

Comparative FTIR Profiling, Phytochemical Content and Antioxidant Potential of Ethanolic Leaf Extracts of *Millettia atropurpurea*

Sathivel Arumugam^{1*} and Durgashini Pichaimtuhu²

^{1,2}Faculty of Medicine, Biochemistry Unit, AIMST University, 08100 Bedong, Kedah, Malaysia
biosakthi@gmail.com*

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Abstract

This study analyzed the comparative FTIR profiling, phytochemical content, and antioxidant potential of ethanolic leaf extracts of *Millettia atropurpurea*. The initial screening showed that both extracts contained flavonoids, alkaloids, phenols, tannins, and saponins, but terpenoids were only found in the 70% of the extract. Using FTIR analysis, it was found that chemical signs that match phenolic and flavonoid compounds. TLC testing showed similar movement patterns to gallic acid and quercetin, which supports the presence of these compounds. To check antioxidant activity, the DPPH method was used, and the 70% extract had better results (IC₅₀ = 94.7 µg/mL) than the 90% extract (IC₅₀ = 124.6 µg/mL), with gallic acid showing the strongest activity (IC₅₀ = 22.54 µg/mL). The 70% extract had higher total phenolic content (23.02 mg GAE/g), while the 90% extract had more total flavonoids (31.80 mg QE/g). Statistical analysis showed that the results between the two extracts were significantly different ($p < 0.05$). Overall, the study provides basic scientific support for the antioxidant qualities of *M. atropurpurea*, making it a promising source of active ingredients from nature.

Keywords: *Millettia atropurpurea*, phytochemical, ethanolic leaf extract, FTIR, antioxidant potential.

Introduction

Many medicinal plants are used as alternative treatments for diseases and most of them don't have the side effects that synthetic drugs often do. Understanding the chemical makeup of the active compounds in these plants can help explain which parts of their structure are responsible for their healing effects (Kumar and Ramaswamy, 2014). Plants usually contain various types of chemical compounds, such as phenolics, flavonoids, glycosides, coumarins, saponins, and terpenoids, which give them their unique properties and help them work in the body (Ajayi *et al.*, 2017). The genus *Millettia* includes over 260 species that belong to the Fabaceae (Leguminosae) family and are found in tropical and subtropical areas around the world. These plants are well-known in traditional medicine for treating a range of human and animal health issues, such as inflammation, menstrual problems, and cancer. Chemically, *Millettia* is rich in various types of isoflavonoids, especially prenylated isoflavones and rotenoids, along with other related compounds like isoflavans, glycosylated isoflavones, pterocarpanes and coumaronochromenes. These active compounds show the therapeutic potential of *Millettia* species and make them important for research into new medicines (Jena *et al.*, 2020). More than 150 isoflavonoids have been found in different parts of selected *Millettia* species, showing a wide range of chemical structures and potential health benefits, including cancer-fighting, anti-inflammatory, antibacterial, and blood sugar-lowering effects. Because of the large number of active compounds they contain, the genus *Millettia* has drawn a lot of attention from scientists for studying their chemical makeup, how to extract them, and their possible uses in developing new drugs and medicines (Rajemiaramiraho *et al.*, 2014). This study offers scientific proof about the chemical makeup and antioxidant abilities of *Millettia atropurpurea* leaves, adding to the small amount of information we have about this plant. FTIR (Fourier Transform Infrared) spectroscopy has been very important in studying medicinal plants. FTIR is one of the most commonly used methods to find out the chemical components, functional groups, and the structure of these chemicals.

*Corresponding author

The results from FTIR were very precise and consistent (Lethika et al., 2013). Identifying key functional groups using FTIR and finding phenolic and flavonoid compounds through TLC gives helpful early insights into the plant's active chemicals. Measuring the total levels of phenolic and flavonoid compounds, along with testing antioxidant activity using the DPPH method, can help link the plant's chemical makeup to its antioxidant effects. The results of this research could support the idea of using *M. atropurpurea* as a natural source of antioxidants and lay the foundation for more studies on its chemical, medical, and industrial uses. The goal of this study was to check the phytochemical content and FTIR spectrum, measure total phenolic and flavonoid levels, and assess antioxidant activity in the ethanolic leaf extract of *Millettia atropurpurea*.

