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Comparative Study on Antioxidant and Antibacterial Activity of Coffee Grounds and Spent Coffee Grounds (Scg) Extract

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The most abundant waste in coffee beverage preparation and coffee production is spent coffee grounds (SCG) which are a rich source of bioactive compound including caffeine, chlorogenic acids and melanoidins. However, improper disposal of SCG in a landfill can contribute to the negative impact to environment such as soil damage, greenhouse gas emissions, loss of nutrients and clogging. SCG have a valuable source of nutrients and energy, but they are regularly disposed of before their full value is extracted. The present study aims to investigate the bioactive compounds, antioxidant and antibacterial activity between ~~a~~ fresh coffee grounds (CG) and SCG. The SCG extract (SCGE) and CG extract (CGE) were carried out using Soxhlet extraction method. Evaluation of bioactive compounds in CG and SCG were carried out through analyses of total phenolic content (TPC) and total flavonoid content (TFC) via UV-Vis Spectroscopy. Further analyses of an antioxidant and antibacterial activity of the extracts were studied using DPPH free radical scavenging assay and agar disc diffusion assay, respectively. The results showed that the extraction yields of the SCG and CG were 6.48 ± 0.40% and 8.13 ± 0.65%, respectively. It shows that the most water-soluble substances are extracted in the brewing process while substances with lower water solubility remain in SCG. The phenolic and flavonoid content of SCGE was slightly lower than CGE. The results from TPC of SCGE and CGE are 86.98 ± 0.27 mg QE/g and 115.71 ± 0.17 mg GAE/g, respectively. While the TFC of SCGE and CGE are 23.73 ± 0.03 mg QE/g and 27.21 ± 0.02 mg QE/g, respectively, due to the lower water solubility of the compound. The caffeine concentration of SCGE and CGE showed a similar pattern, with values of 6.59 ± 0.02 mg/g and 19.36 ± 0.01 mg/g, respectively, due to water-soluble properties of caffeine. SCGE and CGE have IC_{50} values of 11.34 and 19.73 for antioxidant activity, respectively. The findings demonstrate that SCGE and CGE have high in bioactive compounds, which contributes to their strong antioxidant activity. Subsequently, both SCGE and CGE inhibit growth against Escherichia coli, suggesting that both materials possess antibacterial properties. Finally, using SCG offers a new supply of valuable chemicals, encourages the development of sustainable practices for the coffee business, and increases the value of a readily available waste product.

Keywords: Spent coffee grounds (SCG); phytochemicals; antioxidant; antibacterial

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Coffee (Coffea sp) is a food product with highest global consumption and the second most valuable commodity after petroleum. Two types of coffee that are popular in the market are arabica coffee (*Coffea arabica*) and robusta coffee (*Coffea canephora*) while *C. liberica*, *C. stenophylla*, *C. racemosa,* and *C. mauritiana* are less popular species [1]. While the coffee is being processed until it produces the final roasted beans, several by-products are generated during the process such as the pulp, husk, parchment, mucilage, silver skin, and spent coffee grounds (SCG) [2]. Annually, over 10 million tonnes of coffee waste are produced worldwide per year by the coffee industry [3].

SCG are coffee beans that have been roasted and ground but have lost some of their water-soluble compounds [4]. Around 650 kg of SCG is produced by one ton of green coffee on average, and for every kilogram of soluble coffee produced about 2 kilograms of wet SCG [5]. Due to improper waste management, coffee wastes are usually dumped straight into the trash without treatment. As a result, it contaminated landfills and water bodies. Hence, reused SCG has drawn particular attention recently to encourage environmental pollution reduction and global sustainability. As environmental sustainability regarding SCG is becoming increasingly concerning, one of the five recommendations made by the Food and Agriculture Organization (FAO) of the United Nations for a sustainable, environmentally friendly, and circular bioeconomy is avoiding unrecycled biowaste [6].

Although SCG is typically considered as waste, it actually contains an abundance of bioactive

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compounds that may offer numerous benefits to health. SCG have a complex and diverse composition, including various classes of phytochemicals compounds. The exact composition might depend on some factors, including the type of coffee beans, brewing technique, and the duration and temperature of roasting. Some of the organic compounds found in SCG includes carbohydrates, proteins, lipids, as well as phenolic and bioactive compounds, such as chlorogenic acid, caffeine, caffeic acids, cafestol and kahweol [2]. Based on the findings on phytochemical screening conducted by Fadri *et. al* [7], arabica coffee beans extracts included several types of compounds, including steroids, flavonoids, alkaloids, tannins, and saponins.

