RESEARCH PAPER

Qualitative And Quantitative Phytochemical Profiling Of Anogeissus Latifolia (Roxb. Ex DC.) Wall. Ex Guill. & Perr. Bark With Assessment Of Antibacterial Activity

G. Suguna[1*], Ch. Sowmithri[2], P. Rama Rao[3], S. B. Padal[4]

[1]Research Scholar, Department of Botany, Andhra University, Visakhapatnam - 530003, Andhra Pradesh, India.
Email: suguna.g657@gmail.com

[2]Research Scholar, Department of Botany, Andhra University, Visakhapatnam - 530003, Andhra Pradesh, India.
Email: chalugusowmithri@gmail.com

[3]Lecturer in Botany, Government College, Thogaram - 532484, Srikakulam, Andhra Pradesh, India.
Email: paidiramarao27@gmail.com

[4]Professor, Department of Botany, Andhra University, Visakhapatnam - 530003, Andhra Pradesh, India.
Email: sbpadal08@gmail.com

Corresponding Author: G. Suguna, Research Scholar, Department of Botany, Andhra University, Visakhapatnam - 530003, Andhra Pradesh, India. Email: suguna.g657@gmail.com; Phone: 9703894936

Abstract:

The present study focuses on the comprehensive phytochemical profiling and antibacterial evaluation of *Anogeissus latifolia* bark extracts using various solvents. Qualitative and quantitative analyses were conducted to determine the presence and concentration of major bioactive constituents contributing to the plant's ethnomedicinal significance. Preliminary phytochemical screening of bark extracts in acetone, benzene, chloroform, ether, carbinol, and water confirmed the presence of phenolic compounds, flavonoids, terpenoids, steroids, tannins, glycosides, proteins, and quinones. Methanol (carbinol) and aqueous extracts exhibited strong extraction capabilities for phenolics and flavonoids, whereas chloroform and methanol preferentially extracted terpenoids and steroids. These variations highlight the solvent- dependent distribution of phytochemicals influencing the biological properties of the extracts. Quantitative estimation revealed that methanol extract possessed the highest total phenolic content (326.8 ± 22.6 mg GAE/g), total flavonoid content (135.4 ± 10.5 µg quercetin/mg), and total tannin content (95.4 ± 10.4 mg TAE/g), followed by chloroform, water, and acetone extracts. The abundance of phenolics, flavonoids, and tannins underscores the potential of *A. latifolia* as a rich source of antioxidant and antimicrobial compounds. These phytochemicals are known to contribute to various pharmacological effects including anti-inflammatory, wound-healing, and antibacterial activities, thereby supporting the traditional use of the plant in folk medicine. The antibacterial potential of the bark extracts was assessed against *Clostridium perfringens*, *Enterococcus faecalis*, *Pseudomonas aeruginosa*, and *Salmonella typhimurium* using the agar well diffusion method. Among the tested extracts, the carbinol extract exhibited the most pronounced antibacterial activity, with inhibition zones of 22.3 ± 0.36 mm and 21.4 ± 0.45 mm against *P. aeruginosa* and *C. perfringens*, respectively, comparable to the standard antibiotic chloramphenicol. Water and ether extracts showed moderate to low inhibitory effects. The results clearly demonstrate that solvent polarity significantly influences the extraction efficiency and antibacterial efficacy of *A. latifolia* bark. Overall, the study establishes a strong correlation between phytochemical richness and antibacterial potential, highlighting *Anogeissus latifolia* bark, particularly its methanolic extract, as a promising source of natural bioactive compounds for the development of plant- based antimicrobial agents.

Keywords: *Anogeissus latifolia*, phytochemical profiling, antibacterial activity, phenolic content, flavonoids, ethnomedicine.

