

Determination of Caffeine Content in Green *Coffea liberica* Beans by UV–Visible Spectrophotometry and Evaluation of Its Effect on Weight Reduction

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ABSTRACT

Background: Green coffee beans (*Coffea liberica*) are a rich source of bioactive compounds, including caffeine, chlorogenic acid, flavonoids, and polyphenols, which have potential health-promoting properties. Caffeine has been reported to enhance thermogenesis, stimulate fat oxidation, and increase energy expenditure, making it a promising natural compound for weight management. This study aimed to determine the caffeine content of green *Coffea liberica* beans using UV–Visible spectrophotometry and to evaluate the implementation of green coffee extract consumption on body weight reduction. **Methods:** An experimental study was conducted by extracting green coffee beans with 96% ethanol followed by phytochemical screening and quantitative determination of caffeine using UV–Visible spectrophotometry at a maximum wavelength of 272 nm. The analytical method was validated by assessing linearity, accuracy, and precision. A one-group pretest–posttest design was employed to evaluate the implementation of green coffee extract in overweight adults with a body mass index (BMI) ≥ 25 kg/m². Body weight was measured before and after the intervention, and statistical analysis was performed at a 95% confidence level. **Results:** The spectrophotometric method demonstrated satisfactory linearity, accuracy, and precision, indicating its suitability for caffeine determination. Green *Coffea liberica* beans were confirmed to contain caffeine and other bioactive compounds that contribute to antioxidant activity. Consumption of green coffee extract showed a tendency to reduce body weight following the intervention period. **Conclusion:** UV–Visible spectrophotometry is a reliable method for determining caffeine content in green *Coffea liberica* beans. The presence of caffeine and other bioactive compounds supports the potential application of green Liberica coffee as a functional food ingredient for weight management.

Keywords: *Coffea Liberica*; Caffeine; UV–Visible Spectrophotometry; Green Coffee; Weight Reduction.

How to Cite:

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INTRODUCTION

Obesity has become one of the most significant global public health concerns due to its increasing prevalence and its association with numerous chronic diseases, including type 2 diabetes mellitus, hypertension, dyslipidemia, cardiovascular disease, and metabolic syndrome (World Health Organization [WHO], 2024). According to the World Obesity Federation (2024), the global prevalence of overweight and obesity continues to rise, affecting individuals across all age groups. Conventional pharmacological therapies for obesity often produce undesirable side effects and are relatively expensive, leading to growing interest in natural products as safer and more sustainable alternatives for weight management (Naveed et al., 2022).

Coffee is one of the most widely consumed beverages worldwide and represents one of the most economically valuable agricultural commodities. Indonesia is recognized as one of the largest coffee-producing countries, cultivating several commercial species, including *Coffea arabica*, *Coffea canephora* (Robusta), and *Coffea liberica* (International Coffee Organization [ICO], 2024). Among these species, *Coffea liberica* has received relatively limited scientific attention despite its adaptability to tropical environments and its unique phytochemical composition. Green coffee beans, particularly those of *C. liberica*, contain numerous bioactive compounds such as caffeine, chlorogenic acid, flavonoids, diterpenes, and other polyphenols that contribute to their biological activities (Bressani et al., 2021; Tran et al., 2023).

Caffeine (1,3,7-trimethylxanthine) is the principal alkaloid present in coffee beans and has been extensively investigated because of its physiological and pharmacological effects. As a central nervous system stimulant, caffeine enhances alertness, reduces fatigue, and improves physical performance (Heckman et al., 2020). More importantly, caffeine has been shown to increase thermogenesis, stimulate lipolysis, enhance fatty acid oxidation, and elevate resting energy expenditure, making it a promising natural compound for body weight management (Nawrot et al., 2021). These mechanisms contribute to negative energy balance and may support weight reduction when combined with appropriate dietary and lifestyle interventions.

The caffeine content of coffee varies considerably depending on coffee species, geographical origin, cultivation practices, environmental conditions, harvesting time, and post-harvest processing (Farah et al., 2020). In general, green coffee beans retain their original caffeine concentration because they have not undergone roasting, a process that can modify the chemical composition and sensory characteristics of coffee. Therefore, accurate determination of caffeine content is essential for quality control, product standardization, dosage formulation, and evaluation of the biological activity of green coffee products.

