



Analysis of total polyphenol and flavonoid content, antioxidant potential and antibacterial activity of *Parkia speciosa* pod and seed extracts

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ABSTRACT. *Parkia speciosa* Hassk is a legume that is widely utilized as a cooking ingredient and valued for its medicinal properties throughout the Nagaland state in India. This research offers an overview of the phytochemical content, antioxidant properties and antibacterial activity of *P. speciosa* pods and seeds extracted in various solvents. The qualitative analysis revealed the presence of diverse compounds, including tannins, saponins, flavonoids, terpenoids and alkaloids across all extracts. The methanolic extract exhibited the highest levels of phytoconstituents, with total polyphenol content (TPC) and total flavonoid content (TFC) measuring 581.93 mg GAE g⁻¹ and 360.83 mg CE g⁻¹ respectively. Antibacterial activity differed based on the solvent and the bacterial type, with methanolic extracts demonstrating significantly greater effectiveness than acetone and chloroform. Additionally, *Escherichia coli* was more susceptible than *Staphylococcus aureus*. The antioxidant activity was significantly affected by the concentration of extracts and solvents used ($p < 0.05$), with methanolic extract demonstrating the highest scavenging activity, followed by acetone and chloroform extracts. Although pod extracts had higher TPC and TFC as well as stronger antioxidant capacity, seed extracts exhibited higher antibacterial activity. *Parkia speciosa* is not only an edible plant but also a significant source of medicinal properties, offering great potential for use in the pharmaceutical industry.

Keywords: *Parkia speciosa*; phytochemical; solvents; antioxidant; antibacterial.

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Introduction

The modern world presents complex sceneries of health challenges, but it also offers a wealth of solutions through plant-derived natural herbal remedies. Plants are rich in diverse bioactive compounds with medicinal properties, making them stand out as valuable resources for the development of safe and eco-friendly drugs. This vast array of phytochemicals positions plants as exceptional resources for discovering promising new leads in the fields of drug research and development. Despite the widespread availability and effectiveness of synthetic drugs for treating ailments in today's modern world, some people opt for traditional folk remedies using plants due to their less harmful effects and cost-effectiveness along with easy accessibility especially for people residing in rural areas.

Parkia speciosa, an edible legume under the Fabaceae family, is locally called 'Yongchak' in Nagaland and some other areas. It is also goes by other names such as bitter bean, twisted cluster bean, sator bean, stink bean, or petai. The flowers, pods and seeds comprise the consumable parts of the tree. It is eaten as a vegetable and salad in Indonesia, Malaysia, Singapore, Thailand, Borneo, Madagascar, Africa, India and other parts of the world (Singhania et al., 2021). In a local Indian delicacy, the flower is popularly consumed raw as a side dish known as 'singju' or by combining it with traditionally fragmented pickles (Chhikara et al., 2018). In Nagaland, the fresh pods are eaten raw or cooked while seeds in their fresh or dried conditions are consumed purely raw, or after cooking or roasting as a side dish or a vegetable. *P. speciosa* seeds contain protein, fat and carbohydrates and are also a good source of minerals with a considerable amount of vitamin C and alpha-tocopherol (Kamisah et al., 2013). It also possesses hypoglycaemic, antioxidant, anti-inflammatory and anti-hypertensive properties (Azemi et al., 2022) and exhibit anti-cancer, antioxidant, anti-bacterial as well as anti-angiogenic activity (Ghasemzadeh et al., 2018). These abundant bioactive properties found in *P. speciosa*

contribute to a diverse array of health advantages and underline the plant's pharmacological value. The plant has also industrial applications such as the development of activated carbon and the production of dye (Chhikara et al., 2018).

The study aimed to screen phytochemicals and quantify TPC and TFC in *P. speciosa* pod and seed extracts and to explore their potential as an antioxidant and antimicrobial as well as to study the effect of different solvents on these properties. The present study will provide an insight into the diverse phytoconstituent of *P. speciosa* available in Nagaland which will unveil its potential medicinal application. This piece of work will provide valuable insights into untapped avenues for further exploration and research and harness its potential for human health and well-being while also contributing to the conservation and sustainable utilization of biodiversity in Nagaland.