The phenolic compounds present in SCG are from the polyphenol family of esters [8]. These compounds are classified as flavonoids, lignans, stilbenes, and phenolic acids based on their structures. One of the major compounds in SCG is chlorogenic acids (CGA) [9]. It has several potential health benefits affecting cardiovascular health, glucose metabolism and obesity [10-13]. Moreover, polyphenol contains flavonoids that are recognised for their antioxidant, ability to inhibit enzymes and other possible health benefits [14-16].

Another bioactive compound present in SCG is alkaloid, which include caffeine. Caffeine is the alkaloid major component in coffee fruits and responsible for the bitter taste of coffee. It is a stimulant contained in coffee and other beverages, it can change in mood, and provide temporary energy boosts [17]. The health benefits of phytochemicals present in SCG are correlated to their biological properties including antioxidant and antibacterial properties. The second most abundant alkaloid of coffee is trigonelline (N-methylnicotinic acid). Phenolic acids are divided into two types which are hydroxycinnamic acids (caffeic, chlorogenic, cinnamic, p-coumaric, ferulic, and synaptic acids) and hydroxybenzoic acids (gallic, hydroxybenzoic, ellagic, protocatechuic, syringic, and vanillic acids) [18].

Bioactive compounds have a wide range of biological properties in SCG such as antioxidant, anti-inflammatory, antidiabetic, antibacterial, and anticancer activities. Antioxidants present in most phytochemicals including phenolic, flavonoid, and saponin compounds. Antioxidants help to prevent cell damage and improve health by scavenging these free radicals [19]. Moreover, coffee extracts have been shown in previous research to exhibit antibacterial effects against several bacteria, such as *Staphylococcus aureus, Listeria monocytogenes*, and *Escherichia coli* [19,20]. It kills bacteria and inhibits their growth, which has the potential to be new antibacterial products or antibacterial drugs. The antibacterial activity of flavonoids has been reported and much research has been investigated to isolate and identify the structures of flavonoids possessing antibacterial, antifungal, and antiviral activity.

Therefore, the aim of this study is to measure the yield of SCGE and CGE using Soxhlet extraction method, to determine the phytochemicals of SCGE and CGE with total phenolic content (TPC), total flavonoid content (TFC) and caffeine content, also to investigate the antioxidant using DPPH radical scavenging assay and antibacterial activities against *Escherichia coli*. Through this study, the value of a readily available waste product can be boosted while promoting the development of sustainable uses for the coffee industry as well as other beneficial biological activities industries such as pharmaceutical and cosmetic industries.

EXPERIMENTAL

Chemicals and Materials

SCG and CG samples were collected from a local coffee shop in Arau, Perlis. The chemicals used in this study were provided by Science Laboratory Unit of UiTM Perlis. The chemicals include distilled water, 95% ethanol, methanol, Folin-Ciocalteu reagent, sodium carbonate (Na_2CO_3), sodium nitrite ($NaNO_2$), gallic acid, quercetin, sodium hydroxide (NaOH), aluminium chloride ($AlCl_3$), ascorbic acid, 2,2-diphenyl-2-picrylhydrazyl (DPPH), caffeine powder, chloroform, calcium carbonate ($CaCO_3$) Muller-Hinton (MHA) agar, Gram-negative bacteria (*Escherichia coli*) and gentamicin.

Samples Extraction

SCG and CG powder samples were dried in the oven for 8 hours at 80 °C. The dried SCG and CG samples were stored at room temperature in an airtight container to avoid moisture. Next, extraction of the CGE and SCGE was performed using Soxhlet extraction method. Firstly, 15 g of SCG sample was added in a thimble, along with 150 mL of 95% ethanol in round bottom flask. The temperature of heating mantle was set to 210 °C and the extraction time is 6 hours. After the extraction process completed, the ethanolic extract was filtered then allowed to evaporate at 60 °C in a rotary evaporator. The extract was weighed and the recovery yield was calculated. The procedure was repeated for the CG sample. The percentage yield of SCGE and CGE were calculated using Equation (1):

$$\% \text{ yield of sample extract} = \frac{\text{weight of extract (g)}}{\text{dry weight (g)}} \times 100\% \quad (1)$$

Bioactive Compounds Analyses

Determination of Total Phenolic Content (TPC)