Several analytical techniques have been employed to quantify caffeine in coffee, including High-Performance Liquid Chromatography (HPLC), Gas Chromatography (GC), Capillary Electrophoresis, and UV–Visible Spectrophotometry (Jeszka-Skowron et al., 2022). Among these methods, UV–Visible spectrophotometry is widely applied because it is simple, rapid, cost-effective, and suitable for routine laboratory analysis. Caffeine exhibits a characteristic maximum absorbance at approximately 272–274 nm, enabling reliable quantitative determination after appropriate extraction and calibration procedures (Riyanto et al., 2021). Furthermore, analytical method validation, including linearity, accuracy, precision, limit of detection, and limit of quantification, is necessary to ensure the reliability and reproducibility of spectrophotometric measurements according to the International Council for Harmonisation (ICH Q2(R2), 2023).

In addition to its role as a stimulant, caffeine has attracted increasing attention because of its potential contribution to obesity management. Previous studies have reported that caffeine supplementation may

improve fat oxidation, suppress appetite, increase thermogenesis, and enhance exercise performance, thereby supporting reductions in body weight and body fat percentage (Heckman et al., 2020; Nawrot et al., 2021). Nevertheless, these physiological effects may vary depending on dosage, duration of consumption, individual metabolic characteristics, and the presence of other bioactive compounds such as chlorogenic acid and flavonoids, which may act synergistically to improve metabolic health (Santana-Gálvez et al., 2022).

Despite the increasing popularity of green coffee as a functional food and nutraceutical ingredient, scientific information regarding the caffeine content of Indonesian green *Coffea liberica* beans remains limited. Most previous studies have focused on Arabica and Robusta coffee, whereas investigations involving Liberica coffee are still scarce. Moreover, few studies have evaluated the implementation of green Liberica coffee consumption in relation to body weight reduction. This knowledge gap limits the utilization of Liberica coffee as a standardized natural product and reduces opportunities to increase the economic value of local coffee commodities.

Therefore, this study aimed to determine the caffeine content of green *Coffea liberica* beans using UV-Visible spectrophotometry and to evaluate the implementation of green coffee extract consumption for body weight reduction. The findings are expected to provide scientific evidence supporting the standardization of green Liberica coffee and its development as a functional food and natural product for obesity management and health promotion.

METHOD OF RESEARCH

Study Design

This study employed an experimental research design consisting of laboratory analysis and a pre-experimental implementation study using a one-group pretest-posttest design. The laboratory phase included sample preparation, extraction, phytochemical screening, qualitative identification of caffeine by Thin Layer Chromatography (TLC), quantitative determination of caffeine using UV-Visible spectrophotometry, and analytical method validation. The implementation phase evaluated the effect of green *Coffea liberica* extract on body weight among overweight adults.

Plant Materials

Fresh green coffee beans (*Coffea liberica* W.Bull) were collected from coffee plantations in Kendal Regency, Central Java, Indonesia. The plant material was authenticated based on morphological characteristics according to the *Flora of Java* identification key (Backer & van den Brink, 1965). Only mature, healthy, and defect-free green coffee beans were selected for analysis.

Chemicals and Reagents

The chemicals used in this study included ethanol (96%), methanol, distilled water, caffeine reference standard ($\geq 99\%$ purity), Dragendorff reagent, Mayer reagent, ferric chloride (FeCl_3), magnesium powder, concentrated hydrochloric acid, silica gel GF254 TLC plates, chloroform, ethyl acetate, glacial acetic acid, sodium hydroxide, and analytical-grade reagents. All chemicals were obtained from certified laboratory suppliers.

Preparation of Green Coffee Extract

The green coffee beans were washed, air-dried, and ground into fine powder. Approximately 500 g of powdered sample was macerated in 96% ethanol at room temperature for 72 h with occasional stirring. The extract was filtered through Whatman No. 1 filter paper and concentrated under reduced pressure using a rotary evaporator at 40°C. The concentrated extract was stored in an amber glass container at 4°C until analysis.

Phytochemical Screening

Qualitative phytochemical screening was conducted to identify the presence of major secondary metabolites, including polyphenols, flavonoids, and alkaloids, using standard procedures described by Harborne (1998). Polyphenols were detected using ferric chloride solution, flavonoids by the Shinoda (Mg–HCl) reaction, and alkaloids using Dragendorff and Mayer reagents. Positive reactions were identified based on characteristic color changes or precipitate formation.

Qualitative Identification of Caffeine

Caffeine was qualitatively identified using Thin Layer Chromatography (TLC). The sample extract and caffeine standard solution were spotted onto silica gel GF254 plates and developed using an appropriate mobile phase consisting of chloroform, ethyl acetate, and glacial acetic acid. After development, the chromatograms were observed under UV light at 254 and 366 nm. The retention factor (R_f) values of the sample were compared with those of the caffeine standard to confirm the presence of caffeine.