Materials and Methods

Leaves Collection and Preparation: About 500 g of *Millettia atropurpurea* leaves were collected from the parking area at AIMST University, Malaysia. After collection, they were rinsed well with clean water to clear away soil and other particles, and then carefully blotted with tissue or a cloth to take off leftover water on the surface. Next, the washed leaves were laid out in a uniform layer on a table in an open area and left to dry naturally until no visible wetness remained. Once they had partly dried, the leaves were placed into two ovens maintained at 40°C and kept there for 24 hours, so that moisture could be fully eliminated. Their dryness was then assessed; if any dampness was still present, the leaves were put back into the oven and dried longer until completely dry. To verify that all moisture was gone, the dried leaves were lightly pressed, after which they were milled into a fine powder with a clean, dry blender. The resulting powder was placed into sealed, airtight containers, marked with labels, and kept in a dry place at room temperature.

Maceration extraction methods: To extract the leaf material, the maceration method was used with two different ethanol strengths. Each of two 500 mL conical flasks received exactly 50 g of dried leaf powder. Two separate solvent mixtures were made by combining absolute ethanol with distilled water to create 70% and 90% ethanol solutions. The resulting ethanolic extracts were then concentrated using a rotary evaporator.

Preliminary phytochemical screening: An initial survey of phytochemicals in leaf extracts from *Millettia atropurpurea* was performed, aiming to identify key secondary compounds through established qualitative assays. Multiple phytochemical assays were applied to the ethanolic root extract in order to determine whether diverse phytoconstituents were present, including flavonoids, alkaloids, glycosides, polyphenols, tannins, terpenoids, and saponins.

FTIR spectrum analysis: The FTIR spectrum of the ethanolic leaf extract from *Millettia atropurpurea* was recorded using KBr as the sample medium with a computer-controlled FTIR BRUKER spectrometer. A He-Ne laser was used as a reference (Manickam and Mohan, 2014). The FTIR spectrum helps in identifying the functional groups present in *M. atropurpurea*. The extract of *M. atropurpurea* was scanned over the spectral range of 4000 to 400 cm⁻¹.

TLC profiling: Thin layer chromatography (TLC) analysis was carried out to identify the presence of phenolic and flavonoid compounds in the *M. atropurpurea* extracts. The dried E70 and E90 extracts were dissolved in ethanol to obtain a concentration of 10 mg/mL. Gallic acid and quercetin standards were prepared at a concentration of 1 mg/mL in ethanol. A silica gel 60 F254 TLC plate (5 × 10 cm) was prepared and a baseline was drawn at 1 cm from the bottom edge using a pencil. Three equidistant points were marked on the baseline for the standard and extracts. Using capillary tubes, 2–5 µL of each solution (standard, E70 and E90) were spotted onto the marked points, allowing each application to dry before re-spotting to avoid overloading. For phenolic analysis, the mobile phase consisting of chloroform: methanol: formic acid (8:1:1) was prepared and transferred into a TLC chamber. The chamber was covered and allowed to saturate for 10 minutes. The spotted plate was placed inside the chamber and developed until the solvent front reached approximately 8 cm from the baseline. The plate was removed and the solvent front was immediately marked and air-dried. The developed plate was observed under UV light at 254 nm. The plate was then sprayed evenly with 1% FeCl₃ reagent and allowed to dry. The appearance of blue or dark spots indicated the presence of phenolic compounds. For flavonoid analysis, a new TLC plate was prepared and spotted with quercetin, E70 and E90 as described above. The mobile phase consisting of toluene: ethyl acetate: formic acid (5:4:1) was prepared and the chamber was saturated for 10 minutes.

