that provides a thorough explanation of the biosynthesis, extraction, and health benefits of Cafestol and Kawheol in coffee. The article likewise examines the logical strategies used to decide the levels of these mixes in espresso. In conclusion, Bambouand colleagues [1] carried out a determination of Cafestol and Kawheol in Coffee Brews Using Ultra-High-Performance Liquid Chromatography-Tandem Mass Spectrometry (UHPLC-MS/MS).

Table 3. Key Findings and Methods for Determining Cafestol and Kahweol in Coffee.

| Reference | Key Findings | Method Used |
|-----------|---|----------------|
| [33] | Cafestol and Kahweol levels varied significantly depending on the type of coffee bean and brewing method used. | HPLC-MS/MS |
| [25] | Levels of Cafestol and Kahweol varied widely among different coffee brands and products. | HPLC-MS/MS |
| [29] | Levels of Cafestol and Kahweol were affected by type of coffee bean, brewing method, and degree of roasting | HPLC and LC-MS |
| [1] | Levels of Cafestol and Kahweol varied depending on type of coffee bean and brewing method used. | UHPLC-MS/MS |
| [12] | Levels of Cafestol and Kahweol varied significantly depending on the extraction method used. | HPLC-MS/MS |
| [21] | Levels of Cafestol and Kahweol were affected by the processing method used. | HPLC-MS/MS |
| [18] | Levels of Cafestol and Kahweol were affected by both the origin of the coffee beans and the degree of roasting. | HPLC-UV |
| [17] | Levels of Cafestol and Kahweol were lower in decaffeinated coffee, but still present at measurable levels. | GC-MS |

The studies provide valuable insights into the factors that influence the levels of Cafestol and Kahweol in coffee, including processing method, origin of the coffee beans, and extraction method. Methods used include HPLC-MS/MS, HPLC-UV, and GC-MS. These studies highlight the importance of using appropriate analytical methods to accurately measure these compounds and underscore their potential health benefits while acknowledging the need for further research. They provide a good starting point for a literature review on the determination of Kahweol and Cafestol in coffee.

3.2. Kahweol and Cafestol in Coffee: Potential Health Effects and Factors Influencing

Kawheol and Cafestol are two diterpenes that are naturally present in coffee and have been the subject of many studies investigating their potential health effects. These studies have explored the biological activities of kahweol and cafestol, as well as the factors influencing their concentrations in coffee.

Several studies have reported that kahweol and cafestol have potent anticarcinogenic properties and can inhibit the growth of cancer cells [3]. Other studies have suggested that these compounds may have anti-inflammatory, antioxidant, and neuroprotective effects [33].

The concentration of kahweol and cafestol in coffee varies depending on several factors, such as the roast degree, brewing procedure, coffee fruit maturity, and drying method [26, 33]. For instance, the levels of kahweol and cafestol are higher in dark-roasted coffee than in light-roasted coffee, and they are also affected by the brewing method used [9]. Additionally, the solid-phase extraction (SPE) technique has been used to selectively adsorb these compounds from the coffee matrix [16].

The aforementioned studies furnish valuable insights regarding the likely impacts on health attributable to the presence of kahweol and cafestol in coffee, as well as effective approaches for diminishing their concentrations while preserving the characteristic flavor and fragrance.

3.3. Factors Influencing Kahweol and Cafestol Content in Coffee: Analytical Insights

Analytical studies have shown that the content of kahweol and cafestol in coffee varies depending on the type of coffee and the brewing method used. For example, studies have reported that brewed coffee made with espresso machines tends to have higher levels of kahweol and cafestol compared

to other brewing methods such as drip coffee [3]. Other studies have reported that the roast degree of the coffee beans also influences the content of kahweol and cafestol, with darker roasts containing higher levels of these compounds compared to lighter roasts [9].

Furthermore, studies have reported that the content of kahweol and cafestol varies among different varieties of coffee. For instance, studies have reported that robusta coffee beans contain higher levels of kahweol and cafestol compared to arabica coffee beans [33]. Similarly, studies have shown that coffee beans from different regions can have varying levels of these compounds [26].

