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# METHANOLIC BARK EXTRACT OF BRIDELIA FERRUGINEA: PHYTOCHEMICALS, ANTIOXIDANT POTENTIALS AND SPECTROSCOPIC CHARACTERIZATION

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#### ABSTRACT

*Bridelia ferruginea* is commonly utilized in African traditional medicine, yet extensive inquiries into its bioactive constituents remain limited. This study assesses the phytochemical composition, antioxidant properties, and spectroscopic characteristics of the methanolic bark extract of *B. ferruginea*. Standard phytochemical screening showed the presence of saponins, phenols, tannins, flavonoids, terpenoids, steroids, and phlobatannins, with phenols and terpenoids occurring in substantial quantities. Antioxidant properties were evaluated using recognized assays, comprising total phenolic and flavonoid content, ferric reducing antioxidant power (FRAP), Fe<sup>2+</sup> chelation, and DPPH radical scavenging activity with ascorbic acid as control. The extract confirmed remarkable antioxidant capacity, with 50 % inhibition (IC) values for FRAP at 0.412 μg/mL, Fe<sup>2+</sup> chelation activity of 7.12 μg/mL, and DPPH scavenging activity of 12.63 μg/mL, demonstrating significant potential in mitigating oxidative stress. FTIR analysis affirmed functional groups, including hydroxyl, carbonyl, ester, and aromatic systems related to bioactive compounds. UV-Vis spectra showed strong absorption around 230 nm, consistent with phenolic and flavonoid constituents. GC-MS analysis identified 24 compounds, comprising myricetin, quercetin, kaempferol, phytol, and zingiberene, several of which are associated with antioxidant, antimicrobial, and anti-inflammatory activities. These results offer scientific support for the plant's traditional uses and point to its capacity as a natural source of antioxidants and medicinal agents.

Keywords: Bridelia Ferruginea, Phytochemicals, Antioxidant Potential, Traditional Medicine

#### INTRODUCTION

Bridelia ferruginea has received extensive focus for its diverse phytochemical composition and potential therapeutic benefits. It is a remarkable species from the Euphorbiaceae family and widely utilized in traditional medicine in the tropical regions of Africa. The extracts from B. ferruginea have been used for the treatment of diseases covering digestive disorders, infections, to inflammatory conditions (Afolayan et al., 2018; Abubakar et al., 2017). Recent research has underlined the phytochemical richness of this species, highlighting its potential for modern pharmacological applications (Adeleye, 2020; Afolayan, 2023). Among various parts, the bark extract of this plant has shown potential for its medicinal properties, notably its anti-inflammatory, antimicrobial, and antioxidant properties (Mehmood and Murtaza, 2018). The bark holds an array of bioactive compounds, including flavonoids, tannins, alkaloids and phenolic acids, which contribute to its therapeutic importance (Yeboah et al., 2022; Engel et al., 2023; Mahomoodally et al.,

The scientific curiosity in the phytochemical properties of plant extracts serves as a critical basis for exploring their therapeutic applications. Understanding such properties is important for establishing the efficiency and safety of herbal preparations (Haile et al., 2020). The extracts from B. ferruginea bark have revealed significant phytochemical diversity. Two primary groups, alkaloids and flavonoids, identified in the plant bark, have been associated with diverse biological activities, including anti-inflammatory and cytoprotective properties (Gheraissa et al., 2023). Another study confirmed that the crude methanolic extract displayed high concentrations of tannins, flavonoids and phenolic compounds, suggesting its potential as a source of natural antioxidants (Adeleye, 2020). Other therapeutic properties, such as the anticancer potential of the phytochemicals from the plant extract, have garnered attention owing to promising in vitro studies uncovering its efficacy against certain cancer cell lines (Omoboyowa et al., 2020).

A major area of interest in recent research focuses on the extract's antioxidant properties, which stem from its broad range of secondary metabolites (Adeleye, 2020). Oxidative stress, resulting from an imbalance between antioxidants and free radicals, has been linked to a variety of diseases, including cancer, cardiovascular disorders and diabetes. Compounds rich in phenolics and flavonoids demonstrate thorough antioxidant activity by scavenging free radicals and reducing lipid peroxidation processes (Deeh et al., 2024; Jeevitha et al., 2021). The antioxidant mechanisms utilized by compounds like flavonoids include the stabilization of free radicals through donating an electron and chelating of transition metal ions (Labe et al., 2020; Ekanem et al., 2024). Additionally, phenolic compounds, which are prevalent in various plant extracts, serve as vital actors in antioxidant activities, strengthening the necessity for thorough phytochemical analyses of herbal preparations.