How to cite this article: Suguna G, Sowmithri Ch, Rao PR, Padal SB. Qualitative and Quantitative Phytochemical Profiling of Anogeissus latifolia (Roxb. ex DC.) Wall. ex Guill. & Perr. Bark with Assessment of Antibacterial Activity. Int J Drug Deliv Technol. 2026;16(12s): 11-19. DOI: 10.25258/ijddt.16.12s.3

Introduction

Medicinal plants represent a valuable source of therapeutic compounds and continue to play a significant role in traditional and modern healthcare systems. Approximately 80% of the global population relies on herbal medicines for primary healthcare needs due to their accessibility, affordability, and minimal side effects (WHO, 2019). The growing resistance of pathogenic microorganisms to conventional antibiotics has intensified the search for novel antimicrobial agents from plant-based sources (Abreu et al., 2020). Phytochemicals such as phenolics, flavonoids, tannins, alkaloids, and terpenoids are among the major classes of bioactive secondary metabolites responsible for a wide range of pharmacological activities, including antibacterial, antifungal, antioxidant, and anti- inflammatory effects (Panghal et al., 2022; Olagbemide et al., 2021).

Anogeissus latifolia (Roxb. ex DC.) Wall. ex Guill. & Perr., belonging to the family Combretaceae, is a deciduous tree commonly found in dry deciduous forests across India, Sri Lanka, Myanmar, and Thailand. It is locally known as "Axlewood" or "Dhava" and holds a prominent place in Indian traditional medicine systems such as Ayurveda and Siddha. Different parts of the plant, including the bark, leaves, and gum, have been used traditionally for treating diarrhea, dysentery, skin infections, wounds, and inflammation (Jain et al., 2018; Lal et al., 2022). The bark of *A. latifolia* is particularly valued for its high tannin content and strong astringent properties, which have been utilized in tanning industries and in folk remedies for microbial infections and wound healing (Kumar et al., 2021).

Phytochemical investigations on *A. latifolia* and related species such as *Anogeissus acuminata* have revealed the presence of diverse bioactive compounds, including phenolic acids, ellagic acid, flavonoids, steroids, glycosides, and terpenoids (Garba et al., 2025; Mishra et al., 2023). These compounds are known to contribute to antimicrobial and antioxidant activities, which justify the ethnomedicinal uses of the plant. For instance, phenolic and flavonoid constituents exhibit free radical scavenging abilities, while tannins and terpenoids are implicated in antibacterial and anti-inflammatory effects (Stray, 1998; Pandey et al., 2022). Despite the extensive traditional use of *A. latifolia*, systematic scientific validation of its phytochemical profile and antibacterial properties remains limited. Comprehensive studies that combine both qualitative and quantitative screening with bioactivity evaluation are crucial to establish its pharmacological relevance and identify potential therapeutic leads.

Solvent extraction plays a critical role in determining the phytochemical composition and biological activity of plant extracts. Solvents with different polarities extract distinct classes of metabolites, thereby influencing the yield and efficacy of the bioactive compounds (Mishra et al., 2023). Methanol and water are often reported as the most effective solvents for extracting phenolics and flavonoids, which are associated with antimicrobial and antioxidant activities (Lal et al., 2022). Therefore, evaluating the solvent-dependent variation in phytochemical content and antibacterial potential provides insight into optimizing extraction processes for pharmaceutical applications.

Antibacterial resistance poses a major global health concern, making the identification of new natural antibacterial agents an urgent research priority (WHO, 2020; Bassetti et al., 2023). Plant-derived phytochemicals can act as alternative or complementary agents to conventional antibiotics by targeting bacterial cell membranes, enzymes, and genetic materials, thereby disrupting pathogen growth and survival (Abreu et al., 2020). In this context, exploring the antibacterial efficacy of *A. latifolia* bark extracts against clinically relevant bacterial strains such as *Pseudomonas aeruginosa*, *Clostridium perfringens*, *Enterococcus faecalis*, and *Salmonella typhimurium* offers valuable insights into its therapeutic potential. The present study aims to perform a comprehensive phytochemical investigation and antibacterial evaluation of *Anogeissus latifolia* bark extracts obtained using different solvents.