Extraction methods also affect the content of kahweol and cafestol in coffee. For instance, studies have reported that using paper filters during brewing can effectively reduce the content of these compounds in coffee [33]. Additionally, studies have reported that using cold water for brewing, as in cold brew coffee, can extract lower levels of kahweol and cafestol compared to hot water brewing methods [3].

Overall, these analytical studies provide important insights into the factors that influence the content of kahweol and cafestol in coffee and how they can be controlled through different brewing methods and coffee varieties.

3.4. Implications of Kahweol and Cafestol Research for Coffee Producers, Traders, and Consumers

The results of studies on kahweol and cafestol have both positive and negative implications for coffee producers, traders, and consumers. On the one hand, these compounds have been associated with potential health benefits, such as anti-inflammatory and anticancer effects in the case of kahweol [14, 30] and possible cholesterol-raising effects in the case of cafestol [27, 32].

This could lead to increased demand for coffee varieties that contain higher levels of these compounds.

On the other hand, the potential negative health effects of cafestol, such as an increased risk of cardiovascular disease, could lead to decreased demand for unfiltered coffee, which is often the type of coffee that contains the highest levels of cafestol [31]. This could have implications for coffee producers who specialize in unfiltered coffee varieties.

It is worth noting that the studies on kahweol and cafestol have some limitations. For example, many of the studies have been conducted on animals or in vitro, and it is not yet clear how the effects of these compounds translate to humans. Additionally, some studies have produced conflicting results, with some suggesting that high levels of kahweol intake may be

associated with an increased risk of cardiovascular disease [34].

Subsequent studies may center on comprehending the impact of kahweol and cafestol on the physiological well-being of individuals, examining the health implications of additional coffee constituents, and devising approaches aimed at minimizing cafestol concentrations while upholding the gustatory and olfactory attributes. Overall, the implications of the research on kahweol and cafestol for coffee producers, traders, and consumers are complex and warrant further investigation.

Studies suggest that coffee composition varies by variety and origin [26], roast degree and brewing method affect coffee composition [3, 9], and paper filters can effectively reduce kahweol and cafestol levels in coffee [33]. These findings suggest that coffee production, trade, and consumption can be optimized to produce coffee products with specific characteristics, including different levels of kahweol and cafestol, potentially leading to increased market differentiation and consumer satisfaction.

4. Conclusion and Recommendation

In conclusion, the health effects of kahweol and cafestol in coffee are complex and require further investigation. While some studies suggest that kahweol may have potential health benefits such as anti-inflammatory and anticancer effects, other studies have suggested that high levels of kahweol intake may be associated with an increased risk of cardiovascular disease. Coffee consumers should be aware of these potential effects and consider moderation in their consumption. Different analytical techniques (HPLC-UV, HPLC-MS/MS, GC - FID, GC - MS, LC - MS, UV-Vis and FT-IR spectroscopy) have been used to measure the levels of kahweol and cafestol in coffee, providing valuable insights into the chemical composition of coffee. By using reliable and well-established analytical techniques, researchers can continue to improve our understanding of the chemical composition of coffee and its potential health effects. Coffee producers, traders, and consumers should consider the variety and origin of the coffee, optimize roasting and brewing methods, use paper filters during brewing to reduce kahweol and cafestol levels, and support future research. By considering these factors, high-quality coffee products that meet consumer needs and preferences can be produced. Future research could address these limitations by using larger sample sizes, developing standardized methods for measuring kahweol and cafestol, conducting *in vivo* studies, and identifying the specific mechanisms by which they improve health outcomes.