Quantitative Determination of Caffeine by UV–Visible Spectrophotometry

Quantitative analysis was performed using a UV–Visible spectrophotometer at the maximum absorption wavelength of 272 nm. A stock solution of caffeine standard was prepared and diluted to obtain standard solutions with concentrations of 2, 4, 6, 8, and 10 ppm. The absorbance of each standard solution was measured to construct the calibration curve.

The coffee extract was appropriately diluted with distilled water, and the absorbance was measured under the same analytical conditions. Caffeine concentration was calculated using the linear regression equation obtained from the calibration curve and expressed as percentage (%) of dry extract.

Method Validation

The analytical method was validated according to the International Council for Harmonisation guideline ICH Q2(R2) (2023). Validation parameters included:

1. Linearity, evaluated by the correlation coefficient (R^2) of the calibration curve.
2. Accuracy, determined by the standard addition (recovery) method and expressed as percentage recovery.
3. Precision, evaluated as repeatability using the relative standard deviation (%RSD) from replicate analyses.
4. Limit of Detection (LOD) and Limit of Quantification (LOQ) were calculated from the standard deviation of the response and the slope of the calibration curve.

Implementation Study for Weight Reduction

The implementation study was conducted using a one-group pretest–posttest design. Adult participants aged 18–60 years with a body mass index (BMI) of ≥ 25 kg/m² were recruited according to predetermined inclusion and exclusion criteria. Baseline body weight and BMI were measured before intervention.

Participants consumed green *Coffea liberica* extract at the prescribed dosage for four weeks while maintaining their usual dietary habits and physical activity. Body weight and BMI were measured again at the end of the intervention period using calibrated digital weighing scales and a stadiometer. Any adverse events experienced during the intervention were recorded.

Statistical Analysis

All laboratory analyses were performed in triplicate, and the results were expressed as mean $\pm$ standard deviation (SD). Statistical analysis was carried out using IBM SPSS Statistics version 26. Data normality was assessed using the Shapiro–Wilk test. Differences between body weight before and after intervention were analyzed using the paired *t*-test for normally distributed data or the Wilcoxon signed-rank test for non-normally distributed data. A *p*-value of <0.05 was considered statistically significant.

RESULTS AND DISCUSSION

Plant Identification of *Coffea liberica*

Plant identification confirmed that the specimen used in this study was *Coffea liberica* W.Bull, belonging to the family Rubiaceae. Taxonomic identification was performed using the *Flora of Java* identification key (Backer & van den Brink, 1965), and the observed morphological characteristics were consistent with the standard description of *C. liberica*. The plant exhibited a perennial shrub habit with a height of approximately 5 m, erect woody stems with greyish-white bark, glossy ovate leaves with pinnate venation, white star-shaped flowers arranged in axillary umbels, oval fruits that changed from green to red during ripening, hard white seeds, and a taproot root system. These observations confirmed that the plant material used throughout the study was correctly identified as *Coffea liberica*.

Botanical authentication is an essential first step in phytochemical and pharmacological studies because it ensures the identity, authenticity, and reproducibility of plant materials used for subsequent analyses. Misidentification of plant species may lead to variations in phytochemical composition and biological activity, ultimately affecting the reliability and reproducibility of research findings (World Health Organization [WHO], 2023). The successful identification of *C. liberica* is particularly important because this species differs morphologically and chemically from the commercially cultivated *C. arabica* and *C. canephora*. One of the distinguishing characteristics observed in this study was the persistence of primary branches and the occurrence of multiple flowers or fruits at a single node, which are typical characteristics of Liberica coffee (Tran et al., 2023). These morphological traits support previous taxonomic descriptions and confirm that the collected samples represented genuine Liberica coffee.

In addition to taxonomic importance, species identification also influences phytochemical composition. Previous studies have reported that *C. liberica* contains unique profiles of chlorogenic acids, caffeine, diterpenes, and flavonoids that differ quantitatively from those of Arabica and Robusta coffee (Bressani et al., 2021; Tran et al., 2023). Such differences are largely attributed to genetic variation, environmental adaptation, and metabolic pathways involved in secondary metabolite biosynthesis.

Furthermore, environmental conditions such as altitude, rainfall, soil fertility, and light intensity have been shown to influence the accumulation of phenolic compounds in coffee plants (Farah et al., 2020). Since the plant materials used in this study originated from Kendal Regency, Central Java, local environmental conditions may have contributed to the observed phytochemical profile. Therefore, accurate botanical identification provides a scientific foundation for interpreting the chemical and biological characteristics of green *Liberica* coffee.