Materials and Methods:

Plant Collection

The plant material of *Anogeissus latifolia* Roxb. was collected from the Paderu division in Andhra Pradesh, India, during October 2024. The botanical identification and authentication of the specimen were carried out by Prof. Dr. S. B. Padal, Taxonomist, Department of Botany, Andhra University, Visakhapatnam. The authenticated herbarium specimen has been deposited in the Department of Botany, Andhra University.

Preparation of plant extract

Anogeissus latifolia stem bark was gathered, cleaned properly to get rid of dirt and contaminants, and then allowed to dry in the shade at room temperature to retain its phytochemical components. A motor and pastel were used to grind the bark into a fine powder once it had completely dried. About 250 g of the air-dried bark powder was put into a 1-liter conical flask that had been cleaned and dried in preparation for extraction. This was followed by the independent addition of 500 mL of solvents in several sets: distilled water, methanol, and hydroalcoholic combination. To enable the effective extraction of bioactive chemicals, the flasks were set on a rocker shaker and constantly shaken at room temperature for three days in a row. After maceration, the extracts were filtered through Whatman No. 1 filter paper. The resulting filtrates were then concentrated using a

rotary evaporator at lower pressure to produce crude extracts, which were subsequently stored for additional examination.

Qualitative Phytochemical Analysis of *Anogeissus latifolia* Bark Extracts

Using standard recognized techniques, the presence of bioactive secondary metabolites was detected through qualitative phytochemical screening of Anogeissus latifolia stem bark extracts made in a variety of solvents, including acetone, benzene, chloroform, ether, carbinol (methanol), and water.

Preparation of Extracts

Finely powdered bark samples were separately extracted with acetone, benzene, chloroform, ether, carbinol, and distilled water by cold maceration. Approximately 10 g of powdered bark was soaked in 100 mL of each solvent for 72 hours at room temperature with intermittent shaking. The extracts were filtered through Whatman No.1 filter paper and concentrated under reduced pressure using a rotary evaporator at 40–45 °C.

Phytochemical Screening Tests

The following chemical tests were conducted on each concentrated extract to qualitatively detect the presence of major phytochemical groups:

Test for Alkaloids

Mayer's Test: To 1 mL of extract, add a few drops of Mayer's reagent (potassium mercuric iodide). Formation of a cream or white precipitate indicates presence of alkaloids.

Wagner's Test: To 1 mL of extract, add a few drops of Wagner's reagent (iodine in potassium iodide). A reddish-brown precipitate indicates alkaloids.

Dragendorff's Test: Add a few drops of Dragendorff's reagent (potassium bismuth iodide) to 1 mL of extract. Formation of an orange or reddish-brown precipitate confirms alkaloids.

Test for Flavonoids

Alkaline Reagent Test: Add 2 mL of 2% sodium hydroxide solution to 1 mL of extract. A yellow coloration develops, which disappears on addition of few drops of dilute acid, indicating flavonoids.

Test for Tannins

Add 2 mL of 5% ferric chloride solution to 1 mL of extract. Appearance of blue-black or greenish coloration indicates tannins.

Test for Saponins

Dilute 1 mL of extract with 5 mL of distilled water and shake vigorously. Persistent froth formation lasting for at least 10 minutes confirms saponins.

Test for Steroids and Terpenoids

Liebermann-Burchard Test: To 1 mL extract, add 2 mL of acetic anhydride followed by 2 mL of concentrated sulfuric acid slowly down the side of the test tube. Formation of a blue or green ring at the junction indicates steroids; a reddish-brown coloration indicates terpenoids.

Test for Glycosides

Keller-Killiani Test: Add 1 mL of glacial acetic acid containing a trace of ferric chloride to 1 mL of extract followed by 1 mL of concentrated sulfuric acid. A reddish-brown ring at the interface signifies glycosides.

Test for Carbohydrates

Molisch's Test: Add 2 drops of Molisch's reagent (α-naphthol in ethanol) to 1 mL of extract and carefully add 1 mL of concentrated sulfuric acid along the side of the test tube. A violet or purple ring at the interface indicates carbohydrates.