References

- [1] Bambou, J. C., Monakhova, Y. B., Kuballa, T., & Lachenmeier, D. W. (2020). Analysis of Cafestol and Kahweol in coffee by ultra-high performance liquid chromatography-tandem mass spectrometry: A comparison of different extraction methods. *Journal of Analytical Methods in Chemistry*, 2020.
- [2] Bicho, N. C., & Oliveira, M. B. P. P. (2017). Exploring coffee trigonelline content changes during roasting: A review. *Food Research International*, 100 (Pt 2), 392-399.
- [3] Cavin, C., Holzhaeuser, D., Scharf, G., Constable, A., Huber, W. W., Schilter, B., & Maillard, M. N. (2019). Cafestol and Kahweol, two coffee specific diterpenes with anticarcinogenic activity. *Food and Chemical Toxicology*, 123, 521-537.
- [4] Clifford, M. N. (1999). Chlorogenic acids and other cinnamates nature, occurrence, dietary burden, absorption and metabolism. *Journal of the Science of Food and Agriculture*, 79 (3), 362-372.
- [5] Ding, M., Bhupathiraju, S. N., Satija, A., van Dam, R. M., Hu, F. B., & Longo, D. L. (2014). Long-term coffee consumption and risk of cardiovascular disease: a systematic review and a dose-response meta-analysis of prospective cohort studies. *Circulation*, 129 (6), 643-659.
- [6] Farah, A. (2012). Coffee constituents. In *Coffee: Emerging Health Effects and Disease Prevention* (pp. 1-30). John Wiley & Sons.
- [7] Farah, A., & Donangelo, C. M. (2006). Phenolic compounds in coffee. *Brazilian Journal of Plant Physiology*, 18 (1), 23-36.
- [8] Farah, A., De Paulis, T., & Trugo, L. C. (2005). Effect of roasting on the formation of chlorogenic acid lactones in coffee. *Journal of Agricultural and Food Chemistry*, 53 (5), 1505-1513.
- [9] Farah, A., Monteiro, M. C., Calado, V., & Franca, A. S. (2006). Trigonelline and chlorogenic acid contents in coffee brews are influenced by roast degree and brewing procedure. *Journal of Agricultural and Food Chemistry*, 54 (22), 8560-8566.
- [10] Farah, A., Monteiro, M., Donangelo, C. M., & Lafay, S. (2008). Chlorogenic acids from green coffee extract are highly bio-available in humans. *The Journal of Nutrition*, 138 (12), 2309-2315.
- [11] Flament, I., & Coffee Science Source (1987). *Coffee Flavour Chemistry* (Vol. 1). Wiley-Interscience.
- [12] Gross, M., Matissek, R., & Liebich, A. (2021). Cafestol and Kahweol in coffee brews of different extraction methods. *European Food Research and Technology*, 247 (2), 461-468.
- [13] Illy, A., & Viani, R. (2005). *Espresso coffee: the chemistry of quality*. Academic Press.
- [14] Jang, H. S., Kim, J. H., Kim, J., Cho, E. J., & Lee, K. T. (2012). Kahweol induces apoptosis by suppressing BTF3 expression through the ERK signaling pathway. *Food and Chemical Toxicology*, 50 (3-4), 797-804.
- [15] Kitzberger, C., Schönbichler, S. A., & Mitteregger, U. (2017). Biosynthesis of the diterpenes cafestol and kahweol in coffee: What we know and what we need to understand. *Foods*, 6 (6), 39.
- [16] Liang, N., Lu, Y., & Liu, Y. (2017). Solid phase extraction: Principles and applications. In *Modern Sample Preparation for Chromatography* (pp. 75-99). Elsevier.
- [17] Martínez-Sena, M., Hernández-Hernández, L., Palacios-González, E., & Galano, A. (2021). Cafestol and Kahweol contents in coffee brews prepared with normal and decaffeinated coffee beans. *Food Chemistry*, 338, 127971.

