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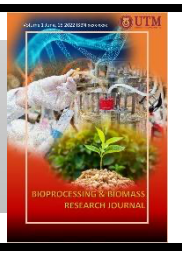

Research Article

Comparative Analysis of Phytochemical Composition in Ethanolic Extracts of *Acalypha Indica* Between Ultrasound-Assisted and Maceration Extraction Methods

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ABSTRACT

Acalypha indica (AI) is one of the edible traditional medicinal herbs used worldwide. Previously, conventional maceration extraction was often used to extract phytochemicals from AI plant material with limitations of prolonged extraction time and high solvent consumption. This study investigates the extraction methods, namely maceration extraction (ME) and ultrasound-assisted extraction (UAE), which were utilised to extract and compare the phytochemical content and antioxidant activity of the AI sample derived from the whole plant. The extraction involved the use of water solvent and ethanolic solvent (20%, 40%, 60%, and 80%). Each extract was subjected to chemical-assay guidance by testing for Total Phenolic Content (TPC), Total Flavonoid Content (TFC), and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay. The study reveals UAE extract gained a low EC_{50} (84.5 µg/ml) at 40% ethanolic solvent, the highest TPC (555.952 ± 41.938 mg GAE/g) using water solvent, and 60% obtained the highest TFC (887.9048 ± 46.085 mg RE/g). As for maceration, 60% obtained a low value of EC_{50} (115.1 µg/ml) and the highest TPC (567.061 ± 16.432 mg GAE/g). Meanwhile, 80% ethanol solvent obtained the highest TFC (577.619 ± 43.008 mg RE/g). Ultimately, maceration achieved higher polyphenolic content while having the better antioxidant outcome. However, the UAE extracted polyphenolic content in a shorter period of time and with lower solvent consumption.

INTRODUCTION

Acalypha indica (AI), commonly known as "Galak Kucing" or "Cika Emas" in Malaysia, is a type of weed that thrives in many residential areas and wet, tropical regions globally, making it indigenous to countries like India and South Africa, among others (Zahidin et al., 2017). As the name implies and notably recognises, AI plant contains a cat attractant compound that affects cats' behaviour with the sense of euphoria (Scaffidi et al., 2016; Wickramaratne et al., 2022). Beyond this unique characteristic, this plant's ethnopharmacological usage has long been etched in history. In Malaysia, this plant is mainly used as a purgative, medicine for worms, acne, and haemorrhoids. Countries such as India utilise this plant in many ways, like wound healing, burn injuries, coughs, laxatives, and many more (Ribeiro et al., 2010; Ibrahim et al., 2021). Intriguingly, some countries in the Arabian Gulf region consume this plant as an everyday food, and many south-east countries like Malaysia commercially marketed as a tea beverage (Ibrahim et al., 2021; Zahidin et al., 2017; Zahidin et al., 2018).

The growing fascination in the AI plant has led to extensive pharmacological research revealing its potential for various medicinal purposes, such as anti-inflammatory, antioxidant, hepatoprotective, and others (Nambiar &

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Varghese, 2023; Sharma et al., 2023). The AI plant extraction method has been studied using conventional and modern extraction. In order to extract phytochemicals, the traditional method of ME is soaking the plant sample in an appropriate solvent for a predetermined amount of time. On the other hand, UAE is a technologically advanced technique that uses ultrasonic waves to improve extraction performance and shorten the extraction time. However, there are limited studies on the usage of UAE on the AI plant. These techniques were selected because they are useful and efficient for extracting phytochemicals from plant sources.

A well-known method for extracting thermolabile components from plants is ME. It might not be the most effective technique in terms of polyphenolic yield and extraction time, despite its ease of use and adaptability (Carmen et al., 2022). Higher yields of polyphenolic compounds and time-effectiveness are amongst the benefits of using ultrasonic waves (Adeeyo et al., 2023). In addition, solvent choice plays a pivotal role in extraction due to consideration of the polarity factor. In exploring this, different polarity may exhibit different bioactive compounds.