Furthermore, earlier literature indicates that the aqueous extracts of B. ferruginea exhibit significant antioxidant activities (Abubakar et al., 2017; Afolayan, 2023). Another recent study highlighted that the aqueous extract effectively inhibits the formation of thiobarbituric acid-reactive substances (TBARS) in ex vivo models (Adeleye, 2020). In addition, studies have shown that its methanol extract can substantially inhibit oxidative stress markers in vitro (Purnamasari et al., 2023; Vivek-Ananth et al., 2023). Also, some studies indicated that the extraction method may play a central role in determining the yield and potency of phytochemical compounds. For example, extracts prepared using hydro-ethanolic methods have been shown to present excellent antioxidant activities compared to aqueous extraction methods, illustrating the importance of solvent choice in phytochemical analysis (Omoboyowa et al., 2020; Akinyele and Dada, 2020). Specific assays, such as DPPA and

FRAP, have shown that these extracts exhibit significant free radical scavenging ability, confirming oxidative stress-related diseases (Jeevitha *et al.*, 2021; Behera and Nayak, 2023).

Spectroscopic characterization provides fundamental insights into the chemical makeup of plant extracts. Techniques such spectroscopy, Fourier-transform infrared as UV-Vis spectroscopy (FTIR), and Gas chromatography-mass spectrometry (GC-MS) are used to identify and differentiate the phytochemical components present in the extracts ((Mehmood and Murtaza, 2018; Jeevitha et al., 2021). FTIR spectroscopic analysis is particularly useful for elucidating functional groups and evaluating purity, while GC-MS correctly determine the chemical composition of plant extracts with exceptional specificity and sensitivity (Fagbohun and Bamikole, 2019; Cangeloni et al., 2022). The combination of these approaches not only confirms the presence of significant bioactive constituents but also assists in standardizing the extracts for clinical applications (Afolayan et al., 2018; Yeboah et al., 2022). Also, through these techniques, researchers can gain an understanding of the chemical profile of the extracts, potentially guiding the development of targeted therapies and enhancing our knowledge of their biological activities (Rahman et al., 2022; Ojo et al., 2022).

Finally, the utilization of *B. ferruginea* bark extracts provide avenues not only for novel therapeutic development but also for promoting traditional African medicine on a global scale. While previous studies have explored aqueous extracts, a comprehensive analysis of the methanolic bark extract, correlating its detailed phytochemical profile from GC-MS with its antioxidant capacity through multiple assays, remain limited. It is expected that this research will not only reassert the traditional uses of B. ferruginea but also lay the foundation for further studies designed to exploit its full medicinal potential through modern scientific methods and set the stage for its potential application in complementary and alternative medicine. This study therefore aims to conduct a detailed phytochemical screening, evaluate the in vitro antioxidant potential using multiple standard assays (FRAP, DPPH, Metal chelation), and characterize the functional groups and specific bioactive compounds using FTIR, UV-Vis, and GC-MS spectroscopy.

# MATERIALS AND METHODS

# **Sample Collection and Extract Preparation**

The bark of *B. ferruginea* was collected from a village in Usi-Ekiti, Nigeria, and was identified at the Department of Plant Science and Biotechnology and a voucher specimen was deposited accordingly at the herbarium of the Department of Plant Science and Biotechnology, Federal University Oye-Ekiti, Nigeria. The collected bark was carefully washed to remove sticking debris, cut into smaller pieces, and air-dried in sunlight for seven days. The dried material was ground into powder and afterwards sieved to obtain particle sizes ranging from 300 to 425 μm. Extraction was performed in a Soxhlet apparatus using methanol, according to the procedure described by Kanthal *et al.*, 2014. The obtained methanolic extract was concentrated with a rotary evaporator and further dried at 40 °C to yield the crude extract. The dried extract was stored at 4 °C until required for further analyses

# Phytochemical Analysis of the Extract

The phytochemical screening of the methanolic extract was carried out using standard procedures as described by Gul *et al.*, 2017 and Ezeonu and Ejikeme, 2016. The secondary metabolites explored included flavonoids, phenols, tannins,

steroids, phlobatannins, anthocyanins, saponins, and terpenoids.