The plate was developed to approximately 8 cm, removed and the solvent front was marked. The plate was observed under UV light at 254 nm and 365 nm. The plate was then sprayed with 1% AlCl_3 reagent and allowed to dry for 2–5 minutes. The plate was re-observed under UV light at 365 nm, where yellow or green fluorescent spots indicated the presence of flavonoid compounds.

Antioxidant activity (DPPH radical scavenging assay): Antioxidant activity of ethanolic leaf extract was determined using DPPH free radical scavenging activity (Blois, 1958). About 1 mL of 0.1 mM solution of DPPH in methanol was added to 2.5 mL of the ethanolic leaf extract (70% and 90%) was prepared at six concentrations: 20, 40, 60, 80, 100, and 200 $\mu\text{g/mL}$ using ethanol as the solvent test extract. The reaction mixture was then allowed to stand for 30 min at room temperature in a dark place. After 30 min, absorbance was measured at 517 nm using a colorimeter. Ascorbic acid was used as a standard. The scavenging activity of DPPH radical (%) was calculated using the following equation: % Inhibition = $[(A_0 - A_1)/A_0] \times 100$ A_0 - absorbance of control A_1 - absorbance of the test sample. All experiments were performed in triplicate.

Total Phenolic Content (TPC): The total phenolic content of ethanolic extract was determined using the Folin-ciocalteu colorimetric method (Panche *et al.*, 2016). The standard solution of gallic acid (5-25 $\mu\text{g/mL}$) was added to 25 mL volumetric flask containing 9 mL of distilled deionized water. A reagent blank was prepared using distilled water instead of a sample. About 1 mL of Folin-Ciocalteu phenol reagent was added to the mixture and shaken well. After 5 min, 10 mL of 7% aqueous sodium carbonate was added to the mixture. The solution was diluted to 25 mL with double distilled water. After incubation for 90 min at room temperature, the absorption against prepared reagent blank was determined at 760 nm using a colorimeter. Quantification was done with respect to the standard gallic acid and expressed as gallic acid equivalents (GAE) in mg per gram of extract. All experiments were performed in triplicate.

Total Flavonoid Content (TFC): Total flavonoids content was determined following a method by Park *et al.* (2008). The standard curve for total flavonoids was made using Quercetin standard solution under the same procedure as earlier described. The total flavonoid was expressed as milligrams of quercetin equivalents per g of dried fraction.

Results

Phytochemical Screening: The ethanolic leaf extract of *Millettia atropurpurea* was subjected to the qualitative chemical analysis for the identification of various plant constituents. The results were given in Table 1. The phytochemical evaluation of ethanolic extract was revealed different phytoconstituents like alkaloids, triterpenoids, tannins, polyphenols, saponins and phenolic compounds were present whereas glycosides were absent.

Table 1. Phytochemical screening of ethanolic leaf extract of *Millettia atropurpurea*.

S.No.	Test	70% ethanolic extract	90% ethanolic extract
1.	Flavonoid	+	+
2.	Alkaloid	+	+
3.	Terpenoid	+	+
4.	Phenol	+	+
5.	Tannins	+	+
6.	Saponins	+	+
7.	Cardiac Glycosides	-	-

+ Presence; - Absence.

FTIR analysis: The ethanolic extract of *Millettia atropurpurea* was subjected to the FTIR spectrum analysis, and data of the FTIR spectrum revealed the presence of various characteristic functional groups in the ethanolic extract of *Millettia atropurpurea* was obtained and depicted in Fig. 1 and 2, which confirmed the presence of functional groups, the frequency ranges, and functional groups obtained from absorption spectra are presented in Table 2 and 3.