Materials and methods

Chemicals and reagents

Gallic acid and catechin were purchased from Sigma Aldrich, India. Folin-Ciocalteu reagent, 2,2-diphenyl-1-picrylhydrazyl (DPPH), ascorbic acid, aluminium chloride, acetone, methanol, sulfuric acid (H₂SO₄), hydrochloric acid (HCl), sodium hydroxide, sodium carbonate, sodium hydroxide, chloroform, ferric chloride, ammonia, glacial acetic acid, potassium dichromate and potassium ferricyanide were procured from Merck, India. Culture media were procured from HiMedia, India. All the chemicals and reagents used were of analytical grade.

Collection and preparation of plant material

Fresh legumes of *P. speciosa* were collected in February 2024 from Samaguri, under Dimapur district (latitude 25.919684° N and longitude 93.785187° E), situated in Nagaland (Figure 1). The collected sample was thoroughly washed under running tap water to eliminate any impurities. The outer layer of the pod was scraped out and the pod with seeds was cut into pieces. The seeds were removed from pods and were then dried in an oven at 35°C for one week. Dried plant samples were blended into fine powder with a kitchen blender and stored under refrigeration for further analysis.



Figure 1. *Parkia speciosa* in its natural habitat.

Preparation of plant extract

The extraction process for seed and pod powder was carried out in methanol, acetone and chloroform in 1:10 ratio (w/v) using the Soxhlet apparatus. The extraction proceeded through five cycles to ensure thorough

extraction of the desired compounds. The solution was collected and the solvent was removed by evaporating it to dryness. The extracted compounds were then carefully stored under refrigeration for subsequent analysis.

Screening of phytochemicals

Extracts were screened for phytochemicals using standard protocols (Iqbal et al., 2015; Shaikh & Patil, 2020; Alemu et al., 2024).

Carbohydrate test: 1 mL each of Fehling's solution A and B along with 1 mL of filtrate was boiled in a water bath. The formation of a red precipitate indicates a positive result.

Test for glycoside: 0.5 mL glacial acetic acid was added to 2 mL of extract followed by 2-3 drops of FeCl_3 and 1 mL of 98 % H_2SO_4 . The formation of deep-blue color at the junction of two liquids, indicates the presence of glycoside.

Test of steroid: 2 mL of extract was transferred to 2 mL of chloroform and 2 mL of 98 % H_2SO_4 . Steroid was detected by occurrence of red color at the lower layer.

Alkaloid test: 15 mg of extract was added to 6 mL of 1 % HCl, warmed in a water bath for 5 min and filtered, then divided into three equal parts.

a. **Dragendorff's test:** 1 mL of Dragendorff's reagent was added to one portion of the filtrate. The mixture was observed for an orange-red precipitate.

b. **Mayer's test:** 1 mL of Mayer's reagent was added to one portion of filtrate. The mixture was observed for cream-colored precipitate.

c. **Wagner's test:** A few drops of Wagner's reagent were added to one portion of the filtrate. The mixture was observed for a brown colored precipitate.

Test for phenols: A few drops of potassium dichromate solution were added to the plant extract. Phenol is detected by the formation of dark color.

Tannin test: 0.5 mL of plant extract was mixed with 5 mL of 10 % NaOH and shaken well. The formation of an emulsion indicates the presence of tannin.

Detection of flavonoids:

a. **Alkaline reagent test:** 1 mL of the extract was mixed with 2 mL of 2 % NaOH solution, the resulting mixture exhibits an intense yellow color, which turns colorless upon the addition of diluted hydrochloric acid.

b. **Ferric chloride test:** A few drops 10 % ferric chloride solution was added to the extract solution, the resulting mixture shows a green precipitate.

Detection of anthraquinones: 10 mL of 10 % ammonia solution was added to a few mL extract. The solution was shaken for 30 seconds. The positive result is indicated by the formation of pink, violet, or red colored solution.