The TPC of the SCGE and CGE were quantified using the Folin-Ciocalteu's reagent method by Diharmi *et. al* [21]. For preparation of the standard curve, a 1000 mg/L

of gallic acid stock solution was prepared by adding 10 mg of gallic acid into a 50 mL volumetric flask with solvent methanol. Then, five different concentrations were prepared by dissolving gallic acid stock solution in methanol. Absorbance was measured in standard caffeine solutions in the range of wavelength between 700-800 nm. The results of the scanned wavelength showed that the maximum absorbance for the samples was 765 nm. TPC is measured in milligrams of gallic acid equivalent per gramme of dried CG and SCG sample (mg GAE/g).

For sample preparation, 0.2 mL of 1000 mg/L of SCGE was added to 15.8 mL of distilled water with 1 mL of Folin-Ciocalteu reagent. After 8 minutes, 3 mL of 10% Na_2CO_3 solution was added to the mixture and shaken homogeneously. At room temperature, the mixture was left in the dark for one hour because some phenolic compounds are light sensitive. The procedure was repeated for the CGE sample. Measure the absorption of samples at 765 nm wavelength with UV-Vis spectrophotometer. The percentage of remained phenolic content in SCGE compared to CGE was calculated as shown in Equation (2):

$$\%\ \text{of remained phenolic content in SCGE}\ (\%) = \frac{\text{SCGE phenolic content}}{\text{CGE phenolic content}} \times 100 \quad (2)$$

Determination of Total Flavonoid Content (TFC)

The TFC of the samples were determined using method by Alam & Sharma [22] with modification in the volume of reagent used. For preparation of the standard curve, a 1000 mg/L of quercetin stock solution was prepared by adding 10 mg of quercetin into a 50 mL volumetric flask with solvent 95% ethanol. Then, 5 different concentrations were prepared by dissolving quercetin stock solution in 95% ethanol. A calibration curve was prepared from the different quercetin concentrations. The absorbance of the mixture was determined at 510 nm wavelength using the UV-Vis spectrophotometer. The TFC is expressed as milligrams of quercetin equivalents per gramme of dried CG and SCG samples (mg QEs/g).

For sample preparation, 0.5 mL of the SCG ethanolic extract (1000 mg/L) was added to 2 mL of distilled water. Then, 0.15 mL of 5% sodium nitrite solution ($NaNO_2$) was added and allowed it to stand for 5 minutes. Next, 0.15 mL of 10% aluminium chloride ($AlCl_3$) was added and let it stand for 6 minutes. Lastly, 1 mL of 1 M NaOH was added and shaken, then let it stand for 15 minutes. 5% of $NaNO_2$ was prepared by adding 5 g $NaNO_2$ to 95 mL volume of distilled water, 10% aluminium chloride ($AlCl_3$) was prepared by adding 10 g of $AlCl_3$ in a 90 mL volume of distilled water and 1 M NaOH was prepared by adding 4 g of NaOH pellets in 25 mL distilled water. The mixture was incubated for 15 minutes at room temperature. The procedure was repeated for the CGE sample. The absorbance of the mixture was determined at 510 nm using the UV-Vis spectrophotometer. The percentage of remained flavonoid content in SCGE compared to CGE was calculated using Equation (3):

$$\%\ \text{of remained flavonoid content in SCGE}\ (\%) = \frac{\text{SCGE flavonoid content}}{\text{CGE flavonoid content}} \times 100 \quad (3)$$

Determination of Caffeine Content

The caffeine content of samples was determined using to the method performed by Vuletic *et. al* [23]. For the standard calibration curve, a 200 mg/L stock solution of caffeine was prepared by adding 20 mg into 100 mL volumetric flask with chloroform as solvent. Five different concentrations were prepared by diluting the stock solution. Absorbance was measured from standard caffeine solutions in the wavelength range between 250-300 nm. The maximum absorbance for the samples was found to be 274 nm. This wavelength was applied then to observe the caffeine calibration curve and caffeine content of SCG and CG.