The intense bands occurring at 3350.36 cm⁻¹, 3195.21 cm⁻¹, 1613.99 cm⁻¹, 1602.17 cm⁻¹, 1444.07 cm⁻¹, 1442.26 cm⁻¹, 1347.01 cm⁻¹, 1206.48 cm⁻¹, 1039.99 cm⁻¹, and 1031.02 cm⁻¹ corresponding to N-H₂, O-H stretch, asymmetric C-H₂ stretching, C=O, C=C, C-H Benzene, C-O stretch and C-Cl. This confirms the presence of functional groups in *M. atropurpurea* like amines, amides, hydroxyl, carbonyl, Benzene, ether, and Halogen. These are active secondary metabolites of the plant. The functional groups that appeared in FTIR were correlated to a qualitative phytochemical screening of the ethanolic extract of *M. atropurpurea*. FTIR spectrum revealed the presence of various characteristic functional groups in ethanolic root extract of *M. atropurpurea*.

Figure 1. FTIR spectrum of the 70% ethanolic leaf extract of *Millettia atropurpurea*.

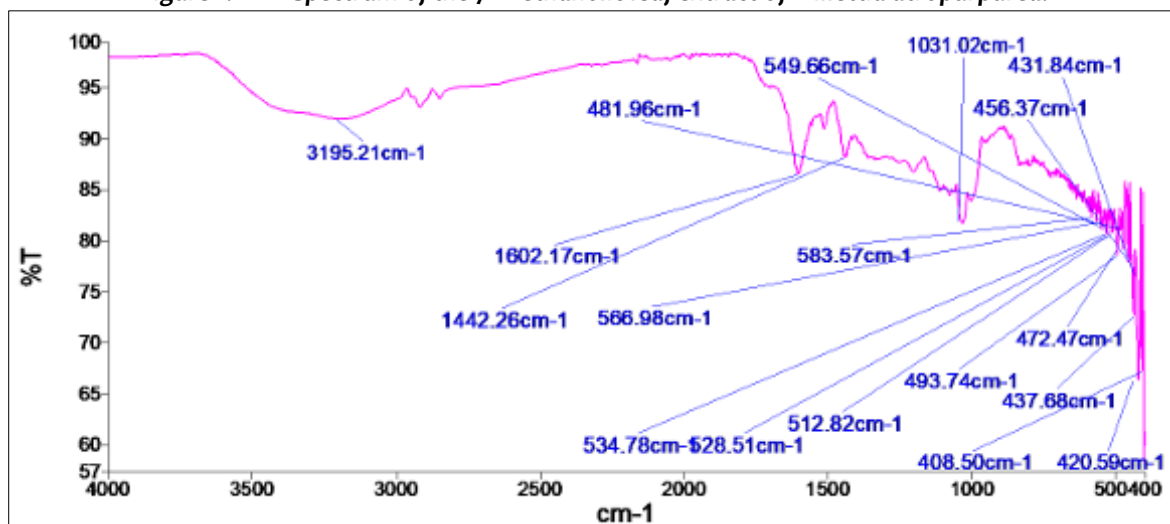


Table 2. Major FTIR absorption peaks and corresponding functional groups identified in 70% ethanolic leaf extract of *M. atropurpurea*.

Peak (cm ⁻¹)	Vibration mode	Functional group
3195.21	O-H stretching	O-H
1602.17	C=C stretching	C=C
1442.26	C-H bending (alkane)	C-H
1031.02	C-F stretching	C-F
2000-1600	Overtone of aromatic rings	Aromatic compound

Figure 2. FTIR spectrum of the 90% ethanolic leaf extract of *Millettia atropurpurea*.