This study compares both methods of extraction while investigating the use of ethanolic extraction solvent (20%, 40%, 60%, and 80%) and water solvent (0% ethanol solvent) on the ground sample extract of the AI plant. The extract was then utilised for further phytochemical profile research employing the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity assay for antioxidant capabilities, the total phenolic content (TPC) and total flavonoid content (TFC). In this study, 2 different extraction techniques, which are ultrasound-assisted and maceration extractions, were used. Each method was used to compare the phytochemicals that were recovered. The antioxidant and total polyphenolic content extracted from both methods could contribute to further extraction studies in targeting polyphenolic-rich extract and potential therapeutic value.

MATERIALS AND METHOD

Materials

Acalypha indica (AI) plant was procured from a local nursery in Sungai Buloh, Malaysia. The whole plant was harvested and air-dried under shade for 3 days and ground until it passed a 40-mesh sieve. For chemical-based assays, ethanol and methanol were acquired from Hmbg Chemicals. The analytical standards, such as gallic acid, rutin, and ascorbic acid, alongside sodium hydroxide (NaOH) and 2,2-diphenyl-1-picrylhydrazyl (DPPH), were procured from Sigma-Aldrich (Taufkirchen, Germany). Folin-Ciocalteu's phenol reagent was purchased from R&M Chemicals (Selangor, Malaysia). Dimethyl sulfoxide (DMSO) and sodium carbonate (Na_2CO_3) were obtained from QRec Asia (Selangor, Malaysia). Finally, aluminium chloride ($AlCl_3{\cdot}6H_2O$) and sodium nitrite ($NaNO_2$) were acquired from Bendosen via O&E Technologies (Terengganu, Malaysia).

Ultrasonic-Assisted Extraction (UAE)

The method from Idris et al. (2022) was utilised with minor adjustments. The AI plant was weighed and mixed into different ethanol-water mixtures (0, 20, 40, 60, and 80% ethanol) with a solvent-solid ratio of 10 ml/g with the lid covered with aluminium foil. The beaker was then placed into a 40 kHz bath sonicator (Chincan, China) with the water level above the mixture in the beaker. The sample was then extracted for 10 minutes at 30 °C. Next, the extract was filtered using gravity with a filter paper (Smith No. 101, 125 mm). Later, the filtrate was dried overnight in an oven dryer at 60 °C and stored at -20 °C for later use.

Maceration Extraction (ME)

The extraction was carried out using Ngibad (2019) with slight modification. The maceration method was performed with water solvent, 20%, 40%, 60%, and 80% ethanol solvent. The solvent-solid ratio was kept at 20 ml/g. The AI plant was weighed and mixed thoroughly with each solvent. An orbital shaker (Protech model 719) operating at 120 rpm at room temperature was used for the extraction process, which required continuous shaking for a whole day. After that, the mixture was filtered via filter paper (Smith No. 101, 125 mm) to produce pulp and filtrate. The filtrate was concentrated and completely dried in a drying oven at 60 °C and stored at -20 °C for later usage.

Total Phenolic Content (TPC)

The method used is modified from Ismail et al. (2017). Initially, 30 µl of 10 mg/ml dissolved extract was inserted in a 96-well plate in triplicate with a blank sample. Next, 60 µl of Folin-Ciocalteu's reagent (2N), 60 µl of 10% sodium carbonate (Na_2CO_3) and 150 µl of distilled water were added and incubated for 60 minutes at room temperature and in the dark. For the sample blank, Folin-Ciocalteu reagent was replaced with distilled water. A gallic acid standard curve was also prepared at different concentrations (0–500 µg/ml) to be used in the regression equation. The plate was then measured via a microplate reader (Biobase, China) at 750 nm. The results were represented as mg GAE/g.

Total Flavonoid Content (TFC)

Abd Rashid et al. (2022) method served as the basis for the approach used here, with some modification. Firstly, 30 µl of 10 mg/ml dissolved extract was inserted in a 96-well plate in triplicate with a blank. Next, 60 µl of 5% sodium nitrite ($NaNO_2$), 60 µl of 10% aluminium chloride hexahydrate ($AlCl_3{\cdot}6H_2O$) and 150 µl of sodium hydroxide (NaOH) were added and incubated for 15 minutes at room temperature and in the dark. For the sample blank, aluminium chloride hexahydrate was replaced with distilled water. The rutin standard curve was also prepared at different concentrations (0–500 µg/ml) to be used in the equation. The plate was then measured via a microplate reader (Biobase, China) at 430 nm. The results were represented as mg RE/g.