For sample preparation, 2 g of SCG and CG samples were weighed precisely. Then, 20 mL of distilled water was added and heated for 10 mins until it boiled. A 2 g of Na_2CO_3 was added to each sample for tannins precipitation. Samples were filtered and the filtrates were heated to a 5 mL concentration. The caffeine was extracted by adding 5 mL of chloroform into the separatory funnel. The separatory funnel was shaken for a several minutes. The bottom layer containing caffeine was separated and analyzed with a UV/Vis spectrophotometer. A 0.1 mL of each SCGE and CGE was added into 10 mL of chloroform and placed in a quartz cuvette with wavelength scanned before (274 nm). The dilution factor of samples into chloroform are 100. The caffeine content in the samples can be calculated using Equation (4) [24]:

$$\text{Caffeine content}(\text{mg/g}) = \frac{(\text{M})(\text{V})(\text{DF})}{\text{m}} \times 100 \quad (4)$$

Where: M = sample concentration (mg/L), V= sample volume (L), DF = Dilution factor, and m = sample weight (g).

The percentage of remained caffeine content in SCGE compared to CGE was calculated as shown in Equation (5).

$$\%\ \text{of remained caffeine content in SCGE}\ (\%) = \frac{\text{SCGE caffeine content}}{\text{CGE caffeine content}} \times 100 \quad (5)$$

Antioxidant Activity Analysis

The antioxidant activity of the CG and SCG was determined using DPPH free radical scavenging assay according to the method obtained from Nerdy & Manurung [25] with modification in DPPH-sample volume ratio. For negative control, 40 mg/L of DPPH stock solution was prepared by adding 2 mg of DPPH into 50 mL volumetric flask, diluted with methanol to the indicated line, and shaken homogeneously. The solution was left in dark environment for 30 minutes in order to prevent light from significantly oxidizing the DPPH radical and interfering with the results. The absorbance at maximum wavelength was measured and obtained for further analysis. The results of the wavelength scanning showed that the maximum absorbance for the DPPH solution was 518 nm.

For sample preparation, 100 mg/L of SCGE and CGE was prepared by weighing 5 mg of SCG and CG then diluted with methanol in a 50 mL volumetric flask, the solution was shaken homogeneously. Five different concentrations (10, 20, 30, 40, and 50 mg/L) were prepared by diluting the sample extract stock solution in methanol. Then, 1 mL of the sample extract was combined with 3 mL of DPPH stock solution. The solution was left in dark environment for 30 minutes. The absorbance of the mixture was measured at 518 nm using the UV-Vis spectrophotometer. The scavenging activity of the DPPH radical can be calculated using the Equation (6):

$$\text{Scavenging activity (\%)} = \frac{(\text{Absorbance control} - \text{Absorbance sample})}{\text{Absorbance control}} \times 100 \quad (6)$$

The IC_{50} is referred as the concentration (mg/L) of samples needed to inhibit the production of DPPH radicals by 50% [26]. The IC_{50} value was determined through linear regression, where the x-axis displays the sample concentration and the y-axis indicates the % inhibition. From the equation y = mx + c, the IC_{50} value can be calculated using Equation (7) [27]:

$$\text{IC50} = \frac{50 - c}{m} \quad (7)$$

Where: y = % inhibition (50), c = Intercept, m = Slope, and x = Concentration of sample (mg/L).

Antibacterial Activity Analysis

The antibacterial competence of CGE and SCGE samples was evaluated against a common Gram-negative bacterium (Escherichia coli) through agar disc diffusion method. The zone of inhibition (clearance around the disc) was measured in order to determine the antibacterial activity of each concentration of CGE and SCGE (50, 25, and 12.5 mg/mL), compared with a positive control (gentamicin) and a negative control (distilled water). Briefly, Mueller-Hinton agar (MHA) agar medium was used for this study and the disc was placed on plate. Inoculate the bacteria on (with respective standard methods) sterilized, and placed disc on plates. Then dip about 12.5, 25, 50 mg/mL concentrations of CGE and SCGE with 6 mm diameter disc and incubate at 38°C for 24 hours. After incubation completed, the zone of inhibition was measured.

RESULTS AND DISCUSSION

The Percentage Yield of the Extracts

Soxhlet extraction was used to extract spent coffee grounds (SCG) and coffee grounds (CG). The percentage yield of both samples is shown in **Table 1**.

Based on **Table 1**, the percentage yield of SCG and CG extract is 6.48 ± 0.40 and 8.13 ± 0.65%, respectively. The data indicates that SCG produces lower yield compared to CG. Generally, SCG has been brewed into coffee beverages while the CG sample has not yet been brewed. This indicates that the sample extract has been lost as a result of the previous brewing process. In the coffee brewing process, the hot water extracts the water-soluble substances, leaving behind substances with lower water solubility in the SCG. However, the extract yield of SCG is still considerable in comparison to CG extract, which can be utilised in the food or non-food industry.