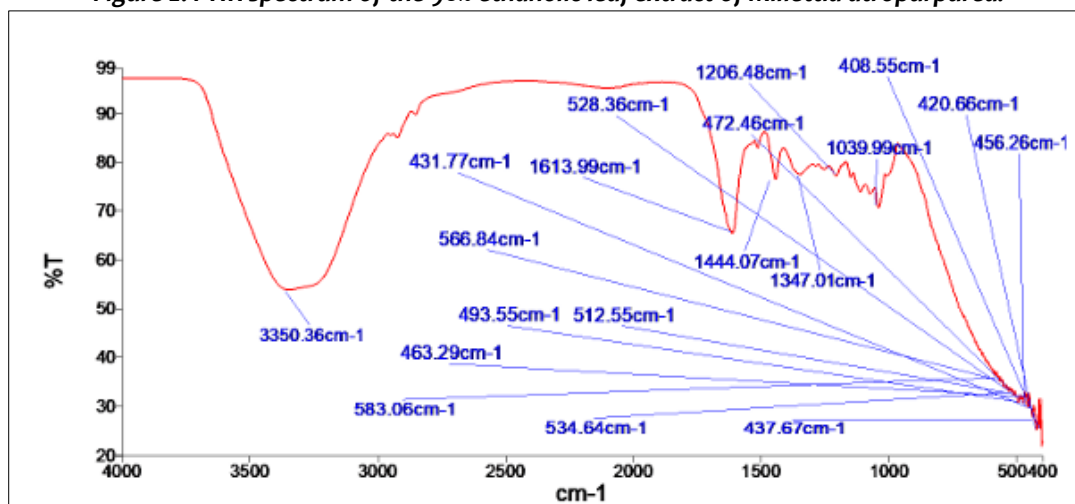


Table 3. Major FTIR absorption peaks and corresponding functional groups identified in 90% ethanolic leaf extract of *M. atropurpurea*.

Peak (cm ⁻¹)	Vibration mode	Functional group
3350.36	O-H stretching	O-H
1613.99	C=C stretching	C=C
1444.07	C-H stretching (sp ³ alkane)	C-H
1347.01	C-O stretching (phenols/alcohols)	C-O
1206.48	C-O-C stretching (ethers)	C-O-C
1039.99	C-O stretching (phenols/alcohols)	C-O

TLC profiling: Results obtained on distances travelled by solvent front and extracts on performing the thin layer chromatography were analyzed and the R_f values were noted. The larger the R_f value, the lower the polarity of natural products or phytochemical. The number of spots and the R_f values clearly indicate that each latex solvent extract with different solvent systems has many phytoconstituents present. Table 4 gives a complete data about the TLC profiling of both the plants leaves solvent extracts.

TLC Analysis of Flavonoids: R_f values obtained from TLC analysis of flavonoids in 70% and 90% ethanolic extracts of *M. atropurpurea* using toluene: ethyl acetate : formic acid (5:4:1) as the mobile phase.

Figure 3. TLC plate observed under UV light and visible light after development.

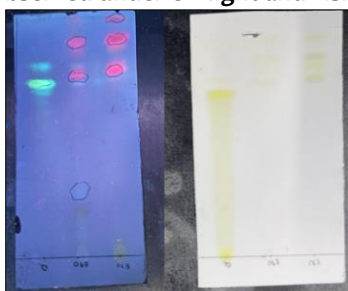


Table 4. R_f values obtained from TLC analysis of flavonoids in 70% and 90% ethanolic extracts of *M. atropurpurea*.

Sample	Distance travelled by spot (cm)	Solvent front (cm)	R _f value
Quercetin	6.8	9.0	0.75
70% extract	6.9	9.0	0.77
90% extract	6.7	9.0	0.74

TLC Analysis of Phenol: R_f values obtained from TLC analysis of phenol in 70% and 90% ethanolic extracts of *Millettia atropurpurea* using chloroform : methanol : formic acid (8:1:1) and visualized after FeCl₃ spraying.

Figure 4. TLC plate observed under UV light and after FeCl₃ spraying.

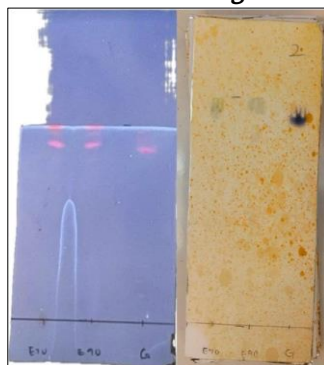


Table 5. Rf values obtained from TLC analysis of phenol in 70% and 90% ethanolic extracts of *M. atropurpurea*.