Test for Phenols

Add 2 mL of 5% ferric chloride solution to 1 mL of extract. Formation of blue, green, or black coloration indicates phenols.

Test for Proteins and Amino Acids

Biuret Test: Add 1 mL of 10% sodium hydroxide and 2-3 drops of 0.5% copper sulfate to 1 mL of extract. Formation of violet or purple color indicates proteins.

Ninhydrin Test: Mix 1 mL extract with 1 mL of 0.2% ninhydrin solution and heat gently. Development of purple or blue color indicates amino acids.

Quantitative Estimation *Anogeissus latifolia* Bark Extracts Preparation of Extracts

The powdered stem bark of *Anogeissus latifolia* was extracted with suitable solvents (e.g., methanol or hydroalcoholic solvent) by cold maceration or Soxhlet extraction. The extracts were filtered and concentrated under reduced pressure. The dried crude extracts were stored at 4 °C until further analysis.

Quantitative Estimation of Total Phenolic Content (TPC)

Using standardized spectrophotometric techniques, the total phenolic, flavonoid, and tannin contents in the bark extracts of *Anogeissus latifolia* were quantitatively estimated. The Folin-Ciocalteu reagent test, which involves treating 0.5 mL of the extract with diluted Folin- Ciocalteu reagent and then sodium carbonate solution, was used to measure total phenolics. The absorbance at 765 nm was measured after the reaction mixture was allowed to sit at room temperature for 30 minutes in the dark. The results were reported as milligrams of gallic acid equivalents per gram of extract (mg GAE/g), with gallic acid serving as the standard (Elvino Nortjie *et al.,* 2022).

Quantitative Estimation of Total Flavonoid Content (TFC)

Total flavonoid content was determined using the aluminum chloride colorimetric method. The extract (0.5 mL) was mixed with methanol, aluminum chloride, sodium acetate,

\

and water, then incubated at room temperature for 30 minutes. Absorbance was measured at 415 nm against a standard curve prepared with quercetin, with results expressed as milligrams of quercetin equivalents per gram of extract (mg QE/g).

Quantitative Estimation of Total Tannin Content (TTC)

For total tannins, the Folin-Denis method was used where the extract was mixed with Folin-Denis reagent and sodium carbonate solution, then incubated for 30 minutes. Absorbance was read at 760 nm, with tannic acid serving as the calibration standard. Tannin content was calculated as milligrams of tannic acid equivalents per gram of extract (mg TAE/g).

Spectrophotometric measurements for all assays were performed using a UV-Vis spectrophotometer, and all determinations were carried out in triplicate for statistical reliability. These quantitative assays provide critical insight into the bioactive composition of Anogeissus latifolia bark relevant to its traditional medicinal properties (Singleton VL, Rossi JA, 1965; Chang et al., 2002; Harborne JB, 1998)

Antibacterial activity of bark extracts of *Anogeissus latifolia*

The antibacterial activity of *Anogeissus latifolia* plant extracts prepared using carbinol (methanol), ether, and water as solvents was evaluated by the agar well diffusion method. Pure bacterial cultures of *Clostridium perfringens* (MTCC-13124), *Enterococcus faecalis* (MTCC- 6845), *Pseudomonas aeruginosa* (MTCC-6363), and *Salmonella typhimurium* (MTCC-443) were procured from the Microbial Type Culture Collection (MTCC) and Gene Bank, Institute of Microbial Technology (IMTECH), Chandigarh, India. The cultures were revived and maintained on nutrient agar slants and preserved at -20ºC with 15% (v/v) glycerol for long- term storage.