DPPH radical scavenging activity

The methodology employed is a modified version of that presented in Sarabo et al. (2021). For the treated sample, 150 µL of dissolved extract was diluted to different concentrations (0.001-1 mg/ml) and added with 150 µL of 0.5 mM methanol dissolved 2,2-diphenyl-1-picrylhydrazyl solution (DPPH) in triplicate in a 96-well plate. As for its blank, the DPPH solution was replaced with methanol. For the untreated sample, there are triplicates of 150 µL water and 150 µL DPPH solution with the sample blank of water and methanol. Ascorbic acid (AA) served as a positive control. Then, incubation was done for 30 minutes in the dark. The plate was then measured via a microplate reader (Biobase, China) at 515 nm. The EC_{50} value was generated

using non-linear regression of 4 parameters in variable format from GraphPad Prism v 9.0.

Statistical evaluation

Each triplicate measurement's average and standard deviation (SD) were calculated. One-way analysis of variance (ANOVA) and Tukey's multiple comparisons test were used to compare 3 or more independent groups. If the p-value is less than 0.05, it is considered statistically significant. Data analysis was done using Microsoft Office Excel and GraphPad Prism v. 9.0. The Pearson correlation was also included for determining the correlation between TPC, TFC, and EC_{50} of the extract obtained using maceration and UAE methods.

RESULTS AND DISCUSSION

Phytochemical screening

Extraction of phytochemicals in the AI plant was obtained using UAE and ME methods by varying ethanol concentration (0%, 20%, 40%, 60%, and 80% ethanol solvent). TPC and TFC assays were conducted for screening of phytochemical profiles present in each extract. **Figure 1** illustrates the TPC and TFC of each extract by both methods. The standard calibration curve for gallic acid ($Y = 0.0004893x + 0.08922$; $R^2 = 0.9982$) and rutin ($Y = 0.0006723x + 0.05899$; $R^2 = 0.9980$) demonstrated excellent linearity, confirming the reliability of quantification.

The quantitative assays confirmed that the AI plant is rich in phenolic compounds and flavonoids, as hypothesised. This phytochemical distribution is consistent with established data reported in several studies, including those from N'Paki et al. (2024), Sahukari et al. (2021) and Ravi et al. (2015). The UAE method obtained the highest TPC using aqueous solvent. Notably, a sharp decline was observed at 20% ethanol, followed by a steady decrease as ethanol concentration increased. In contrast, ME followed an upward trend, peaking at 60% ethanol solvent before declining at the 80% mark. As for TFC, UAE peaked at 60% ethanol before plummeting at the 80% mark. Conversely, the ME method showed a positive correlation with ethanol concentration with a maximum peak at 80%. Overall, UAE was most effective for TPC in water with TFC at 60%, while ME showed maximum TPC at 60% and TFC at 80%.

The extraction efficiency of polyphenols is highly dependent on solvent polarity, following the general principle of "like dissolves like". In this study, variation of TPC and TFC across different ranges of ethanol concentration reflects the diverse, unique chemical structures of polyphenols and polarity in the AI plant. Variations of ethanol content is widely used for the extraction of phenolics due to ethanol possessing greater affinities for phenolic compounds (Kumar et al., 2021). This can be seen from both the TPC and TFC profiles of either method. Notably, this observation was also observed and reported by Salma Iza et al. (2023). In changing ethanol concentrations, the amount of polyphenolic reported peaks at 70% ethanol concentration. Other than that, this study is similar to the previous report that found the highest extraction yield of phenolic and flavonoid compounds from soybean was obtained by 60% ethanol with 2.75 ± 0.13 mg GAE/g and 1.02 ± 0.04 mg CE/g, respectively, as compared to 70% and 80% ethanol (Teffane et al., 2022).