Table 1. Percentage yield of SCG and CG extracts.

Sample	Yield (%)
SCG	6.48 ± 0.40
CG	8.13 ± 0.65

Table 2. Total phenolic content (TPC) of SCG and CG extracts.

Sample	TPC (mg GAE/g)
SCG	86.98 ± 0.27
CG	115.71 ± 0.17

Bioactive Compounds Analyses

Determination of Total Phenolic Content (TPC)

The phenolic content of the SCG and CG extracts are reported in **Table 2**. The data are presented as mg of gallic acid equivalents (GAE) per gram of dry weight of the sample, using spectrophotometric measurement of the TPC. The levels of phenolic compounds found in SCG and CG is 86.98 ± 0.27 and 115.71 ± 0.17 mg GAE/g, respectively. SCG has slightly lower phenolic compounds compared to CG. The percentage of phenolic compounds remaining in SCG is 75.17%, which means that most of the phenolic compounds remain in the samples instead of being extracted during the brewing process. Abdeltaif *et al.* [15] also reported that phenolic content in SCG is less than raw CG.

According to Haminiuk *et al.* [28], polyphenols are frequently more soluble in organic solvents that are less polar than water. That means that phenolic compounds are highly soluble in organic solvents compared to water. Therefore, when coffee is brewed, barely any of the phenolic compounds was extracted into coffee beverages. Thus, it is shown that phenolic content in SCG is comparable with CG and holds potential as products with high phenolic content.

Determination of Total Flavonoid Content (TFC)

The total flavonoid content (TFC) of SCG and CG extracts are shown in **Table 3**. The TFC was determined using a spectrophotometer, and the results are expressed as mg of quercetin equivalents (QE) per gram of dry weight of the SCG and CG.

Flavonoids have been considered the most common, crucial, and easily available type of phenolics found in plants with a high antioxidant effect [15]. The flavonoid content in SCG and CG is 23.73 ± 0.03 and 27.21 ± 0.02 mg QE/g, respectively. The SCG sample showed that flavonoid content was slightly lower than CG. The percentage of remaining flavonoid content in SCG is 87.21% after being brewed. The percentage is high because flavonoid compounds are mostly not extracted during the brewing process.

As polar substances, flavonoids are less soluble in water and highly soluble in polar solvents like ethanol, methanol, butanol, acetone, dimethylformamide, and dimethyl sulfoxide [29,30]. Similar studies were reported by Abdeltaif *et al.* [15] that flavonoid content in SCG is lower than raw CG. As SCG has an abundance of flavonoid compounds as in CG, SCG has an opportunity as a product rich in flavonoids.

Table 3. Total flavonoid content (TFC) of SCG and CG extracts.

Sample	TFC (mg QE/g)
SCG	23.73 ± 0.03
CG	27.21 ± 0.02

Table 4. Caffeine content of SCG and CG extracts.

Sample	Caffeine concentration (mg/g)
SCG	6.59 ± 0.02
CG	19.36 ± 0.01

Determination of Caffeine Content

Caffeine content (mg/g) in the sample of SCG and CG are shown in **Table 4**. The results show that the caffeine content in SCG is 6.59 ± 0.24 mg/g, while CG produces 19.36 ± 0.04 mg/g of coffee content. The caffeine content in SCG is drastically lower than the caffeine content in CG, only 34.04 % of the caffeine content remained in the SCG extract, while another 65.96% of the caffeine was extracted during the brewing process. This is because caffeine is readily soluble in water [31]. Thus, most of the caffeine has been extracted during the brewing process as it uses a hot water temperature.

Antioxidant Activity Analysis

The absorbance value of the control obtained was 1.224. The scavenging activity and IC_{50} for both sample extracts were calculated. **Figure 1** displays the antioxidant activity in terms of DPPH scavenging activity of SCG and CG samples, while **Table 5** shows the IC_{50} value for SCG and CG extracts.

As shown in **Figure 1**, the scavenging activity of SCG extract ranged between 49.76 ± 0.13% to 56.57 ± 0.03% while CG ranged from 47.48 ± 0.47% to 59.25 ± 0.28%. Overall, the results indicated that as the concentration of SCG and CG increased, the scavenging activity increased. The results showed that the scavenging activity of CG and SCG at 30 mg/mL is slightly comparable. The slight difference in scavenging activity can be correlated with the level of phytochemicals present in the sample extract. The measured antioxidant activity is positively correlated with the levels of phenolic and flavonoid compounds in the SCG and CG extracts.