#### Test for Flavonoids

The alkaline reagent test was used to detect flavonoids. 2 mL of 2% NaOH solution was mixed with the crude aqueous extract. The progression of an intense yellow coloration, which became colourless upon the addition of two drops of dilute HCl, suggested the presence of flavonoids.

# Test for Phenols

The presence of phenols was determined by mixing 2 mL of 2% FeCl<sub>3</sub> solution with 5 mL of the methanolic extract. The form of a blue-green or black coloration affirmed the presence of phenolic compounds.

#### Test for Tannins

Tannins were tested by adding 0.5 g of the aqueous extract to 10 mL of bromine water. The decolorization of bromine water indicated the presence of tannins.

#### Test for Steroids

5 mL of the aqueous extract was treated with 2 mL each of chloroform and concentrated H<sub>2</sub>SO<sub>4</sub>. Steroids were confirmed by the formation of a red colour at the lower chloroform layer.

# Test for Phlobatannins

5 mL of the extract was added to 1% aqueous HCl, and the mixture was boiled. Phlobatannins were confirmed by the presence of red precipitate in the sample.

# Test for Cyanogenic Glycosides

The sodium picrate (Guignard) test was used to detect the presence of anthocyanins. Filter paper strips soaked in 10% sodium carbonate and 10% picric acid were dried and suspended inside a closed bottle containing the extract, with part of the strip fixed to the lid. The formation of a maroon or brick-red colour indicates the presence of anthocyanins.

### **Test for Saponins**

Aqueous extract was mixed vigorously with 5 mL of distilled water in a test tube. Then a few drops of olive oil were added to the resulting froths, and the mixture was shaken further. The formation of persistent foam suggested the presence of saponins.

#### **Test for Terpenoids**

5 mL of the aqueous extract was added to 2 mL of chloroform, and the mixture was evaporated in a water bath. Afterwards, 3 mL of concentrated  $H_2SO_4$  was added and allowed to boil. The formation of a grey coloration confirmed the presence of terpenoids.

#### **Antioxidant Properties of the Extract**

The antioxidant potential of the methanolic extract of *B. ferruginea* bark was assessed using the following standard assays.

# **Determination of Total Phenolic Content**

The total phenolic content was determined using the method of Singleton *et al.*, 1999. Briefly, 2.0 mL of 7.5% sodium carbonate solution and 2.5 mL of 10% Folin–Ciocalteu reagent were mixed with 0.2 mL of the extract. The reaction mixture was incubated at 45 °C for 45 min, and the absorbance was taken at 700 nm using a spectrophotometer. The standard used for the test was Gallic acid, and results were expressed as gallic acid equivalents (GAE).

#### **Determination of Total Flavonoid Content**

Total flavonoid content was calculated using the colorimetric method of Boa *et al.*, 2005. In brief, 0.3 mL of 5% NaNO<sub>2</sub> was mixed with 0.2 mL of extract and allowed to stay for 5 min. Thereafter, 0.6 mL of 10% AlCl<sub>3</sub> was added to the mixture and allowed to wait for another 6 min before 2.1 mL of distilled water and 2.0 mL of 1 M NaOH were added to the mixture. The absorbance of the mixture was measured at 510 nm compared to a reagent blank, and flavonoid content was expressed as mg of rutin equivalent (RE).

#### **Determination of Ferric Reducing Power**

Ferric reducing antioxidant power was estimated according to the method of Pulido *et al.*, 2006. A mixture of 0.25 mL 200 mM sodium phosphate buffer (pH 6.6), 0.25 mL of 1% potassium ferricyanide [K<sub>3</sub>Fe(CN)<sub>6</sub>] and 0.25 mL of extract was prepared and incubated at 50 °C for 20 min; thereafter, 0.25 mL of 10% trichloroacetic acid was added. The mixture was centrifuged at 200 rpm for 10 min, and 1.0 mL of distilled water and 0.1 mL of 0.1 M FeCl<sub>3</sub> solution were mixed with 1.0 mL of the supernatant. The absorbance was taken at 700 nm

### **Determination of Free Radical Scavenging Activity**

The free radical scavenging activity of the extract was determined utilizing the DPPH assay, as described by Gyamfi *et al.*, 1999. 1.0 mL of 0.4 mM methanolic DPPH solution was mixed with 1.0 mL aliquot of extract, and the mixture was incubated in the dark for 30 mins. The absorbance was taken at 516 nm.