Test for protein

a. **Xanthoproteic test:** Extract was mixed with a few drops of concentrated nitric acid. A positive result is indicated by transformation to yellow-colored solution.

b. **Biuret Test:** 2 mL of plant extract was mixed with 1 drop of 2 % copper sulphate solution followed by 1 mL of 95 % ethanol and few KOH pellets. A positive result is indicated by formation of a pink-coloured solution in the ethanolic layer.

Saponin test: 0.5 g of plant extract was shaken vigorously with 10 mL of distilled water and kept in water and bath for 5 min. The formation of froth indicated the presence of saponin.

Terpenoid test: 5 mL plant extract was dispensed in 2 mL chloroform followed by 3 mL concentrated H_2SO_4 . The formation of the grey-colored solution when the solution was boiled in a water bath indicated the presence of a terpenoid.

Quantitative estimation of phytochemicals

Total polyphenol content

TPC in the extract was determined using Folin-Ciocalteu reagent as described by Wonghirundecha et al., (2014) after some modification. Solvent extracts of seed and pod were diluted from stock solutions of 10 mg mL^{-1} and standard gallic acid solutions ($10\text{-}180 \text{ mg mL}^{-1}$) were prepared from the stock solution of 200 mg mL^{-1} . 4 mL of 10 % Folin-Ciocalteu reagent was added to 400 μL of each extract, as well as, standard solutions and mixed thoroughly. The mixtures were allowed to stand at room temperature for 5 min and 3.2 mL of 10 %

Na₂CO₃ was added. The final solution was made up to 8mL with distilled water and incubated in dark at room temperature for 90 min. Absorbance was measured at 750nm using a UV spectrophotometer. TPC was determined from the standard curve of gallic acid and expressed as mg gallic acid equivalent (GAE) per gram dry weight.

Total flavonoid content

TFC was determined following Ghasemzadeh et al., (2018) after slight modification. 10mg of pod and seed extract were separately dissolved in 10 mL methanol. Simultaneously, various concentrations of catechin standard solutions were prepared from a stock solution of 10 mg mL⁻¹. From each sample, 3 mL of the solutions was mixed with 3mL of 2 % aluminium chloride solution. The mixture was then incubated in darkness for 10 min and the absorbance was measured at 415 nm using a spectrophotometer. A calibration curve was established from the standard solutions and TFC was expressed in milligrams of catechin equivalent (CE) per gram dry weight.

Antibacterial activity

Culture of bacteria and agar well diffusion assay

Two bacterial strains, namely *Escherichia coli* (*E. coli*) ATCC 25922 and *Staphylococcus aureus* (*S. aureus*) ATCC 25932 were used as test organisms. Bacteria were sub-cultured by inoculating from stock to freshly prepared Muller Hilton agar plates and incubated for 24 hours at 37°C. Broth cultures of each test bacteria were prepared in Muller Hilton broth.

The ability of seed and pod extracts to inhibit bacteria was determined by agar-well diffusion assay as outlined by Wonghirundecha et al., (2014) after some modifications. 1mL of freshly prepared bacterial culture was added under aseptic condition to sterilized Muller-Hinton agar, mixed gently, poured into petri-dish and allowed to solidify at room temperature of laminar air-flow. After the media solidified, wells with a diameter of 6mm were created in each plate. Wells were then filled with 20 µL each of seed and pod extract pre-prepared at a concentration of 20 mg mL⁻¹ and 1mg mL⁻¹ of kanamycin which served as a positive control. Plates were kept for 30 min at room temperature and were incubated at 37°C for 24 hours. The zones inhibited by bacteria around wells were measured.

1,1-diphenyl-2-picrylhydrazylfree-radical scavenging assay

The antioxidant ability of various extracts of *P. speciosa* seed and pod to scavenge free radical DPPH was carried out following Adil et al., (2024) after some modifications. Stock solution in 100 µg mL⁻¹ concentration of plant samples was prepared by dissolving 25mg extracts in 250 mL of methanol. Various concentrations of extracts (10-100 µg mL⁻¹) were prepared from the stock solution. Ascorbic acid was taken as a reference standard and methanol served as a control. 5mL of each concentration as well as standard and control were taken separately and 1 mL of 0.1M of DPPH solution was added to it. The solutions were kept at room temperature under dark for 30 min and observed at a wavelength of 517 nm. The percentage of DPPH inhibition was determined by using the formula:

$$\text{Scavenged DPPH (\%)} = \frac{\text{Absorbance of control} - \text{Absorbance of test or standard sample}}{\text{Absorbance of control}} \times 100$$

The IC₅₀ value is determined using the regression curve.