The polarity changes as the ethanol concentration increases. The same could be said when using different solvents of lesser polarity. The findings align with reports by Gantala et al. (2023) that use the ME method with 60% ethanol solvent and acetone on AI plant leaves. The less polar acetone extract obtained a TPC of 116.26 ± 2.84 equivalent to mM of tannic acid, and a TFC of 84.07 ± 1.44 equivalent to mM of quercetin. As for the hydro-ethanol extract, it shows a higher profile with a TPC of 148.57 ± 2.1 equivalent to mM of tannic acid, and a TFC of 63.86 ± 1.46 equivalent to mM of quercetin. The most common polyphenols are flavonoids and phenolics, which is the reason both phytochemicals were analysed in this study. The amount of TPC and TFC was affected by the extraction method and ethanol percentage from the observation of the results. 60-80% ethanol has been shown as the most favourable solvent for phenolic groups. This aligns with the characteristic of phenolic compounds that poorly dissolve in water due to the dissimilarity in polarity.

However, water solvent is also able to have high efficiency in extracting phenol. This is because the water content helps to weaken the hydrogen bonds connecting the phenols and the cell matrix. This facilitates the mass transfer of the compounds concurrently, enhancing the solubility of polyphenols (Mikucka et al., 2022). Conversely, another study using AI plant also reported that using Soxhlet extraction with aqueous solvent resulted in a higher TPC of 1016.73 ± 41.10 mg GAE/g but a lower flavonoid content of 24.33 ± 2.96 mg QE/g (Zahidin et al., 2018). This suggests that water solvent can be used to extract phenolic compounds like gallic acid and caffeic acid but may lack flavonoid content, although using different methods due to its polarity. While water is cheaper, more environmentally friendly, and safer, it can attract most polar compounds; polyphenols that are moderate and less polar might be more soluble in organic solvents (Lim et al., 2019). Hence, a combination of aqueous and organic solvents can aid further in extraction. Water can swell up the plant matrix and aid the diffusion of polyphenols throughout the plant matrix, while ethanol's low viscosity helps in enhancing mass transfer (Borges et al., 2020; Brglez Mojzer et al., 2016). Water as a solvent is characterised as a solvent with high polarity and demonstrates greater efficacy in extracting polar compounds like phenolic acids. Conversely, ethanol, a less polar solvent, is more adept at extracting flavonoids, which exhibit a lower degree of polarity.

Extraction method also plays a pivotal role in obtaining targeted compounds. Discovery of bioactive compounds is typically achieved through different extraction techniques. The techniques used in extracting the compounds should confer their chemistry and uneven distribution within the plant matrix. Both conventional maceration and modern UAE methods were used in this study. ME works through continuous shaking, which then breaks the surface tension (Nurcholis et al., 2022). Whereas the cell wall structure of plants is lysed through acoustic cavitation by microbubbles of the liquid phase in the UAE method (Debiasi et al., 2021). The working operation clearly states ultrasound potentially gives better TPC and TFC, which are considerably alike to the results obtained from this study at the several ethanol solvent extractions. However, at certain solvent concentrations, ultrasound-assisted extract recorded a lower yield than macerated extract, which may be due to decreased diffusion rates, reduced diffusion areas, and the elevation of diffusion distances, leading to a decrease in polyphenol levels due to prolonged extraction duration (Nurcholis et al., 2022).

As the phytocompound of chemistry varies, different extraction concentration solvents may also have different effects and impacts on the extraction yield, solubility, and activity of the antioxidants of the phytochemical. Extraction techniques and the type of solvents used play a significant role in determining the phytochemical components responsible for the biological activities of the propolis (Yıldırım, 2022). The amount of TPC and TFC obtained may be different depending on the chosen extracted plant, which is strongly related to polarity and solubility properties. Prominently, for both TPC and TFC for both methods, a decline can be observed at the 80% mark. This reduction can be attributed to the decreased swelling and diffusion throughout the plant matrix, leading to impeding the solubilising of polyphenolic content due to dehydration of the plant tissues and denaturation of protein, consequently decreasing the polyphenol content (Kumar et al., 2021). Other studies, such as Jacotet-Navarro et al. (2018), reported that rosmarinic acid, a significant contributor to TPC, has poor solubility in pure ethanol compared to 30% ethanol, which was optimal for its extraction, thereby causing a decrease in TPC at higher concentrations. A study by Venkatesan & Muniyan (2025) also reported Soxhlet extraction using pure ethanol as a solvent for AI plant leaves obtained a TPC of 28.24 mg GAE/g and a TFC of 23.40 mg QE/g. This indicates that the decrease in global extraction yield observed beyond 80% ethanol is accompanied by a decrease in the extraction of certain compounds, which contributes to the lower TPC at higher ethanol concentrations. To summarise, the UAE achieved higher TPC using aqueous solvent and TFC at the 60% mark. As for ME, ME peaks at 60% and TFC at 80%.