Sample	Distance travelled by spot (cm)	Solvent front (cm)	Rf value
Gallic Acid	4.50	7.5	0.60
70% extract	4.70	7.5	0.52

Antioxidant activity (DPPH radical scavenging assay): The DPPH assay showed a concentration-dependent increase in radical scavenging activity for the 70% ethanolic extract. The percentage inhibition increased as the extract concentration increased from 20 to 200 µg/mL. The IC₅₀ value obtained from interpolation was 94.7µg/mL, indicating the antioxidant potential of the extract (Table 6). The DPPH assay showed a concentration-dependent increase in radical scavenging activity for the 90% ethanolic extract. The percentage inhibition increased as the extract concentration increased from 20 to 200 µg/mL. The IC₅₀ value obtained from interpolation was 118 µg/mL, indicating the antioxidant potential of the extract (Table 6). The DPPH assay showed a concentration-dependent increase in radical scavenging activity for the Gallic acid. The percentage inhibition increased as the extract concentration increased from 20 to 200 µg/mL. The IC₅₀ value obtained from interpolation was 22.54 µg/mL indicating the antioxidant potential of the extract (Table 7).

Table 6. Mean absorbance of 70% extract and gallic acid (mean ± SD).

Concentrations (µg/mL)	Mean ± SD		
	70% Extract	90% Extract	Gallic acid
20	0.483 ± 0.006	0.542 ± 0.017	0.192 ± 0.007
40	0.428 ± 0.023	0.436 ± 0.026	0.137 ± 0.011
60	0.421 ± 0.020	0.422 ± 0.014	0.078 ± 0.006
80	0.338 ± 0.016	0.372 ± 0.023	0.068 ± 0.004
100	0.255 ± 0.038	0.344 ± 0.023	0.042 ± 0.002
200	0.107 ± 0.003	0.156 ± 0.009	0.026 ± 0.004

Table 7. DPPH radical scavenging activity (% inhibition)

Concentrations (µg/mL)	DPPH radical scavenging activity %		
	70% Extract	90% Extract	Gallic acid
20	12.82	9.97	48.11
40	22.92	27.57	62.97
60	24.01	29.95	78.92
80	38.99	38.21	81.62
100	53.97	42.86	88.65
200	80.75	71.84	92.97

Total Phenolic Content (TPC): Table 8 shows the presence of phenolic contents in all the plant solvent extracts. The 70% ethanolic extract of *M. atropurpurea* possessed the highest phenolic content (23.02) mg GAE/gm while 90% ethanolic extracts of *M. atropurpurea* comprised of high amount of phenolics (18.95) mg GAE/gm.

Table 8. Total phenolic content of *M. atropurpurea* extracts.

Sample	Absorbance (Mean ± SD)	TPC (mg GAE/g extract)
70% ethanol extract	0.874 ± 0.048	23.02
90% ethanol extract	0.706 ± 0.045	18.95

Total Flavonoid Content (TFC): Table 9 shows the presence of maximum flavonoids content were recorded in 90% ethanolic extracts of *Millettia atropurpurea* (31.80) mg QE/gm dry weight. While 70% ethanolic extracts of *M. atropurpurea* comprised of high amount of flavonoids (23.57) mg QE/gm dry weight.

Table 9. Total flavonoid content of *M. atropurpurea* extracts.

Extract	Absorbance (Mean ± SD)	TFC (mg QE/g extract)
70% ethanolic extract	0.310 ± 0.017	23.57
90% ethanolic extract	0.403 ± 0.020	31.80

Discussion

Preliminary testing of the 70% and 90% ethanolic extracts showed the presence of several active plant chemicals. Both extracts had flavonoids, alkaloids, phenols, tannins, and saponins, which means that ethanol is a good solvent for extracting these compounds. These chemicals are often linked to antioxidant and healing effects in plants. Terpenoids were found only in the 70% ethanolic extract, not in the 90% one. This difference might be because the polarity of the solvent affects how well it can dissolve certain compounds. Studies show that moderately polar solvents like 70% ethanol are better at extracting terpenoids than higher concentrations of ethanol. Cardiac glycosides were not found in either extract, which suggests they may not be present in significant amounts in the sample.