From the scavenging activity of different concentrations of SCG and CG, a graph was plotted. From the graph equation, the IC_{50} was calculated. The IC_{50} of SCG and CG was 11.34 and 19.73 mg/L, respectively. According to Gülçin and Alwasel [32], lower IC_{50} values indicate a higher DPPH free radical scavenging ability and higher antioxidant activity. The IC_{50} value of SCG was slightly different from CG, showing that both samples have great antioxidant capacity. Thus, SCG can be used in the production of antioxidant products such as food additives to extend shelf life and personal care to protect skin from free radical damage.

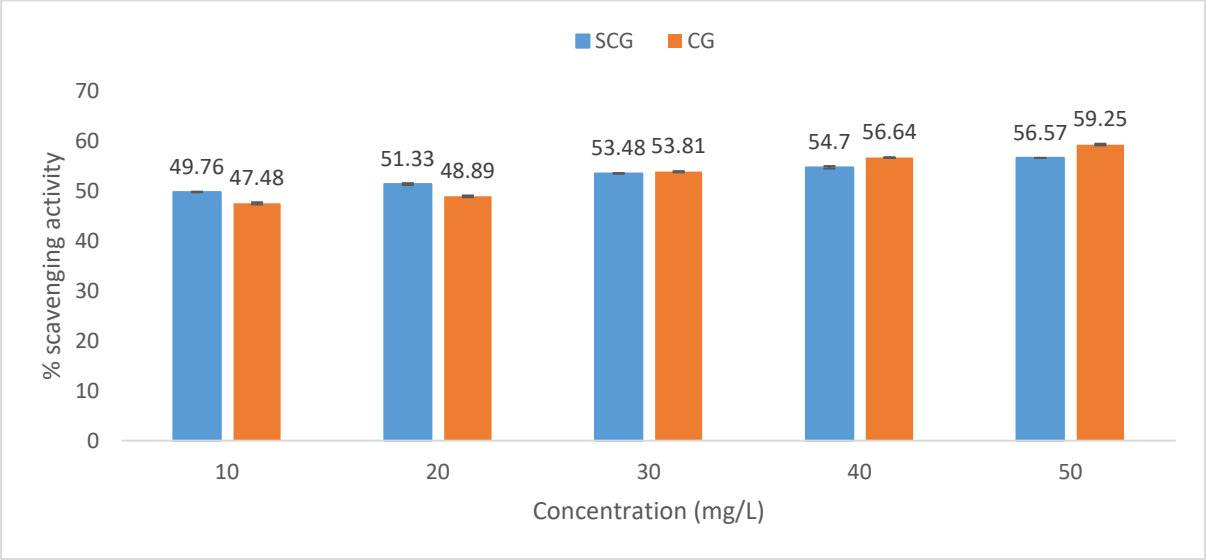
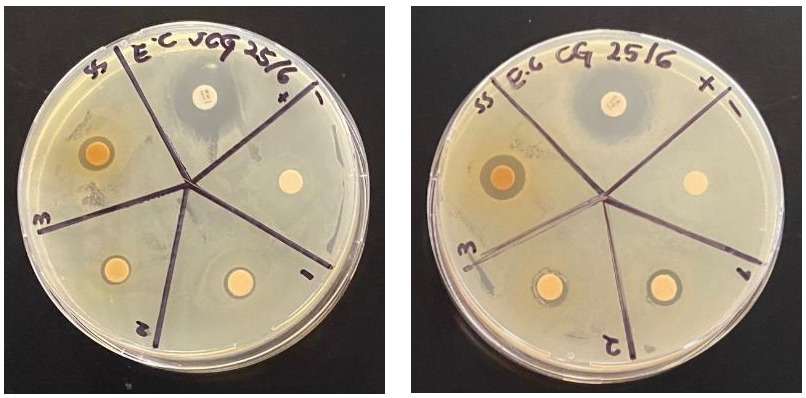


Figure 1. DPPH scavenging activity of SCG and CG extracts.

Table 5. IC_{50} of SCG and CG extracts.

Sample	IC_{50} (mg/L)
SCG	11.34
CG	19.73



(a) (b)

Figure 2. Antibacterial activity of (a) SCG and (b) CG against *Escherichia coli.*

Table 6. Inhibition zone of different concentration of SCGE and CGE with negative and positive control against *Escherichia coli*.