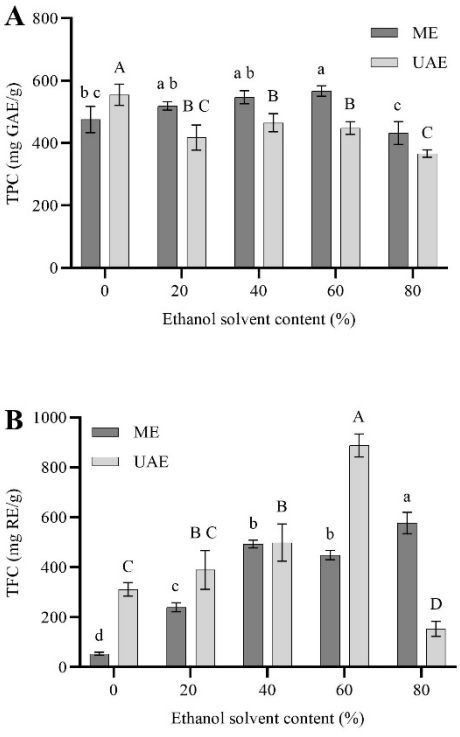

Figure 1 Phytochemical content of each AI extract. **A**) TPC for both methods. **B**) TFC for both methods. Both assays were performed in triplicate and represented as mean ± SD (n=3) with relative SD less than 10%. Different letters above the bars indicate statistically significant differences (p < 0.05), with small letters representing ME and capital letters representing UAE.

DPPH Radical Scavenging Activity Evaluation of Extracts

To evaluate the biological potential, DPPH radical scavenging activity was performed. This assay serves as a functional validation of the capability of each extract by both methods to nullify or scavenge free radicals. **Figure 2** depicts DPPH radical scavenging activity of each extract in a concentration-dependent manner. AA served as a positive control due to its strong antioxidant capacity. Much literature relates the polyphenolic content extracted with radical scavenging capability, linking AI plant with antioxidant capability, such as Sahukari et al. (2021), Kusrini et al. (2019) and Zahiddin et al. (2018). As demonstrated in **Figure 2**, all extracts follow a dose-dependent manner. AA outperformed all of the extract. **Table 1** below lists down all EC_{50} values of each extract by both methods with AA. The EC_{50} of AA recorded was 42.94 µg/ml, the lowest compared to all extracts by both methods, signifying a more prominent effect. As for extraction by the UAE, the lowest recorded was by 40% ethanol concentration with 84.5 µg/ml, while for ME, it was 115.1 µg/ml.

Table 1 EC_{50} values for DPPH RSA of each extract of each method of extraction

EtOH Conc (%)	EC_{50} (µg/ml)	
	Maceration	Ultrasonic-assisted
AA	42.94	
0	183.8	86.7
20	270.9	151.0
40	349.6	84.5
60	115.1	169.3
80	487.5	2228

This observation is similar to findings by Akshayaa et al. (2021), illustrated that increasing the concentration of different plant extracts of AI results in higher free radical scavenging activity. Another study reported by Salma Iza et al. (2023) recounted a low EC50 at aqueous solvent and 70% ethanol solvent with 61.975 µg/ml and 47.064 µg/ml, respectively, using the ME method. Venkatesan & Muniyan (2025) also reported that optimisation of Soxhlet extraction of AI plant leaves using ethanol concentration (60, 70, 80%) reveals higher DPPH radical scavenging activity using Box-Behnken design (BBD). Variations in the metabolites extracted might affect biological activities, including antioxidant function, due to antagonistic or synergistic effects on one another (Salma Iza et al., 2023).