Statistical analysis

Data of each sample are expressed as the mean ± standard error mean (SEM) of triplicates. Significant differences in parameters studied were analyzed using one-way analysis of variance (ANOVA) followed by Tukey's post-hoc test. A value of p < 0.05 was considered to be statistically significant. The analyses were performed using the SPSS software version 23.

Results and discussions

Screening of phytochemicals

Phytochemical screening is the identification of different groups of phytoconstituents that are present in various parts of a plant. Phytochemicals are renowned for their medicinal benefits with primary metabolites

driving essential biochemical processes like respiration and photosynthesis, while secondary metabolites protect plants and enhance their aroma, color and flavor (Saleh et al., 2021). Screening of phytochemicals aids in revealing the components of the plant extracts and contributes to searching for bioactive agents that can be utilized as dietary supplements and medicine. In this study, alkaloids, saponins, flavonoids, carbohydrates, phenols and tannins were detected in all extracts (Table 1). Similarly, Balaji et al., (2015) also reported the presence of alkaloid, saponin, flavonoids, phenols and tannins in pod extracts of *P. speciosa*. These phytochemicals are recognized for their beneficial properties, for instance, alkaloids can lower total cholesterol, triglycerides, low-density lipoprotein and atherogenic indices while raising high-density lipoprotein and flavonoids and tannins also exert antioxidant effects by reducing triglycerides and low-density lipoprotein (Tandi et al., 2020). Among the extracts, methanolic extracts yielded the highest number of positive results, when compared with acetone and chloroform showcasing the importance of solvent in extracting phytochemicals from *P. speciosa*. Anthraquinones and starch were detected only in pod while terpenoid was found only in seed. When seed and pod were compared, higher numbers of positive results were observed in pod extracts which indicate the presence of a richer phytochemical profile in pods and the need for emphasizing the significance of exploring more into pod-derived phytoconstituents for medicinal applications. Further exploration into the isolation, characterization and pharmacological evaluation *P. speciosa* could lead to the development of new therapeutic agents with extensive health benefits.

Table 1. Phytochemical screening of seed and pod extracts of *Parkia speciosa*.

Phytoconstituents	Test	SM	SA	SC	PM	PA	PC
Alkaloids	Dragendorff	-	-	+	-	+	+
	Mayer's	+	+	-	-	+	+
	Wagner's	++	++	+	+	++	++
Saponin		++	++	++	++	++	++
Flavonoids	Alkaline reagent	+	+	+	+	+	+
	Ferric chloride	+	-	-	+	+	+
Anthraquinones	Borntrager's	-	+	-	-	-	-
Protein	Xanthoproteic	++	++	++	-	+	+
	Biuret	+	-	+	-	-	-
Terpenoid		-	-	-	+	-	-
Tannin		+	+	+	+	+	+
Carbohydrates		+	++	++	++	++	++
Phenols		++	++	+	+	+	+
Starch		+	-	-	-	-	-

++ indicates highly positive; + indicates positive; - indicates negative result; SM, SA and SC indicates methanolic, acetone and chloroform extracts of seed; PM, PA and PC indicates methanolic, acetone and chloroform extracts of pod.