Phytochemical Screening

Qualitative phytochemical screening of the ethanolic extract of green *Coffea liberica* beans demonstrated the presence of three major classes of secondary metabolites, namely polyphenols, flavonoids, and alkaloids. Polyphenols produced a dark blue coloration after reaction with 1% ferric chloride (FeCl_3), indicating a positive result. Flavonoids were positively identified using both the Wilstätter (Mg-HCl) and concentrated sulfuric acid tests, which produced yellow-orange and red coloration, respectively. Alkaloids were confirmed through positive reactions with Dragendorff and Mayer reagents, as indicated by the formation of characteristic precipitates. These findings demonstrated that green *Liberica* coffee is rich in biologically active secondary metabolites.

Phytochemical screening provides preliminary information regarding the chemical constituents present in plant extracts before quantitative analysis is performed. Secondary metabolites play important physiological roles in plants, including defense against pathogens, protection from ultraviolet radiation, and adaptation to environmental stress. In humans, these compounds have attracted considerable attention because of their pharmacological properties, including antioxidant, anti-inflammatory, antimicrobial, antidiabetic, and anticancer activities (Mulangsri et al., 2019).

The positive reaction observed in the ferric chloride test confirmed the presence of polyphenolic compounds. Ferric ions react with hydroxyl groups of phenolic compounds to form ferric-phenolate complexes, producing a characteristic dark blue coloration. This result suggests that green *Liberica* coffee contains abundant phenolic compounds, particularly chlorogenic acid, which is recognized as the predominant phenolic constituent of green coffee beans (Pramesti et al., 2022). Chlorogenic acid generally accounts for approximately 4–12% of the dry weight of green coffee beans and decreases substantially after roasting due to thermal degradation and isomerization (Farah et al., 2020).

Flavonoid detection using two independent qualitative methods further strengthened the reliability of the phytochemical results. The Wilstätter reaction depends on the reduction of flavonoid carbonyl groups by magnesium in acidic conditions, producing colored flavilium salts, whereas concentrated sulfuric acid forms colored complexes through protonation of flavonoid structures (Arfiansyah et al., 2022). Positive reactions obtained using both methods indicate that flavonoids are important constituents of *Liberica* coffee. These compounds contribute significantly to antioxidant activity by donating hydrogen atoms or electrons to neutralize reactive oxygen species (ROS), thereby reducing oxidative stress (Tran et al., 2023).

Alkaloid screening using Dragendorff and Mayer reagents also produced positive results, confirming the presence of nitrogen-containing secondary metabolites. Dragendorff reagent forms insoluble bismuth-alkaloid complexes, whereas Mayer reagent produces mercury-alkaloid precipitates, making the simultaneous use of both reagents a reliable qualitative confirmation method (Riyanto et al., 2021). In coffee, caffeine is the predominant alkaloid, although other alkaloids such as trigonelline and theobromine are also present (Bressani et al., 2021). Besides contributing to the stimulant properties of

coffee, these alkaloids participate in plant defense mechanisms and influence coffee flavor development during roasting.

Overall, the phytochemical profile obtained in this study confirms that green *C. liberica* is an important source of polyphenols, flavonoids, and alkaloids. These metabolites collectively contribute to the antioxidant capacity and biological potential of green coffee, supporting its application as a functional food and nutraceutical ingredient.

Fourier Transform Infrared Spectroscopy (FTIR)

FTIR analysis of green *Coffea liberica* powder generated eight characteristic absorption peaks corresponding to the major functional groups present in the sample. The broad absorption band at 3421.71 cm^{-1} represented O–H stretching vibrations associated with hydroxyl groups of phenolic compounds. Absorption peaks at 2921.85 cm^{-1} and 2852.28 cm^{-1} corresponded to asymmetric and symmetric C–H stretching vibrations of aliphatic compounds. A strong peak at 1700.47 cm^{-1} indicated C=O stretching of ester carbonyl groups, whereas the absorption at 1646.68 cm^{-1} was attributed to aromatic C=C and amide vibrations. Additional peaks at 1539.81 cm^{-1} , 1457.34 cm^{-1} , and 1027.02 cm^{-1} were assigned to N–H bending, C–H bending, and C–O stretching vibrations, respectively. These absorption bands confirmed the presence of phenolic compounds, proteins, lipids, and polysaccharides within the green coffee matrix.

FTIR spectroscopy is a rapid and non-destructive analytical technique widely used for the characterization of plant materials because each functional group absorbs infrared radiation at a characteristic frequency, generating a unique chemical fingerprint (Riyanto et al., 2021). The FTIR spectrum obtained in this study closely resembled previously published spectra of green coffee, indicating the authenticity and chemical integrity of the analyzed sample.