% of inhibition activity =  $[(A_c - A_s)/A_c] \times 100$  where,  $A_c$  is Control reaction absorbance;  $A_s$  is Testing specimen absorbance

# Determination of Fe<sup>2+</sup> Chelating Activity

The Fe<sup>2+</sup> chelating activity was evaluated following the amended method of Minotti and Aust, as reported by Puntel *et al.*, 2005. Briefly, 168 mL of 0.1 M Tris-HCl buffer (pH 7.4) in a reaction mixture was added to 150 mM FeSO<sub>4</sub>. To

the mixture, the extract (0.2 mL) and 218 mL of saline solution were added, and the total volume was brought up to 1 L with distilled water. 13 mL of 1,10-phenanthroline solution was added to the resulting mixture after incubation for 5 min. Afterwards, the absorbance of the mixture was measured at 510 nm.

#### **Characterization of the Extract**

The functional groups present in the extract were determined using Fourier Transform Infrared (FTIR) spectroscopy on a SHIMADZU FTIR-8400S spectrometer, operated within the range of 400-4000 cm $^{-1}$ . The sample was prepared by pelletizing with KBr. The optical properties of the extract were analyzed using a UV-Visible spectrophotometer (U-2010). Before scanning, the absorbance of the solvent used to dissolve the extract was first determined and used as a blank. The Gas Chromatography-Mass Spectrometry (GC-MS) analysis was performed on a Varian 3800/4000 GC system equipped with an Agilent MS capillary column (30 m  $\times$  0.25 mm). Nitrogen was utilized as the carrier gas at a flow rate of 1.0 mL/min. The extract was methylated before injection, and analysis was carried out after the column temperature was stabilized.

# RESULTS AND DISCUSSION

### Phytochemical Analysis

The phytochemical analysis of the methanol extract of *B. ferruginea* bark reveals a complex profile of bioactive compounds that may play critical roles in its therapeutic efficiency. The presence of various phytochemicals such as tannins, steroids, phlobatannins, saponins, flavonoids, and phenols, combined with the absence of anthocyanins, underscores the multifunctional role these metabolites could play in both traditional and contemporary medicine. Given the varying concentrations of these phytochemicals, a thorough analysis of their presence can reveal the extract's potential as a medicinal plant.

Table 1: Phytochemical Composition of the Methanolic Bark Extract of B. Ferruginea

Phytochemical	Present	Slightly present	Heavily present	Absent
Flavonoids	+			
Phenols			+++	
Tannins		++		
Steroids		++		
Phlobatannins	+			
Anthocyanins				-
Terpenoids			+++	
Saponins	+			

Tannins are polyphenolic compounds characterized by their astringent properties, which have been associated with therapeutic benefits in gastrointestinal disorders, including diarrhea. Galalain and Aliyu, 2022 reported that tannin-rich extracts possess significant antidiarrheal activity through their action on the digestive tract. The moderate presence of tannins in the *B. ferruginea* extract is significant, as tannins are widely recognized for their antioxidant potential, which plays a critical role in reducing oxidative stress within the body (Adeleye, 2020; Ojo *et al.*, 2022). Several studies have further shown a definite correlation between tannin content and antioxidant activity, emphasizing their role in neutralizing free radicals and mitigating oxidative damage (Engel *et al.*, 2023; Galalain and Aliyu, 2022). Beyond their antioxidant effects, tannins also exhibit antimicrobial and anti-

inflammatory properties (Fagbohun and Bamikole, 2019). For instance, Afolayan, 2023 reported that tannins can hinder the growth of pathogenic microorganisms, contributing to the antimicrobial activity ascribed to *B. ferruginea* extracts. Additionally, steroids, although present in moderate quantities in the *B. ferruginea* extract, have been involved in various bioactive roles, including anti-inflammatory and immune-boosting properties, as these compounds are commonly associated with such properties in various plants (Ojo *et al.*, 2022). Previous studies support that steroid compounds found in medicinal plants can control biochemical pathways associated with inflammation and immunity, boosting therapeutic uses in traditional medicine (Bakoma *et al.*, 2019; Kuevi *et al.*, 2024). Such properties may contribute to the general health benefits linked with *B. ferruginea*.