Total polyphenol and total flavonoid content

Phenolics and flavonoids are naturally occurring phytochemicals with broad-spectrum biochemical activities that account for most of the antioxidant activities in plant products (Cheong et al., 2024). As given in Figure 2 and Figure 3, the TPC and TFC in pod and seed of *P. speciosa* differed significantly ($p < 0.05$) from 264.35 to 581.93 mg GAE g^{-1} and 252.80 to 360.83 mg CE g^{-1} respectively. TPC was higher than the prior reports of 2464.32 mg GAE $100 g^{-1}$ of edible portion in methanolic extract (Ayub Ali et al., 2011) and 14.16 mg GAE $g^{-1}m$ (Balaji et al., 2015). This could be due to various factors such as differences in cultivar, stage of plant, harvesting period and techniques, climatic conditions and methods of extraction. A comparison between pod and seed samples displayed higher TPC in pod (285.45 mg GAE g^{-1} to 581.93 mg GAE g^{-1}) than in seed (264.35 mg GAE g^{-1} to 525.65 mg GAE g^{-1}). Likewise, TFC was higher in pod (270.83 to 360.83 mg CE g^{-1}), as compared to seed (252.50 to 344.17 mg CE g^{-1}). This observation proposes that *P. speciosa* pods might serve as richer reservoirs of polyphenols and flavonoids within the plant.

The assessment of TPC and TFC was carried out in various solvent extracts which displayed significant differences ($p < 0.05$) depending on the solvent used for extraction. The methanolic extract exhibited the highest TPC in pod (581.93 mg GAE g^{-1}), as well as in seed (525.65 mg GAE g^{-1}). A similar result was recorded for TFC where the methanolic extract exhibited the highest TFC in pod (360.83 mg CE g^{-1}), as well as in seed (344.17 mg CE g^{-1}). The high efficiency of methanol to extract TPC and TFC from samples may be due to its ability to interact easily with the major polar phenolic compounds owing to its strong polarity. Contrarily, the chloroform extracts showed the lowest TPC and TFC, suggesting that chloroform may not be as effective as methanol and acetone in the extraction of phenolic compounds from *P. speciosa* seed and pod.

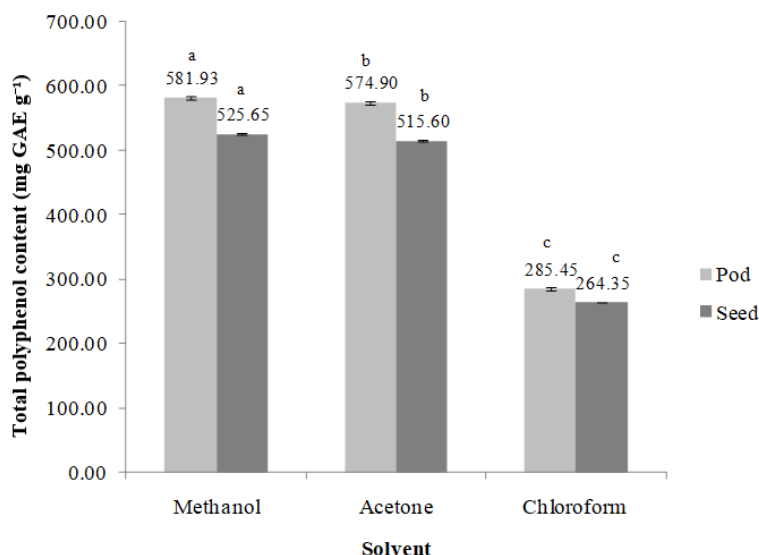


Figure 2. Total polyphenol content of *Parkia speciosa* seed and pod. Values with different letters in each sample indicate significant difference at $p < 0.05$; error bars represent mean \pm standard error mean.

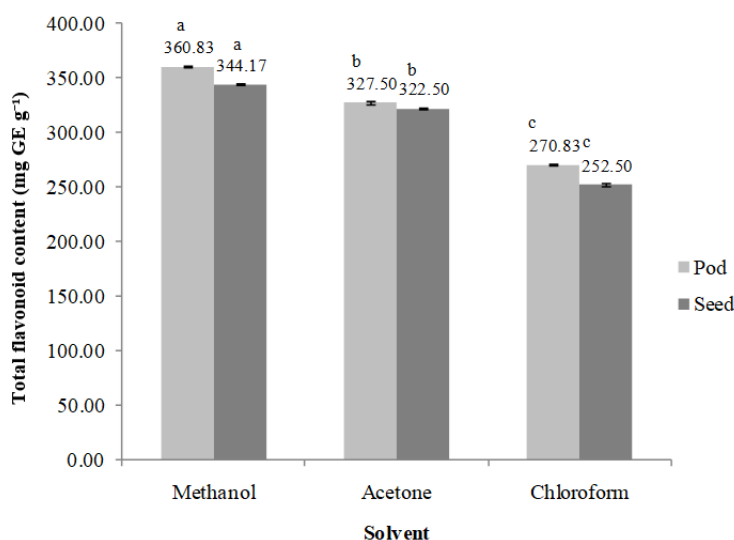


Figure 3. Total flavonoid content of *Parkia speciosa* seed and pod. Values with different letters in each sample indicate significant difference at $p < 0.05$; error bars represent mean \pm standard error mean.