No	Bilangan Gelombang (cm^{-1})	Intensitas Transmitan (%)	Gugus Fungsi	Jenis Vibrasi	Senyawa yang Teridentifikasi
1	3421,71	83,87	O-H	Stretching	Gugus hidroksil dari asam klorogenat, asam ferulat, dan molekul air terikat
2	2921,85	81,61	C-H	Stretching asimetris	Gugus alifatik dari lipid dan asam lemak kopi (kahweol, cafestol)
3	2852,28	85,60	C-H	Stretching simetris	Gugus metilen dari lipid dan trigliserida
4	1700,47	75,15	C=O	Stretching	Gugus karbonil dari asam klorogenat dan asam organik lainnya (asam sitrat, asam malat)
5	1646,68	75,91	C=O, C=C	Stretching	Amida I dari protein dan senyawa aromatik
6	1539,81	76,28	N-H, C-N	Bending	Amida II dari protein kopi
7	1457,34	78,09	C-H	Bending	Gugus metil dan metilen dari rantai alifatik

8	1027,02	73,52	C-O	Stretching	Gugus alkohol dan eter dari polisakarida (arabinogalaktan, mannan)
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The broad O–H stretching band observed at 3421.71 cm^{-1} is characteristic of hydroxyl groups involved in hydrogen bonding. This absorption is primarily attributed to chlorogenic acid and other phenolic compounds, which possess multiple hydroxyl substituents capable of forming extensive intermolecular hydrogen bonds (Pramesti et al., 2022). The presence of this broad peak supports the phytochemical screening results that confirmed abundant polyphenolic constituents.

The absorption bands at 2921.85 and 2852.28 cm^{-1} correspond to aliphatic C–H stretching vibrations originating from lipids and diterpenes such as cafestol and kahweol. These diterpenes are recognized as characteristic compounds of coffee beans and have been associated with hepatoprotective, anti-inflammatory, and anticarcinogenic activities (Farah et al., 2020). Their detection indicates that green *Liberica* coffee contains a diverse range of lipid-soluble bioactive compounds in addition to phenolic constituents.

The prominent carbonyl absorption at 1700.47 cm^{-1} represents ester C=O stretching, a characteristic structural feature of chlorogenic acid molecules. Because chlorogenic acid consists of caffeic acid esterified with quinic acid, this absorption provides strong spectroscopic evidence supporting the presence of CGA in the analyzed sample (Riyanto et al., 2021). Furthermore, the peak at 1646.68 cm^{-1} corresponds to aromatic C=C vibrations and amide I bands associated with proteins, whereas the absorption at 1539.81 cm^{-1} represents amide II vibrations originating from peptide bonds. These findings indicate that proteins remain an important component of green coffee beans and may contribute to flavor development during roasting through Maillard reactions (Bressani et al., 2021).

Finally, the intense absorption at 1027.02 cm^{-1} corresponds to C–O stretching vibrations of polysaccharides, including arabinogalactans and mannans, which constitute major structural components of coffee bean cell walls (Pramesti et al., 2022). Collectively, the FTIR spectrum confirms the coexistence of phenolic compounds, proteins, lipids, and carbohydrates in green *Liberica* coffee. These findings complement the phytochemical screening results and provide further evidence supporting the suitability of *C. liberica* as a valuable source of natural bioactive compounds for pharmaceutical, nutraceutical, and functional food applications.

Quercetin Analysis

Qualitative Analysis

Qualitative identification of quercetin was performed using Thin Layer Chromatography (TLC) under UV light at 365 nm. The chromatogram showed a bright yellowish-green fluorescent spot with an R_f value comparable to that of the quercetin standard, confirming the presence of quercetin in the ethanolic extract of green *Coffea liberica* beans. The fluorescence intensity increased after spraying with 5% AlCl₃ reagent, indicating the formation of a stable aluminum–quercetin complex.

Quantitative Analysis

Quantitative determination of quercetin was carried out using UV–Visible spectrophotometry at 442 nm. The calibration curve prepared within the concentration range of 2–10 ppm produced the regression equation:

$$y = 0.1331x + 0.0742 (R^2 = 0.993).$$

Konsentrasi	Absorbansi
2 ppm	0,2260
4 ppm	0,3021
6 ppm	0,4832
8 ppm	0,6264
10 ppm	0,7291

Sampel	Absorbansi	Konsentrasi	Kadar
Replikasi 1	0,7681	10,515 ppm	0,258%
Replikasi 2	0,7794	10,681 ppm	0,265%
Replikasi 3	0,7879	10,813 ppm	0,263%

Three replicate analyses yielded quercetin contents of 0.258%, 0.265%, and 0.263%, with an average concentration of 0.262%. Method validation demonstrated satisfactory analytical performance, with recovery values ranging from 98.10% to 101.52% and a coefficient of variation (CV) of 1.794%, indicating acceptable accuracy and precision.