Nutrient agar medium was prepared by dissolving the required quantity of HiMedia nutrient agar powder in distilled water and sterilized by autoclaving at 121ºC under 15 lbs pressure for 15 minutes. The sterilized molten agar was poured aseptically into Petri plates and allowed to solidify. The bacterial inocula were uniformly spread over the agar surface using a sterile glass spreader. Wells of equal diameter were bored into the agar medium using a sterile cork borer at equidistant positions. The *Anogeissus latifolia* extracts prepared in carbinol, ether, and water were concentrated to a stock solution of 100 mg/mL. Different concentrations (40 µg/mL, 60 µg/mL, and 80 µg/mL) of these extracts were loaded into the wells. Chloramphenicol was used as a positive control to compare antibacterial efficacy. The plates were incubated at 37ºC for 24 hours. Afterwards, the zones of inhibition around each well were measured in millimeters. Comparative analyses of the antibacterial activity of carbinol, ether, and water extracts were performed based on the diameter of these zones, with experiments carried out in triplicate to ensure reproducibility.

Results and Discussion

Qualitative screening of bark extract of *A. latifolia*

The phytochemical screening of *Anogeissus latifolia* bark extract reveals the presence of several bioactive compounds, including phenolic compounds, flavonoids, terpenoids, steroids, tannins, glycosides, proteins, and quinones. Different solvents, such as acetone, benzene, chloroform, ether, methanol, and water, each extract unique chemical constituents affecting their pharmacological relevance (Lal et al., 2022).

Both phenolic compounds and flavonoids were present in most solvent extracts, with methanol and water showing strong extraction capabilities for these chemicals. These compounds are known for their antioxidant and antimicrobial activities, supporting the use of

A. acuminata in traditional medicine for wound healing and infection management. The high levels of tannins (13–24%) and flavonoids (14–57%) in bark and leaf extracts reflect the plant's therapeutic potential and ability to scavenge free radicals. Terpenoids were selectively present, primarily in chloroform and methanol extracts, suggesting a higher affinity for non-polar solvents. Steroids and tannins, however, were widely extracted with acetone, benzene, chloroform, ether, and methanol, but less so in water. These compounds contribute to anti-

inflammatory properties and are implicated in the bioactivity against bacterial pathogens like

E. coli and Shigella, affirming the bark's ethnomedicinal applications.

Glycosides were observed mostly in acetone, ether, and methanol, aligning with their moderate polarity. Proteins and quinones also showed solvent-dependent variability, generally absent in benzene and water extracts. Quinones are linked to antimicrobial and antioxidant activities.

Table.1 Phytochemical screening of *Anogeissus latifolia Bark extract*

S. No	Chemical components	Acetone	Benzene	Chloroform	Ether	Carbinol	Water
1	Phenolic compounds	+	-	+	+	+	+
2	Flavonoids	+	+	+	+	+	-
3	Terpenoid	-	-	-	-	+	-

\

	s						
5	**Steroids**	+	+	+	+	+	-
6	**Tannins**	+	+	+	+	+	+
7	**Glycosides**	+	-	-	+	+	-
8	**Proteins**	+	-	+	+	+	-
9	**Quinones**	+	-	+	+	+	-

"+" Present, "-" Absent

The qualitative phytochemical screening of the bark extracts of *A. latifolia* and *A. acuminata* confirmed the presence of various bioactive compounds, including alkaloids, flavonoids, phenols, proteins, saponins, steroids, tannins, and triterpenoids. Alkaloids, in particular, hold significant medicinal value and are widely recognized for their therapeutic properties. Pure isolated alkaloids and their synthetic derivatives serve as essential medicinal agents, demonstrating analgesic, antispasmodic, and antibacterial effects (Stray, 1998). These compounds exhibit notable physiological activity when administered to animals, highlighting their potential applications in pharmacology. This diverse phytochemical composition underscores the medicinal importance of *A. latifolia* and *A. acuminata* and its potential for developing health-promoting and therapeutic products. These phytochemicals contribute significantly to the therapeutic applications of the plants, with tannins and saponins showing the highest and lowest concentration respectively in aqueous extracts. Such findings support the ethnomedicinal use and ongoing development of these plants in herbal formulations and pharmacological research (Garba et al., 2025).