Phlobatannins, while slightly present, are linked with diverse biological activities, including antioxidant and antiinflammatory effects, although much is still to be understood about their specific roles within the B. ferruginea context (Bakoma et al., 2019). Their secondary role corresponds closely with the plant's overall profile of health benefits. On the other hand, the data concerning saponins and flavonoids, although in small quantities, further amplify the pharmacological outlook of this plant. Saponins are widely acknowledged for their immunostimulatory and cholesterollowering effects, feasibly supporting cardiovascular health (Yeboah et al., 2022), although their lower concentration in B. ferruginea may limit their direct therapeutic impacts. Concurrently, flavonoids demonstrate antioxidant and antiinflammatory capabilities, which not only relieve oxidative stress but also possess antimicrobial properties that can help tackle various infections (Al-Ansari et al., 2019). Previous investigations into other plants have indicated that increased flavonoid content correlates positively with enhanced antioxidant activity, emphasizing the significance of these compounds in B. ferruginea (Ojo et al., 2022; Kristiani and Kasmiyati, 2022).

The substantial presence of phenols indicates a robust antioxidant capacity, which is central to the therapeutic profile of *B. ferruginea*. Phenolic compounds can scavenge free radicals, protecting cells from oxidative damage, with effects for chronic diseases like diabetes and neurodegenerative conditions. *B. ferruginea* has been identified for its phenolic content and related health benefits, including protective effects on brain and liver tissues in experimental models (Mahomoodally *et al.*, 2021). The high total phenol content reported (9.83 mg/g) in Table 2 is consistent with outcomes regarding their capacity in neutralizing reactive species effectively (Juwitaningsih *et al.*, 2022). Meanwhile, terpenoids, also present in substantial

quantities, have been connected to several pharmacological activities, including analgesic, anti-inflammatory, and antimicrobial properties that contribute to the extract's general therapeutic efficacy (Engel et al., 2023; Omoboyowa et al., 2020; Kuevi et al., 2024). High levels of terpenoids are suggestive of the plant's medicinal qualities, which have drawn attention in various studies for their versatile roles in traditional medicine systems (Silalahi, 2024; Assogba et al., 2021). The synergistic impact of high phenolic and terpenoid content in plants elevates their bioactive potential, making them credible candidates for drug development and nutraceutical applications. The absence of anthocyanins is significant as it illustrates the phytochemical landscape of B. ferruginea. While anthocyanins are usually linked to potent antioxidant activities and the coloring of plant tissues, their absence does not weaken the potential health benefits of this particular species. The lack of anthocyanins, combined with the existence of other potent antioxidant compounds like phenols and flavonoids, implies that B. ferruginea may possess sufficient antioxidant properties through alternative pathways, justifying detailed investigations on its biochemical and pharmacological effects (Bakoma et al., 2019; Kuevi et al., 2024).

#### **Antioxidant Properties**

The antioxidant properties of the methanol extract of *B. ferruginea* bark represent an important research contribution to understanding this plant's phytochemical strength and related health benefits. The results from different parameters measured through different assays, which include Ferric Reducing Antioxidant Power (FRAP), flavonoid content, phenol content, Fe<sup>2+</sup> chelation, and DPPH radical scavenging activity, serve as a critical clue to the antioxidant capacity and highlight the importance of this species in herbal medicine.

Table 2: Antioxidant Assay of the Methanolic Bark Extract of B. Ferruginea

Antioxidant Parameters	Concentration
FRAP (μg/mL)	0.412
Flavonoid (mg/g)	3.72
Phenol (mg/g)	9.83
Fe <sup>2+</sup> Chelation (μg/mL)	7.12
DPPH (μg/mL)	12.63

The FRAP value of 0.412 µg/mL, as shown in Table 2, suggests a considerable capacity of the extract to donate electrons, thus reducing ferric iron (Fe $^{3+}$ ) to ferrous iron (Fe $^{2+}$ ) and further emphasize its capacity to reduce oxidants and provide protection against oxidative damage (Kuevi et al., 2024). Also, the FRAP assay functions under acidic conditions, which provides understanding into the electron transfer capacity of the extract's potent antioxidants (Hsieh and Vani, 2021). Antioxidants neutralize oxidative stress through electron donation, and an elevated FRAP value suggests improved redox potential. Supporting this finding is the study from Afolayan, 2023 that emphasize the effective antioxidant roles of phenolics and flavonoids in *B. ferruginea*. The Fe<sup>2+</sup> chelation value of 7.12  $\mu$ g/mL, as shown in Table 2, offers further evidence of the extract's ability to sequester iron ions, reducing Fenton reactions that can aggravate oxidative stress (Ferrante et al., 2020). Chelation of transition metals such as  $Fe^{2+}$  reduces oxidative damage by deterring free radical generation, which confirms B. ferruginea as a source of bioactive constituents with prospective medicinal applications (Mahomoodally et al., 2021). The ability of B. ferruginea to chelate iron ions gives it leverage in preventing peroxidation, as free iron catalyzes hydroxyl radical formation through the Fenton reaction (Silalahi, 2024). This quality gives it added benefit, which makes it necessary for maintaining redox balance within the cells, thereby highlighting the protective characteristics of the extract against impending oxidative damage (Kamarauskaitė *et al.*, 2021).