Antibacterial activity

The ability of *P. speciosa* to inhibit bacteria was carried out using the agar well diffusion method, which was indicated by the formation of a transparent zone around the wells inoculated with the sample (Figure 4). *P. speciosa* exhibited significant antibacterial activity against gram-positive and gram-negative bacteria, as shown by the zones of inhibition presented in Table 2. The solvent used significantly impacted the antibacterial effectiveness ($p < 0.05$). Methanolic extract demonstrated the strongest inhibitory effect, with the inhibition zones measuring 3.4cm (pod) and 1.3cm (seed) against *E. coli* and 3.5cm (pod) and 1.2cm (seed) against *S. aureus*. Strong antimicrobial potential of methanolic extracts has been associated with their high polarity, which facilitated the extraction of all phenolic compounds (Mehmood, & Murtaza, 2018). Interestingly, the seed extract exhibited greater antibacterial activity than the pod extract, which contrasts with the TPC and TFC results, where the pod had higher contents of both. This result suggests that, in addition to polyphenols and flavonoids, other phytochemicals, potentially more abundant in the seeds than in the pods may contribute to the observed antibacterial effect against the tested bacterial strains. The resistance of bacteria to antimicrobial agents has been a major challenge in treating infectious diseases, where new strains of gram-positive and gram-negative are armored with alarmingly high levels of resistance. These findings offer a potential solution in the ongoing battle against resistant bacterial strains.

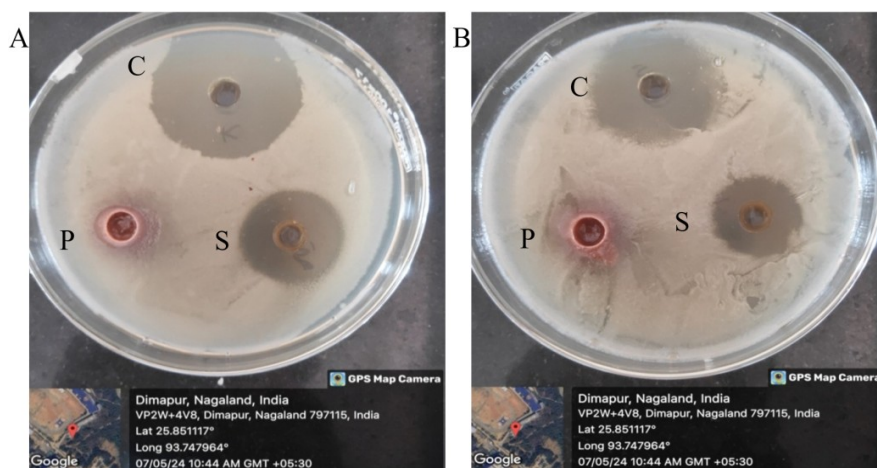


Figure 4. Some representative images of Petri plates showing zone of inhibition. Antibacterial activity of methanolic extracts of *Parkia speciosa* against (A) Antibacterial activity of methanolic extracts of *Parkia speciosa* against *Escherichia coli* and (B) Antibacterial activity of methanolic extracts of *Parkia speciosa* against *Staphylococcus aureus*. C - control (kanamycin); S - seed extracts; P - pod extracts.

Table 2. Inhibition by *Parkia speciosa* extracts against bacteria.