The presence of phenolic compounds, flavonoids, and tannins suggests that the plant extract might have antioxidant properties. These initial findings show that the plant may contain secondary metabolites that could be responsible for its biological effects and support the need for more detailed analysis. The FTIR spectra of both the 70% and 90% ethanolic extracts of *M. atropurpurea* showed specific absorption patterns linked to functional groups found in phenolic and flavonoid compounds. Phenolic compounds are secondary metabolites with an aromatic ring and one or more hydroxyl groups, which are typical of both phenols and flavonoids. In both extracts, a broad absorption band around 3195–3350 cm^{-1} indicated the presence of hydroxyl groups, which are important for antioxidant activity. Both extracts also had absorption peaks around 1600–1614 cm^{-1} , which correspond to C=C stretching in aromatic rings, a common feature in phenolic compounds and flavonoids. Additional peaks between 1340–1440 cm^{-1} (C–H bending) and 1000–1200 cm^{-1} (C–O stretching) further support the presence of these compounds, as these functional groups are often found in hydroxylated aromatic compounds and flavonoid structures.

Thin layer chromatography (TLC) was used to check for flavonoids and phenolic compounds in the 70% and 90% ethanolic extracts of *Millettia atropurpurea*. For flavonoids, the R_f value of the standard quercetin was 0.75. The 70% extract had an R_f of 0.77, and the 90% extract had 0.74. These values are close to the standard, suggesting both extracts contain flavonoids. For phenolic compounds, the standard gallic acid had an R_f of 0.60. The 70% extract showed an R_f of 0.52, and the 90% had 0.57. These are relatively close to the standard, showing both extracts have phenolic compounds. When sprayed with ferric chloride, dark spots appeared, confirming the presence of phenolic compounds, as ferric chloride reacts with hydroxyl groups in phenols to form colored complexes. R_f values can vary due to differences in solvent polarity and compound composition, as different solvents can affect how phytochemicals move through the TLC plate. Overall, the TLC results confirm the presence of flavonoids and phenols in the extracts, suggesting they might be responsible for the plant's antioxidant activity.

The DPPH radical scavenging graphs for the 70% and 90% ethanolic extracts of *Millettia atropurpurea* showed a clear pattern where higher concentrations led to greater percentage inhibition. The 70% ethanolic extract had stronger antioxidant activity than the 90% extract. This is shown by the steeper rise in percentage inhibition and the lower IC_{50} value for the 70% extract (94.7 $\mu\text{g/mL}$) compared to the 90% extract (124.6 $\mu\text{g/mL}$). A statistical analysis confirmed that the difference in effectiveness at 200 $\mu\text{g/mL}$ was significant ($p < 0.05$), showing that the polarity of the solvent played a role in how well antioxidant compounds were extracted. However, both extracts had lower antioxidant activity than gallic acid, which is a pure phenolic compound. The gallic acid graph showed a strong concentration-dependent increase in percentage inhibition with a much lower IC_{50} value of 22.54 $\mu\text{g/mL}$. This highlights that pure phenolic compounds tend to have stronger antioxidant effects than mixtures found in crude plant extracts. Numerous studies suggest that ethanol-water mixtures, especially around 70% concentration, are better at extracting phenolic and flavonoid compounds and show stronger antioxidant activity than higher ethanol concentrations or pure solvents (Nurhasanah *et al.*, 2024b; Sun *et al* 2025a). The total phenolic content (TPC) analysis showed that the 70% ethanolic extract had a higher phenolic content (23.02 mg GAE/g extract) compared to the 90% ethanolic extract (18.95 mg GAE/g extract). Statistical analysis using an independent samples t-test showed that the mean absorbance of the 70% extract was significantly higher than that of the 90% extract ($p < 0.05$), indicating that the 70% extract had a much greater phenolic content.