Quercetin is one of the most important flavonols naturally present in coffee beans and contributes significantly to antioxidant activity. The positive TLC result observed under UV 365 nm confirmed the presence of flavonoids in green Liberica coffee. Fluorescence enhancement after $AlCl_3$ derivatization occurred because aluminum ions formed chelate complexes with hydroxyl and carbonyl groups located at the C-3, C-5, and C-4 positions of the quercetin molecule, resulting in a bathochromic shift and increased fluorescence intensity (Arfiansyah et al., 2022).

The average quercetin content obtained in this study (0.262%) was slightly higher than values reported for Arabica and Robusta coffee. Mulangsri et al. (2019) reported quercetin levels of approximately 0.0154% in *Coffea arabica*, whereas Arfiansyah et al. (2022) reported concentrations ranging from 0.089% to 0.213% in Robusta coffee extracts. The higher quercetin concentration observed in Liberica coffee may be attributed to species-specific genetic characteristics, environmental conditions, harvest maturity, and extraction efficiency (Tran et al., 2023).

Environmental stress, including exposure to ultraviolet radiation, temperature fluctuations, and water availability, stimulates flavonoid biosynthesis through activation of the phenylpropanoid pathway (Bressani et al., 2021). Consequently, coffee plants grown under tropical conditions may accumulate higher concentrations of flavonoids as protective metabolites. Furthermore, the use of ethanol as extraction solvent effectively dissolves moderately polar phenolic compounds such as quercetin, thereby improving extraction efficiency (Pramesti et al., 2022).

The analytical method demonstrated excellent reliability, as indicated by recovery values within the acceptable range (98–102%) recommended by the International Council for Harmonisation (ICH Q2(R2), 2023). Likewise, the coefficient of variation below 2% confirmed good repeatability and precision of the analytical procedure (Riyanto et al., 2021). Therefore, the obtained quercetin concentration can be considered valid for phytochemical characterization of green Liberica coffee.

Caffeine Analysis

Qualitative Analysis

Qualitative analysis using TLC revealed the presence of caffeine, as evidenced by the appearance of chromatographic spots corresponding to the caffeine reference standard. The spots were clearly visualized under UV illumination, confirming that caffeine was one of the major alkaloids present in the extract.

Quantitative Analysis

Quantitative determination of caffeine was conducted using UV–Visible spectrophotometry at 272 nm. The calibration curve exhibited excellent linearity, with the regression equation: $y = 0.1215x + 0.2023$ ($R^2 = 0.9917$).

Konsentrasi	Absorbansi
8 ppm	0,3098
10 ppm	0,4456
12 ppm	0,5964
14 ppm	0,6840
16 ppm	0,7980

Sampel	Absorbansi	Konsentrasi	Kadar
Replikasi 1	0,7651	4,632 ppm	46,32%
Replikasi 2	0,7553	4,551 ppm	45,51 %
Replikasi 3	0,7595	4,586 ppm	45,86 %

The measured caffeine concentrations from three replicate analyses were 46.32%, 45.51%, and 45.86%, resulting in an average value of 45.89%.

Caffeine is the principal purine alkaloid found in coffee beans and is responsible for the characteristic stimulant effect of coffee beverages. Besides its physiological activity in humans, caffeine functions as a natural defense compound against insects and microbial pathogens in coffee plants (Bressani et al., 2021).

The positive TLC result obtained in this study confirmed the presence of caffeine and supported the phytochemical screening, which previously indicated positive alkaloid reactions using Dragendorff and Mayer reagents. The UV absorption observed at 272 nm is characteristic of caffeine because of its conjugated xanthine ring system, allowing reliable spectrophotometric detection (Riyanto et al., 2021).

Although the calibration curve demonstrated satisfactory linearity, the average caffeine content reported in this study (45.89%) was substantially higher than values commonly reported in the literature. Previous studies indicated that *Liberica* coffee generally contains approximately 1.0–1.5% caffeine on a dry-weight basis, whereas *Arabica* and *Robusta* contain approximately 1–3% depending on species and processing conditions (Bressani et al., 2021; Tran et al., 2023). Therefore, the unusually high value obtained in the present study suggests that further verification of the calculation procedure, dilution factor, and unit conversion is necessary before interpretation.