Sample	<i>Escherichia coli</i>			<i>Staphylococcus aureus</i>		
	Methanol	Acetone	Chloroform	Methanol	Acetone	Chloroform
Seed	3.33 ± 0.09 ^a	2.40 ± 0.06 ^b	1.03 ± 0.03 ^c	3.27 ± 0.07 ^a	2.13 ± 0.03 ^b	1.33 ± 0.03 ^c
Pod	1.30 ± 0.06 ^a	1.07 ± 0.03 ^b	1.03 ± 0.03 ^b	1.13 ± 0.03 ^a	1.03 ± 0.03 ^a	0.83 ± 0.03 ^b

Means with different lowercase in the same row are significantly different.

Antioxidant activity

The DPPH assay is a commonly used method for assessing the free radical scavenging capacity in plant extracts. Antioxidants can scavenge radicals like DPPH through several mechanisms, such as transferring a single electron followed by a proton, single-step transfer of hydrogen atom or by sequential proton loss electron transfer (Yamauchi et al., 2024). As displayed in Table 3, the antioxidant activity of *P. speciosa* was significantly influenced by the concentration of extracts as well as the type of solvents used ($p < 0.05$). Sample extracts as well as the standard exhibited its highest DPPH scavenging activity at 100 $\mu\text{g mL}^{-1}$ with inhibition ranging from 76.52 to 89.90 %. The ascorbic acid had a significantly higher scavenging activity followed by methanolic, acetone and chloroform extract ($p < 0.05$). On the contrary, IC₅₀ which represents the concentration at which the solvent extract or ascorbic acid scavenges the formation of radicals by 50 % displayed lower IC₅₀ by ascorbic acid than those of all extracts tested (Figure 5) which explains its strong radical scavenging activity. The antioxidant activity followed a similar pattern to TPC and TFC, with methanolic extracts showing the highest activity among the solvents used and pod exhibiting greater activity than seed, indicating the influence of phenolic compounds on antioxidant activity. Phenolic compounds act as powerful in vitro antioxidants by donating hydrogen atoms and electrons, thereby stabilizing free radicals (Roy et al., 2024).

Table 3. Antioxidant activity of different extracts of pod and seed of *Parkia speciosa*.

Concentration ($\mu\text{g mL}^{-1}$)	Inhibition (%)						
	Standard	PM	PA	PC	SM	SA	SC
10	18.17 ^{fA} ± 0.15	16.96 ^{fB} ± 0.30	14.49 ^{fC} ± 0.28	10.64 ^{fD} ± 0.22	14.40 ^{fC} ± 0.18	13.85 ^{fC} ± 0.17	9.48 ^{fE} ± 0.15
20	26.36 ^{eA} ± 0.13	24.96 ^{eB} ± 0.21	22.17 ^{eC} ± 0.28	18.84 ^{eE} ± 0.28	21.73 ^{eD} ± 0.15	20.99 ^{eD} ± 0.15	17.24 ^{eF} ± 0.17
40	45.63 ^{dA} ± 0.30	42.40 ^{dB} ± 0.26	39.61 ^{dC} ± 0.37	36.54 ^{dE} ± 0.28	40.32 ^{dC} ± 0.19	38.00 ^{eD} ± 0.20	34.32 ^{dF} ± 0.23
60	60.94 ^{cA} ± 0.17	59.58 ^{cA} ± 0.35	56.57 ^{cB} ± 0.43	51.73 ^{cC} ± 0.34	55.23 ^{cB} ± 0.18	53.14 ^{cC} ± 0.24	49.46 ^{cD} ± 0.26
80	75.01 ^{bA} ± 0.28	71.56 ^{bB} ± 0.32	68.02 ^{bD} ± 0.43	63.31 ^{bF} ± 0.26	69.88 ^{bC} ± 0.19	65.04 ^{bE} ± 0.24	61.23 ^{bG} ± 0.22
100	89.90 ^{aA} ± 0.15	87.21 ^{aB} ± 0.30	84.03 ^{aC} ± 0.47	78.99 ^{aE} ± 0.30	84.62 ^{aC} ± 0.24	80.59 ^{aD} ± 0.15	76.52 ^{aF} ± 0.26

Means with different lowercase in the same column are significantly different; means with different uppercase in the same row are significantly different; SM, SA and SC indicates methanolic, acetone and chloroform extracts of seed; PM, PA and PC indicates methanolic, acetone and chloroform extracts of pod; S indicates ascorbic acid