The antioxidant activity of each extract was compared to AA. The antioxidant activity of each extract exhibits the same pattern as the standard AA, indicating the presence of phytochemicals comparable with antioxidant activities in AI plants. A previous study on PDB extraction indicated that 40% ethanol extract has the highest antioxidant activity (Venkatesan et al., 2019). The results indicate that the rosemary extracted with 40% ethanol had the lowest IC50 value (Ziani et al., 2022). In this study, the high discovery of phenolic and flavonoid compounds causes a high 55% of DPPH scavenging activity. Different solvent percentages along with the type of extraction used caused differences in the composition and antioxidant activities of the extract. This is because the solubility of antioxidant compounds in a

solvent depends on the characteristics of the phytochemicals present in the sample.

A water extract of kanuka leaves was concluded to contain major trace phenolic-based compounds, which include gallic acid, catechin trans-ferulic acid, syringic acid, 4-hydroxybenzoic acid, chlorogenic acid, 2-hydroxycinnamic acid, and quercetin (Majid & Silva, 2021). Most of which are small and more polar than flavonoid compounds, which could contribute to water extract for both methods that demonstrates a low EC_{50} value and a similar polyphenolic profile. The solubility of antioxidant compounds in a solvent depends on the characteristics of the phytochemicals present in the sample (Morales-Olán et al., 2020). The compound extracted from plants affects the antioxidant activity, as significant antioxidant activities revealed by the extract is in relation to the presence of flavonoids, tannins, and other alkaloids (Akshayaa et al., 2021).

As mentioned above, these findings align with a previous study where an ethanol-water solvent system via maceration of an AI plant reported that 70% ethanol in water gave the lowest EC50 value of 47.064 µg/ml as compared to other ethanol concentrations, given that DPPH concentration was 0.2 mM of DPPH. The report conducted further analysis using UHPLC-Q-Orbitrap HRMS, which revealed 6 out of 10 is phenolic, including quinic acid, and the rest is flavonoids (Salma Iza et al., 2023). This highlights the critical role of solvent polarity in extracting antioxidative polyphenolic compounds. Other research also supports the link between high phenolic content and antioxidant activity. For example, a Soxhlet extraction of water and the whole AI plant reported an EC_{50} of 0.089±0.003 mg/ml due to a high TPC of 1016.73±41.10 mg GAE/g (Zahidin et al., 2018). Similarly, a Soxhlet extraction using 30% ethanol of aerial parts had an EC_{50} of 62 µg/ml and roots had an EC_{50} of 206 µg/ml using 1 mM of DPPH, which suggested that by varying the ethanol-water ratio, the EC_{50} lowered when using aerial parts. This further reinforces that polyphenol plays a major role in antioxidant activity (N'Paki et al., 2024).

Phenols interact with the free radical by donating H-atoms; meanwhile, flavonoids reduce by electron transfer or hydrogen atom donation (Gulcin, 2020; Gülçin & Alwasel, 2023). Polyphenols like phenols and flavonoids are indeed major contributors to antioxidant activity. However, compound structure plays a role in scavenging capability. This can be further attributed to the diverse structures of polyphenolic compounds. The antioxidant activity is significantly influenced by specific structural characteristics, such as the number of hydroxyl groups, the position of the hydroxyl chain, glycosylation patterns, and the presence of double bonds in the C-ring (Huynh et al., 2025; Safe et al., 2021).

Oxidative stress is fundamental for cellular survival in all living beings. Cellular mechanisms secrete reactive oxygen species (ROS), and natural endogenous antioxidants, such as superoxidase dismutase and catalase, nullify them. This balance creates a harmony of health. However, imbalance can occur with many factors, such as ultraviolet (UV) radiation, pollution, alcohol intake, and others. This will lead to an uptake of ROS levels, which then can lead to inflammation and many more cascades of negative effects (Koksal et al., 2011; Kedare & Singh, 2011). It can be mitigated by natural exogenous antioxidants, such as plant-derived antioxidants. In this study, AI plant extracts demonstrated a potent ability to neutralise free radicals in the DPPH model, characterised by the distinct purple-to-yellow colorimetric shift. This scavenging efficiency is due to the high phenolic and flavonoid yields observed in the 40-80% ethanol solvent extracts. The low value of EC_{50} values achieved, particularly via the UAE, validate that these phytochemicals act as effective H-atom donors, providing a functional defence against the oxidative cascades described in the literature. To summarise, our findings confirm that varying ethanol-water binary solvent systems significantly extracts the polyphenol content and enhances radical scavenging activity.