The remarkable levels of flavonoids and phenolic compounds identified in the methanol extract, as shown in Table 2, correlate with studies highlighting the protective mechanisms of these phytochemicals against oxidative stress. The calculated flavonoid concentration of 3.72 mg/g in the methanol extract aligns with findings of several studies reaffirming flavonoids as important bioactive compounds capable of scavenging reactive oxygen species (ROS) and mitigating oxidative damage (Engel *et al.*, 2023; Mahomoodally *et al.*, 2021). Flavonoids also exhibit additional health benefits, including antimicrobial properties, which also confirms the therapeutic potential of *B. ferruginea* (Omoboyowa *et al.*, 2020). The total phenolic content measured at 9.83 mg/g further supports the antioxidant potential of the extract. Phenolic compounds are well-

documented for their ability to act as antioxidants by engaging in multiple mechanisms, including the chelation of metal ions, which can catalyze free radical formation. Studies have shown significant radical scavenging abilities of phenolics which are consistent with Adeleye's findings (Adeleye, 2020) on the protective effects of phenol-rich extracts on lipid peroxidation in brain and liver tissues (Kajszczak *et al.*, 2022). The role of phenolics in antioxidant defense mechanisms is supported by their structural capabilities to delocalize electrons, critical in neutralizing free radicals.

Additionally, the DPPH value of 12.63 µg/mL, as shown in Table 2, mirrors the significant radical-scavenging capacity of *B. ferruginea* bark. DPPH assay results confirm the extract's capacity to donate hydrogen atoms to free radicals, thereby reflecting its protective measure against oxidative damage (Kuevi *et al.*, 2024). The effect of such activity agrees with various health benefits, particularly in managing conditions like diabetes and cardiovascular diseases, where oxidative stress is a leading pathology (Song *et al.*, 2020). The evident DPPH scavenging ability of the methanol extract can be attributed to its high contents of flavonoids and phenols, as noted in studies on antioxidant activities of various botanical extracts (Arsul *et al.*, 2022).

#### Characterization

The presence of functional groups in the methanol extract using Fourier Transform Infrared (FTIR) spectroscopy (Fig. 1) confirms the complex nature of its chemical composition. A broad peak around 3394.83 cm<sup>-1</sup> suggests the presence of a hydroxyl (-OH) group, commonly found in phenolic compounds, which play a central role in antioxidant activities by scavenging free radicals (Adetutu et al., 2011). Further analysis revealed a peak at 2931.90 cm<sup>-1</sup>, indicating C-H stretching, suggesting the presence of aliphatic -CH<sub>2</sub>- or -CH<sub>3</sub> groups within the extract, which are commonly found in plant oils and lipid cellular structures (Abubakar et al., 2017). The observed C=C and C=O stretching peaks at around 1635.69 cm-1 further suggest a complex mixture of unsaturated compounds within conjugated carbonyl groups, likely including polyphenolic derivatives. These functionalities are known to boost antioxidant activities through mechanisms such as electron and hydrogen atom transfer (Engel et al., 2023). The peak at 1450.52 cm<sup>-1</sup>, corresponding to the C-H bending, suggests the presence of aliphatic chain bending. Also moving further, the peak at 1149.61 cm<sup>-1</sup> indicates C-O stretching associated with esters or ethers found within the extract, supporting the presence of flavonoids and tannins (Kuevi et al., 2024).