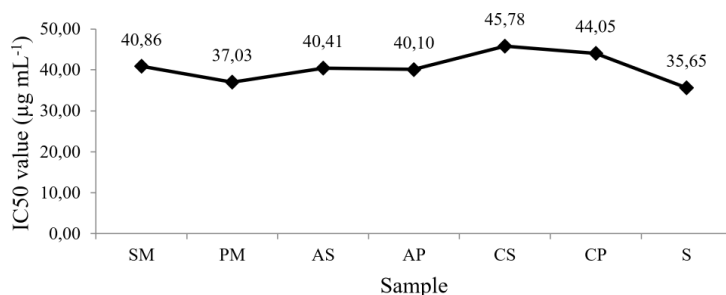


Figure 5. IC₅₀ of standard and different solvent extracts of *Parkia speciosa*. SM, SA and SC indicate methanolic, acetone and chloroform extracts of seed; PM, PA and PC indicates methanolic, acetone and chloroform extracts of pod; S indicates ascorbic acid.

Correlation analysis

Pearson's correlation study revealed a significantly positive relation among various parameters at $p < 0.01$ (Table 4). A significantly strong correlation was established between TPC and TFC ($r^2 = 0.902^{**}$), antibacterial activity ($r^2 = 0.651^{**}$) and antioxidant activity ($r^2 = 0.766^{**}$) and between TFC and antioxidant activity ($r^2 = 0.957^{**}$). A strong positive correlation was also observed between TPC and TFC ($r^2 = 0.902^{**}$). This is expected, as both compound classes are known to contribute in plant antioxidant activity (Muflihah et al., 2021). Another study demonstrated a strong correlation between TPC, TFC and antioxidant activity (El Mannoubi, 2024). The higher level of TPC and TFC in *P. speciosa* might have been one of the reasons for high antioxidant activity as well as antibacterial activity because, in both seed and pod extract, the highest DPPH and antibacterial activity and the lowest IC₅₀ value had the highest TPC and TFC. *P. speciosa* extracts contain great DPPH radical scavenging activity and their high antioxidant activities is associated with high TPC and TFC (Ko et al., 2014). Muflihah et al., (2021) demonstrated that there is a strong linear relationship between antioxidant activity and TPC, highlighting TPC as the predominant antioxidant compound. Another study also correlates the antioxidant activity of plant extract with the presence of phenolic compounds (Chlif et al., 2022). Phenolic compounds work through various mechanisms, such as interfering with microbial cell wall formation, inhibiting the synthesis of proteins and nucleic acids, disrupting metabolic processes and compromising the integrity of cell membranes (Komolafe et al., 2024).

Table 4. Correlation between phenolics, antioxidant and antibacterial activity of *Parkia speciosa*.

	Total polyphenol content	Total flavonoid content	Antibacterial activity
Total flavonoid content	0.902 ^{**}		
Antibacterial activity	0.651 ^{**}	0.45	
Antioxidant activity	0.766 ^{**}	0.957 ^{**}	0.24

^{**}: Correlation is significant at the 0.01 level (2-tailed).

Conclusion

Parkia speciosa is a powerhouse of phytochemicals and a strong source of antioxidants with commendable antibacterial activities. The amount and efficiency of phytoconstituents are significantly influenced by the type of solvents. Methanol was the most effective solvent in extracting phytochemicals, yielding a higher quantity of polyphenol and flavonoid with potent antioxidant properties and antibacterial activity, compared to acetone and chloroform. The high levels of TPC and TFC in *P. speciosa* are likely responsible for its strong antioxidant activities highlighting the significance of phenolics in enhancing bioactivity. Although the pod extract had higher levels of TPC and TFC and demonstrated stronger antioxidant activity, the seed extract exhibited greater antibacterial activity suggesting the along with polyphenols and flavonoids other compounds also contribute to its antibacterial effects. These findings offer baseline information for deeper investigation into the potential of *P. speciosa* as a valuable plant. Further research could explore more into recognizing and distinguishing distinct phenolic compounds present in these extracts and exploring their potential bioactivities and remedial applications.

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