This suggests that 70% ethanol was more effective at extracting phenolic compounds due to its higher polarity compared to 90% ethanol. A similar trend was observed in the DPPH radical scavenging assay. Both extracts showed increasing percentage inhibition with higher concentrations, but the 70% extract had stronger antioxidant activity with a lower IC₅₀ value (94.7 µg/mL) compared to the 90% extract (124.6 µg/mL). The stronger antioxidant activity of the 70% extract matches its higher phenolic content, which indicates that phenolic compounds are a major factor in free radical scavenging. Many studies show a strong positive link between higher total phenolic content in plant extracts and their enhanced antioxidant or free radical scavenging activity (Dayal *et al.*, 2024). This is backed by research showing that polyphenolic compounds, with their various functional groups, are mostly responsible for the strong antioxidant capacity seen in these extracts (Bezghiche *et al.*, 2025).

Phenolic and flavonoid compounds contain hydroxyl groups on aromatic rings, which allow them to act as hydrogen donors and help stabilize free radicals. This is supported by phytochemical screening, FTIR, and TLC analyses, which confirmed the presence of phenols and flavonoids in both extracts. The total flavonoid content (TFC) of the 70% and 90% ethanolic extracts of *M. atropurpurea* was measured using the aluminium chloride colorimetric method with quercetin as the standard. The results showed that the 90% ethanolic extract had a higher flavonoid content (31.80 mg QE/g extract) than the 70% ethanolic extract (23.57 mg QE/g extract). This suggests that the higher ethanol concentration was more effective at extracting flavonoid compounds, which are generally less polar than other phenolic compounds. Flavonoids, as a diverse group of polyphenolic compounds, usually have lower polarity than many other phenolic compounds due to differences in their structure and number of hydroxyl groups (Camargo *et al.*, 2025). This characteristic often leads to better solubility in higher ethanol concentrations or less polar solvents, which helps improve extraction efficiency (Boateng, 2023). The higher flavonoid content in the 90% extract suggests that less polar ethanol concentrations may be better for extracting certain flavonoids. Therefore, both phenolic and flavonoid compounds contribute significantly to the overall antioxidant potential of *Millettia atropurpurea* extracts. These findings align with the TPC and antioxidant results and further support the presence of active phytochemicals in these plant extracts.

Conclusion

This study was conducted to investigate the phytochemical composition, total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activity of *Millettia atropurpurea* leaf extracts prepared using 70% and 90% ethanol. The findings demonstrated that both extracts contained important secondary metabolites, including flavonoids, alkaloids, phenols, tannins, and saponins, while terpenoids were detected only in the 70% extract. These results confirm that *M. atropurpurea* leaves are a rich source of bioactive phytochemicals and support their potential medicinal value.

FTIR analysis further supported the presence of functional groups associated with phenolic and flavonoid compounds in both extracts, indicating that these phytochemicals are major constituents of the plant. Thin layer chromatography (TLC) profiling showed R_f values comparable to gallic acid and quercetin standards, confirming the presence of phenolic and flavonoid compounds. The total phenolic and total flavonoid content analysis revealed that solvent polarity influenced phytochemical extraction efficiency. Antioxidant activity assessed using the DPPH radical scavenging assay showed that both extracts exhibited concentration-dependent antioxidant activity. However, the 70% extract demonstrated stronger activity with a lower IC₅₀ value (94.7 µg/mL) compared to the 90% extract (124.6 µg/mL). Statistical analysis confirmed a significant difference between the extracts ($p < 0.05$), indicating that solvent concentration significantly affects antioxidant potential. Overall, this study provides baseline scientific evidence on the phytochemical profile and antioxidant potential of *Millettia atropurpurea* leaves. The results suggest that the plant is a promising natural source of antioxidant compounds and may have potential applications in pharmaceutical and nutraceutical fields.

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