Sample	Concentration (mg/mL)	Inhibition Zone (mm)
SCG	50	9.73 ± 0.11
	25	8.63 ± 0.18
	12.5	8.40 ± 0.07
	Positive control	22.75 ± 0.21
	Negative control	-
CG	50	13.6 ± 0.14
	25	10.43 ± 1.11
	12.5	9.58 ± 0.25
	Positive control	22.1 ± 0.14
	Negative control	-

Antibacterial Activity Analysis

The clear zone formed around the paper disc revealed inhibitory activity against *Escherichia coli*, showing that the samples possess antibacterial properties as shown in **Figure 2**. The inhibition zone of positive control, negative control and each concentration of both extracts (12.5, 25, 50 mg/mL) was shown in **Table 6**. According to the data, positive control (gentamicin) has the largest inhibition zone as an antibiotic, while negative control (distilled water) does not form an inhibition zone because it does not possess antibacterial properties. Also, the data showed that the higher the concentration of SCG and CG extract (50 mg/mL), the larger the diameter of the inhibition zone which are 9.73 ± 0.11 mm and 13.6 ± 0.14 mm, respectively.

There is a correlation between the level of phytochemicals in the sample and the diameter of the inhibitory zone. The extract with higher levels of flavonoids and phenolics has more potent antibacterial activity [33]. Based on the result, the antibacterial activity against *Escherichia coli* correlates with the phenolic and flavonoid content of CG and SCG. Based on a study conducted by McConnell and Bakermans [34] stated that caffeine content and growth rate showed a strong negative correlated in all circumstances, indicating that caffeine may have antimicrobial properties when consumed. Previous research also

reported that SCG extract has antibacterial properties against various bacteria such as *Aeromonas hydrophila* and *Porphyromonas gingivalis* [35, 36]. Thus, there is potential for SCG extract to be developed as a natural food preservative or food packaging, as *Escherichia coli* is a common foodborne bacterium that contaminates foods.

CONCLUSION

The SCGE and CGE extract were successfully extracted by Soxhlet extraction method which produced percentage yield 6.48 ± 0.40 % and 8.13 ± 0.65 %, respectively. The phytochemicals in SCGE and CGE such as phenolic, flavonoid and caffeine were successfully accomplished by total phenolic content (TPC), total flavonoid content (TFC), and caffeine content determination utilising UV/Vis-spectrophotometer. The phenolic content of SCG and CG are 86.98 ± 0.27 mg GAE/g and 115.71 ± 0.17 mg GAE/g, respectively while TFC of SCG and CG are 23.73 ± 0.03 mg QE/g and 27.21 ± 0.02 mg QE/g, respectively. The results shown that SCG and CG have caffeine content at 6.59 ± 0.02 mg/g and 19.36 ± 0.01 mg/g, respectively. For antioxidant activity, SCG and CG were tested with DPPH radical scavenging assay employing UV/Vis-spectrophotometer. The percentage scavenging activity of both samples increased when the concentration of sample increases. The IC_{50} of SCG and CG were calculated and produced 11.34 and 19.73, respectively. Finally, both SCG and CG extracts possess antibacterial properties against bacteria *Escherichia coli.* Three different concentrations of SCG 50, 25, 12.5 mg/mL, have inhibition zone 9.73 ± 0.11, 8.63 ± 0.18, and 8.40 ± 0.07 mm, respectively while CG extract at 50, 25, 12.5 mg/mL produced an inhibition zone 13.6 ± 0.14, 10.43 ± 1.11 and 9.58 ± 0.25, respectively. An antibiotic, gentamicin was used as positive control and produced 22.75 ± 0.21 mm and 22.1 ± 0.14 mm. Overall, SCG has a significant amount of phenolic and flavonoid compounds compared to CG. In order to improve this research, the temperature for Soxhlet extraction should be changed to the solvent boiling point to ensure efficient extraction and avoid degradation of certain compounds and more assay and types of bacteria and fungi should be tested to provide different insights. This study has demonstrated that the phytochemicals in SCG possess antioxidant and antibacterial properties. There is great potential in employing the valuable compounds found in SCG in the food, cosmetic, pharmaceutical, and personal care industries. Thus, the utilization of SCG also helps to raise the value of a readily available waste product and reduce the amount of waste that ends up in landfills.

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