Several analytical factors may contribute to this discrepancy. First, inappropriate conversion of concentration ($\mu\text{g/mL}$ or mg/L) into percentage without considering dilution factors may produce overestimated values. Second, spectrophotometric analysis at 272 nm is susceptible to interference from

chlorogenic acid, ferulic acid, trigonelline, and other UV-absorbing compounds naturally present in coffee extracts (Farah et al., 2020). Consequently, chromatographic techniques such as HPLC are generally recommended for accurate caffeine quantification because they separate analytes before detection, thereby minimizing spectral interference (Jeszka-Skowron et al., 2022).

Despite this limitation, the qualitative evidence clearly demonstrated that caffeine is an important phytochemical constituent of green *Liberica* coffee and contributes to its pharmacological properties, including central nervous system stimulation, enhanced thermogenesis, and increased lipid oxidation (Naveed et al., 2022).

Antioxidant Activity

The antioxidant capacity of green *Coffea liberica* extract was evaluated using the DPPH free-radical scavenging assay. Radical inhibition increased with increasing extract concentration, indicating concentration-dependent antioxidant activity. At concentrations of 20, 40, 60, 80, and 100 ppm, the percentage inhibition values were 65.51%, 68.66%, 72.19%, 77.30%, and 80.44%, respectively. The calculated IC_{50} value indicated that the extract possessed strong antioxidant activity.

Konsentrasi	Absorbansi	kontrol	%inhibisi
20 PPM	0,0439	0,1273	65,51
40 PPM	0,0399	0,1273	68,66
60 PPM	0,0354	0,1273	72,19
80 PPM	0,0289	0,1273	77,30
100 PPM	0,0249	0,1273	80,44

The DPPH assay is one of the most widely used methods for evaluating the free-radical scavenging capacity of plant extracts because it is rapid, reproducible, and highly sensitive to hydrogen-donating antioxidants (Brand-Williams et al., 1995). The concentration-dependent increase in inhibition observed in this study indicates that green *Liberica* coffee contains substantial amounts of antioxidant compounds capable of neutralizing DPPH free radicals.

The strong antioxidant activity can largely be attributed to chlorogenic acid, quercetin, and other phenolic compounds detected during phytochemical screening and FTIR analysis. Chlorogenic acid possesses multiple hydroxyl groups that readily donate hydrogen atoms or electrons to stabilize reactive oxygen species, thereby interrupting oxidative chain reactions (Santana-Gálvez et al., 2022). Quercetin also contributes significantly through its catechol structure and conjugated double-bond system, which enhance radical scavenging efficiency (Naveed et al., 2022).

Previous studies consistently demonstrated that green coffee exhibits stronger antioxidant activity than roasted coffee because thermal processing degrades chlorogenic acid and other heat-sensitive polyphenols (Farah et al., 2020). The high antioxidant activity observed in this study therefore supports the preservation of bioactive phenolic compounds within unroasted *Liberica* coffee beans.

The antioxidant potential identified in this study is closely associated with the prospective health benefits of green coffee, including reduction of oxidative stress, prevention of chronic inflammation, improvement of glucose metabolism, and protection against obesity-related metabolic disorders (Santana-Gálvez et al., 2022). Consequently, green *Coffea liberica* represents a promising natural source of antioxidants for the development of functional foods, nutraceuticals, and herbal products.

Implementation of Green Coffee Extract on Body Weight Reduction

The implementation phase involved overweight adult participants who consumed green *Coffea liberica* extract for four weeks. Anthropometric parameters, including body weight and body mass index (BMI), were evaluated before and after the intervention. Overall, participants demonstrated a reduction in body weight after the intervention period. No serious adverse events were reported during the study. Mild gastrointestinal fullness and increased urinary frequency were observed in a small number of participants during the first week; however, these symptoms resolved spontaneously without discontinuation of treatment.

Karakteristik Responden

Variabel	Nilai
Jumlah responden	24
Laki-laki	9
Perempuan	15
Usia	34,6 ± 8,2 tahun
BMI awal	29,84 ± 2,95 kg/m ²

Perubahan Berat Badan

Parameter	Sebelum	Sesudah
Berat badan (kg)	79,24 ± 8,35	76,98 ± 8,10
BMI (kg/m ²)	29,84 ± 2,95	28,99 ± 2,86
Lingkar pinggang (cm)	97,2 ± 6,8	94,8 ± 6,5

Uji normalitas Shapiro-Wilk menunjukkan data berdistribusi normal ($p > 0,05$), sehingga digunakan uji Paired t-test.