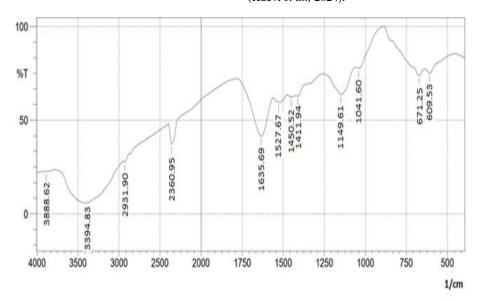


Figure 1: FTIR of The Methanolic Bark Extract of B. Ferruginea

The FTIR identification of a peak at 1041.60 cm<sup>-1</sup>, associated with C-O-C stretching, supports the idea of ether functionalities within the extract. Ethers can influence the solubility and bioavailability of active compounds, thus increasing the extract's antioxidant profiles (Yusufzai *et al.*, 2019). Lower wavenumbers, particularly around 671.25 cm<sup>-1</sup> and 609.53 cm<sup>-1</sup>, indicate out-of-plane bending vibrations of C-H bonds in aromatic systems, suggesting the presence of substituted aromatic rings, which contribute to various biological activities, including antioxidant properties (Mehmood and Murtaza, 2018).

The ultraviolet-visible (UV-Vis) spectrum of the methanol extract from the bark of *B. ferruginea* in Fig. 2 shows an absorption peak at approximately 230 nm, indicating significant electronic transitions within the extract's

molecular structure. This peak suggests the presence of  $\pi \rightarrow \pi^*$  transitions commonly related to conjugated double bonds, such as those found in aromatic compounds especially phenolic compounds (Rajabi *et al.*, 2013). In the context of *B. ferruginea*, such configurations may relate to flavonoids, tannins, and phenolic compounds, all of which are dominant in medicinal plants. The absorption at this wavelength indicates unsaturated functional groups, including carbonyls (C=O) and phenolic groups, both of which are known for their antioxidant properties. Studies on similar phytochemical extracts demonstrate significant absorption in this UV region, supporting the idea that such compounds may contribute to antioxidant activities through their conjugated systems (Dong *et al.*, 2021; Yu *et al.*, 2021).

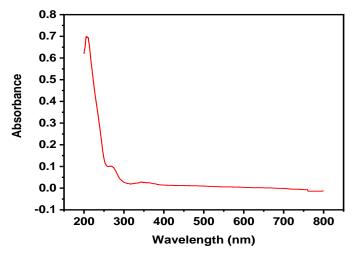


Figure 2: UV-Vis Spectrum of the Methanolic Bark Extract of B. Ferruginea

The GC-MS analysis of the methanolic extract, as shown in Fig. 3, confirms the presence of numerous phytochemicals, ranging from simple aromatics and esters to complex flavonoids, phenols, terpenoids, and fatty acids. A total of 23

compounds were identified based on their retention times and mass spectral matching with the NIST library, and the identified compounds are summarized in Table 3.

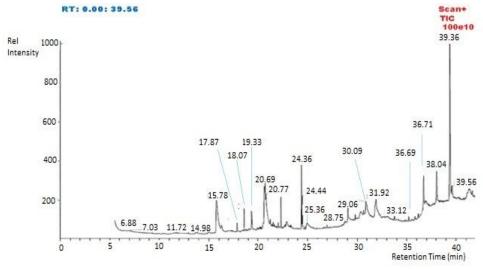


Figure 3: GC-MS Chromatogram of the Methanolic Bark Extract of B. Ferruginea

B. ferruginea bark encompasses various compounds highlighted in the GC-MS analysis, such as myricetin and quercetin, which have recognized antioxidant activities owing to their ability to scavenge free radicals and inhibit lipid peroxidation (Adetutu et al., 2011). Also, compounds like

vanillin may contribute to these activities, with studies showing that vanillin has potential antioxidant effects, thus supporting the pharmacological potential of this plant's extract (Ezike *et al.*, 2011).

Table 3: Identified Compounds in the GC-MS Analysis

RT	Compound Name	Molecular Formula	Mol. Wt.	Peak Area (%)	Chemical Class
6.88	Benzoic acid, methyl ester	C <sub>8</sub> H <sub>8</sub> O <sub>2</sub>	136	1.62	Esters
7.03	1-ethyl-2-methyl benzene	$C_9H_{12}$	120	1.52	Toluene
11.72	Curcumene	$C_{15}H_{22}$	202	1.42	Sesquiterpenoids
14.98	Feruloyl	$C_{11}H_{12}O_3$	192	1.46	Phenols
15.78	3-tert-butyl-4-hydroxyanisole	$C_{11}H_{16}O_2$	180	1.62	Phenols
17.87	Vanillin	$C_8H_8O_3$	152	2.02	Phenols
18.07	Myricetin	$C_{15}H_{10}O_{8}$	318	3.41	Flavonoids
19.33	2,4-di-tert-butylphenol	$C_{14}H_{22}O_2$	206	3.24	Phenols
20.69	Coumarin	$C_8H_8O$	120	5.87	Benzopyrone
20.77	Tyrosol	$C_8H_{10}O_2$	138	5.46	Phenols