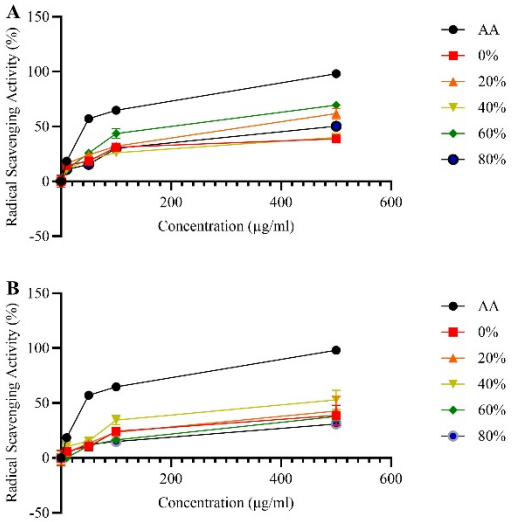

Figure 2 DPPH radical scavenging activity of each AI extract using **A**) ME and **B**) ultrasonic-assisted extraction. The antioxidant assay was performed in triplicate and represented as mean ± SD (n=3) with relative SD less than 10%. AA was utilised as a positive control.

Correlation between TPC, TFC, and Antioxidant Activity of Different AI Extracts

The correlation between TPC and TFC along with antioxidant activity with respect to each method and ethanol extract of AI plant extract was analysed by Pearson's correlation analysis. UAE & maceration exhibited a positive correlation between TPC & TFC as well as TFC & EC_{50} and a negative correlation between TPC & EC_{50} as depicted by **Table 2**. Positive correlation indicates a linear relationship, whereas negative correlation defines an opposite relationship. However, the correlation between the TPC, TFC, and the radical scavenging activity levels of the extracts was not significant. This could be due to the presence of bioactive compounds that possess antioxidant properties that were not determined within the scope of this study. The contribution to antioxidant activity of AI plant is attributed not only to phenolic compounds and flavonoids; other phytochemicals, such as alkaloids, saponins, and terpenoids, can also contribute to the antioxidant activity of plant extracts (Nyero et al., 2023). Besides, the specific extraction method used can also affect the correlation between TPC, TFC, and antioxidant activity. The utilisation of different solvents, extraction durations, and temperatures can result in differences in the extraction efficiency of various phytochemicals. These results indicated that the antioxidant activity of extracts of AI may be related, at least in part, to the presence of flavonoid compounds and phenolics.

Table 2 Pearson's correlation coefficient of TPC, TFC, and EC_{50} of ultrasound-assisted and maceration extracts

Method	Assay	Correlation		
		TPC	TFC	EC_{50}
Ultrasonic-Assisted	TPC	1	0.1172	-0.4171
	TFC	0.1172	1	0.5982
	EC_{50}	-0.4171	0.5982	1
Maceration	TPC	1	0.02106	-0.1284
	TFC	0.02106	1	0.6766
	EC_{50}	-0.1284	0.6766	1

CONCLUSION

A comparison of both methods of extraction reveals UAE was found to be more efficient as compared to maceration, with a high yield of phenolic and flavonoid recovery in a shorter period of time and less consumption of solvent. It is recommended to perform optimisation on the extraction parameters to determine the factors, such as solvent concentration, extraction time, and sample-to-solvent ratio, that significantly affect the measurable variable. Response surface methodology (RSM) is a statistical and mathematical technique that can be used to optimise the extraction conditions by analysing the influence of multiple factors on the yield of the target compounds. Additionally, both spectroscopy and chromatography analysis need to be considered to discover detailed profiles of the bioactive constituents in the AI plant that contribute to the antioxidative system.

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Conflict of Interest

The author(s) declare(s) that there is no conflict of interest regarding the publication of this paper.

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