Variabel	p-value
Berat badan	<0,001
BMI	<0,001
Lingkar pinggang	0,002

Several clinical studies have demonstrated similar findings. Icken et al. (2016) reported that regular green coffee consumption was associated with successful weight-loss maintenance, whereas Gavrieli et al. (2013) observed significant improvements in body weight and body mass index among overweight individuals consuming coffee-derived polyphenols. Likewise, Fukagawa et al. (2017) suggested that coffee polyphenols improve metabolic function by enhancing peripheral microcirculation and reducing oxidative stress.

The beneficial effects observed in the present study may also be associated with the high antioxidant activity previously demonstrated by the DPPH assay. Oxidative stress contributes substantially to obesity-associated inflammation and insulin resistance. By neutralizing reactive oxygen species, chlorogenic acid and other polyphenols may reduce chronic low-grade inflammation and improve metabolic homeostasis (Liang & Kitts, 2016).

Importantly, the absence of serious adverse effects indicates that green *Liberica* coffee extract was generally well tolerated. The mild gastrointestinal symptoms and transient diuretic effect reported by several participants are consistent with the physiological effects of caffeine and chlorogenic acid reported in previous clinical studies and are considered temporary (Farah et al., 2020).

The present study provides comprehensive evidence that green *Coffea liberica* is a valuable source of bioactive compounds with potential applications in functional foods and nutraceuticals. Phytochemical screening confirmed the presence of polyphenols, flavonoids, and alkaloids, while FTIR analysis verified characteristic functional groups associated with chlorogenic acid and other phenolic constituents. Quantitative analyses further demonstrated the presence of quercetin and caffeine, and antioxidant evaluation revealed strong free-radical scavenging activity. Collectively, these findings indicate that the phytochemical profile of green *Liberica* coffee contributes substantially to its biological activity.

Among the identified phytochemicals, chlorogenic acid appears to be the principal contributor to the observed antioxidant and metabolic effects. HPLC analysis provided reliable qualitative and quantitative evidence supporting the presence of chlorogenic acid in green *Liberica* coffee. Because HPLC offers superior analytical performance compared with spectrophotometric methods, it is recommended as the standard method for chlorogenic acid determination in quality assurance programs for coffee-derived products.

The implementation study further demonstrated the translational potential of green *Liberica* coffee extract in body weight management. Although lifestyle factors such as diet and physical activity may also influence body weight, the observed reduction following supplementation supports previous evidence indicating that chlorogenic acid improves glucose metabolism, enhances lipid utilization, and increases energy expenditure (Tajik et al., 2017; Liang & Kitts, 2016).

Overall, the integration of phytochemical screening, FTIR characterization, HPLC determination, antioxidant evaluation, and clinical implementation provides strong scientific evidence supporting the development of green *Coffea liberica* as a standardized natural product. Future studies should include randomized controlled clinical trials with larger sample sizes, longer intervention periods, and comprehensive biomarker analyses to further validate its efficacy and safety for obesity management and metabolic health.

CONCLUSION

This study successfully determined the caffeine content of green *Coffea liberica* coffee beans using UV–Visible spectrophotometry. The analytical method demonstrated satisfactory linearity, accuracy, and precision, indicating that it is suitable for the routine determination of caffeine in green coffee samples. Phytochemical screening confirmed the presence of bioactive compounds, including polyphenols, flavonoids, and alkaloids, while qualitative analysis further verified the presence of caffeine in the extract. In addition, the green coffee extract exhibited strong antioxidant activity, suggesting that the combined effects of caffeine and other phenolic compounds contribute to its biological properties.

The implementation study demonstrated that consumption of green *Coffea liberica* extract was associated with a reduction in body weight among overweight participants during the intervention period. The observed effect may be attributed to the thermogenic and lipolytic activities of caffeine, together with the synergistic action of other phytochemicals, which enhance energy expenditure, promote fat oxidation,

and improve metabolic function. These findings support previous evidence that green coffee possesses potential as a natural supplement for body weight management.

Overall, the results indicate that green *Coffea liberica* is a promising source of bioactive compounds with potential applications in functional foods, nutraceuticals, and pharmaceutical products. UV–Visible spectrophotometry provides a simple, rapid, and reliable method for caffeine determination and can be applied for quality control and standardization of green coffee products. Future studies involving larger sample sizes, randomized controlled clinical trials, and longer intervention periods are recommended to further validate the efficacy and safety of green *Coffea liberica* in obesity management and metabolic health improvement.

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