RT	Compound Name	Molecular Formula	Mol. Wt.	Peak Area (%)	Chemical Class
24.36	Phytol	C <sub>20</sub> H <sub>40</sub> O <sub>2</sub>	296	8.09	Terpenoids
24.44	Zingiberene	$C_{15}H_{24}$	204	4.86	Sesquiterpenoids
25.36	Kaempferol	$C_{15}H_{10}O_6$	286	2.81	Flavonoids
28.75	Benzoic acid, 3,4-dimethoxy-, methyl ester	$C_{10}H_{12}O_4$	196	1.72	Esters
29.06	Ylangene	$C_{15}H_{24}$	204	3.34	Sesquiterpenoids
29.96	Quercetin	$C_{15}H_{10}O_7$	302	3.21	Flavonoids
30.89	Tetradecanoic acid	$C_{14}H_{28}O_2$	228	4.05	Alkanoic acids
31.92	Azulene	$C_{15}H_{24}$	204	4.15	Terpenoids
33.12	Caryophyllene oxide	$C_{15}H_{24}O$	220	2.43	Sesquiterpenoids
36.71	1,2-benzenedicarboxylic acid	$C_8H_6O_4$	166	6.68	Ester
38.04	n-hexadecanoic acid	$C_{16}H_{32}O_2$	256	6.98	Alkanoic acids
39.36	Trans-Octadec-9-enoic acid	$C_{18}H_{34}O_2$	282	20.23	Alkanoic acids
39.56	Tetratriacontane	$C_{34}H_{70}$	478	5.65	Alkanes
42.08	Pentatriacontane	C <sub>35</sub> H <sub>72</sub>	492	5.06	Alkanes

The identification of multiple beneficial compounds, such as kaempferol, phytol, zingiberene and fatty acids, suggests a multi-targeted effect that might elicit a broad spectrum of biological activities. The various sesquiterpenoids and fatty acids correlate with anti-inflammatory and antimicrobial properties documented in other studies (Oluwagbamila *et al.*, 2023). Compounds like phytol contribute to anti-cancer and antioxidant properties. The combination of these bioactives supports the therapeutic value of the sample. Therefore, *Bridelia ferruginea* holds promise not only for its therapeutic potentials in traditional medicine but also for its biochemical characteristics elucidated through modern analytical methods like GC-MS.

#### CONCLUSION

Bridelia ferruginea exhibits remarkable potential as a source of bioactive compounds with diverse therapeutic effects. Comprehensive analyses of its phytochemical properties, antioxidant capability, and detailed spectroscopic characterization were done to offer valuable insights into its efficacy and applicability in treating oxidative stress-related ailments. The phytochemical profile indicated the moderate presence of tannins and steroids, while phlobatannins, saponins, and flavonoids were present in lower concentrations; phenols and terpenoids were heavily present in the extract. The antioxidant parameters measured underpin the potential therapeutic applications through their radical scavenging abilities. For instance, the FRAP value of 0.412 μg/mL indicates a robust capacity for reducing iron in the presence of antioxidants, while the DPPH scavenging activity, measured at 12.63 µg/mL, suggests potential efficacy of the extract in neutralizing free radicals. The findings of the FTIR characterization reinforce the rich source of phytochemicals present in the extract, and the various functional groups identified correlate with established findings on antioxidant and phytochemical profiles. The UV-Vis spectra showed a strong absorption peak around 230 nm, which was linked to a conjugated system and unsaturated functional groups. The GC-MS profile confirmed the presence of numerous bioactive compounds based on their retention times. Compounds like myricetin, quercetin, transoctadec-9-enoic acid, phytol, and ylangene indicate the extract may possess antioxidant, antimicrobial and antiinflammatory potentials. These findings inferred that the methanol extract exhibited a rich profile of antioxidant and phytochemical properties, which significantly support its traditional applications in medicine. Future studies should focus on isolating the major active compounds, testing the extract's efficacy in in vivo models of oxidative stress, and evaluating its toxic profile.

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