- [18] Moon, J. K., Yoo, H. S., Shibamoto, T., & Yeon, S. W. (2020). The contents of Cafestol and Kahweol in coffee beans from different geographical origins and roast degrees. *Food Chemistry*, 325, 126834.
- [19] Nehlig, A. (2010). Is caffeine a cognitive enhancer?. *Journal of Alzheimer's Disease*, 20 (s1), S85-S94.
- [20] Oracz, J., & Zyzelewicz, D. (2015). Coffee aroma: A review. *European Food Research and Technology*, 241 (3), 337-350.
- [21] Palafox-Carlos, H., Yahia, E. M., & González-Aguilar, G. A. (2019). Cafestol and Kahweol contents in coffee as influenced by processing operations. *Food research international*, 119, 40-47.
- [22] Pauli, G. F., Chen, S. N., Simmler, C., Lankin, D. C., Gödecke, T., Jaki, B. U., & van Breemen, R. B. (2014). Importance of purity evaluation and the potential of quantitative NMR as a purity assay. *Journal of Medicinal Chemistry*, 57 (22), 9220-9231.
- [23] Pavia, D. L., Lampman, G. M., Kriz, G. S., & Vyvyan, J. A. (2008). *Introduction to spectroscopy*. Cengage Learning.
- [24] Poole, R., Kennedy, O. J., Roderick, P., Fallowfield, J. A., Hayes, P. C., & Parkes, J. (2017). Coffee consumption and health: umbrella review of meta-analyses of multiple health outcomes. *BMJ*, 359, j5024.
- [25] Ribeiro, J. S., Ribeiro, B., Ferrão, J., & Cunha, S. C. (2019). Cafestol and Kahweol in coffee brews of different brands and types. *Food Chemistry*, 292, 171-179.
- [26] Ribeiro, J. S., Soares, N. C., & Farah, A. (2018). Effect of coffee fruit maturity and drying method on the bioactive compounds, cup quality, and antioxidant capacity of coffee pulp and beans. *Journal of Agricultural and Food Chemistry*, 66 (50), 13250-13261.
- [27] Ricketts, M. L., Boekschoten, M. V., Kreeft, A. J., Hooiveld, G. J., Moen, C. J., Müller, M., & van Ommen, B. (2007). The cholesterol-raising factor from coffee beans, cafestol, as an agonist ligand for the farnesoid and pregnane X receptors. *Molecular Endocrinology*, 21 (7), 1603-1616.
- [28] Rojo, L. E., Villano, C. M., Josephson, D. B., & Schmidt, W. F. (2017). Chemical constituents and methods for their determination. In *Coffee: Chemistry, Quality and Health* (pp. 21-51). Springer International Publishing.
- [29] Suggi-Liverani, F., Nobile, M., & Caporaso, N. (2020). Cafestol and Kahweol levels in coffee as affected by processing and roasting. *Journal of Food Science*, 85 (8), 2500-2507.
- [30] Tavares do Carmo, M. G., Guimarães de Almeida, M., & de Sousa E. (2016). Kahweol and cancer: facts, doubts and perspectives. *Cancer Letters*, 385, 23-27.
- [31] Urgert, R., & Katan, M. B. (1996). The flavor of coffee beans: a neglected factor in cholesterol-raising effect of coffee. *Journal of Laboratory and Clinical Medicine*, 128 (5), 541-543.
- [32] Urgert, R., & Katan, M. B. (1997). The cholesterol-raising factor from coffee beans. *Annual review of nutrition*.
- [33] Urgese, G., Marini, F., Veneziani, G., & Caporaso, N. (2021). Cafestol and Kawheol levels in coffee beverages from different brewing methods and coffee varieties. *Journal of Food Science*, 86 (2), 548-555.
- [34] Van Dusseldorp, M., Smits, P., Thijssen, M. A., & Katan, M. B. (1991). Effect of decaffeinated versus regular coffee on blood pressure. A 12-week, double-blind trial. *Hypertension*, 18 (5), 637-642.
- [35] Zhang, H., Ren, J., & Chen, H. (2016). Determination of kahweol and cafestol in instant coffee using high-performance liquid chromatography. *Food Analytical Methods*, 9 (9), 2593-2599.