Quantitative screening of bark extract of *A. latifolia*

Total phenolic contents

The quantitative phytochemical screening of *Anogeissus latifolia* bark extracts for total phenolic content (TPC) demonstrates significant solvent-dependent variations, revealing the critical role of extraction methods in maximizing the yield of antioxidant compounds. Phenolic content, measured as mg gallic acid equivalents (GAE) per gram of dry extract, was highest in methanol extracts (326.8 ± 22.6), followed by chloroform (265.4 ± 17.6), water (216.3 ± 14.8), and acetone (174.3 ± 12.5) (Table.2 and Fig.1), aligning with recent studies confirming the strong antioxidant potential of methanol-extracted bark.

Phenolic compounds exert diverse physiological effects**:** anti-allergenic, anti- atherogenic, anti-inflammatory, antimicrobial, antioxidant, anti-thrombotic, cardioprotective, and vasodilatory actions (Benavente-Garcia, 2000; Gupta *et al.,* 2009). Their antioxidant capability largely stems from their redox properties, including free radical scavenging and metal chelation, which protect biological systems from oxidative stress and ROS-induced damage (Deeparani *et al.,* 2025). Recent 2025 research has highlighted the presence of major phenolics such as gallic acid, corilagin, ellagic acid, and various glycoside flavonoids in A. latifolia bark, which contribute directly to its functional food and pharmaceutical relevance. Gum ghatti, a product of A. latifolia, also contains notable monosaccharide and protein content, adding to the plant's health-promoting properties ((Garba *et al.,* 2025).

Table-2 Total Phenolic content (TPC) of the prepared bark extract of *A. latifolia*

S. No	Name of the plant extract	TPC (µg eq. of Gallic Acids/mg of extract) (Mean ± SD)
1	**Acetone**	**174.3 ± 12.5**

\

2	Chloroform	265.4 ± 17.6
3	Methanol	326.8 ± 22.6
4	Water	216.3 ± 14.8

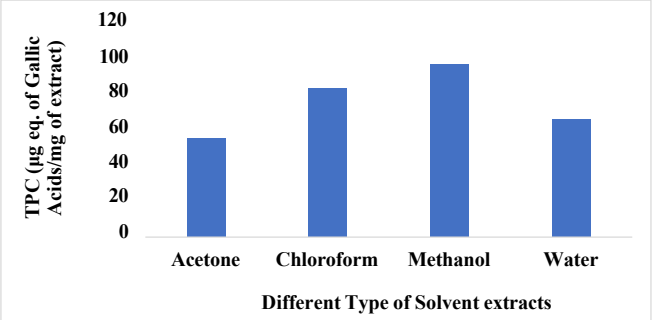

Fig. 1 Phenolic content (TPC) of the prepared bark extract of *A. latifolia*

Total Flavonoid Contents

Quantitative analysis of total flavonoid content (TFC) in the bark extract of *Anogeissus latifolia* highlights significant solvent-dependent differences, with methanol proving to be the most efficient solvent (135.4 ± 10.5 µg quercetin/mg extract), followed by chloroform (122.3 ± 9.8), water (90.2 ± 8.6), and acetone (85.7 ± 6.8) (Table.3 and Fig.2). Flavonoids, the dominant polyphenolics in these extracts, were confirmed through advanced chromatographic techniques, with the active methanol fraction containing orientin, vitexin, and isovitexin, recognized for potent biological activities. This extraction trend mirrors findings from similar research, where polarity plays a critical role in solvent efficiency for flavonoid isolation

Flavonoids possess anti-inflammatory, hepatoprotective, anti-ulcer, antiviral, and anticancer properties, underpinning their wide dietary and pharmaceutical significance. Recent studies have shown that *A. latifolia* extracts, especially those derived from methanol, exhibit notable anti-inflammatory and analgesic effects in vivo, linked to their high flavonoid content. The differential extraction efficiency underscores the practical importance of choosing appropriate solvents to maximize both yield and bioactivity, essential for pharmaceutical and nutraceutical applications.

Table-3 Total Flavonoid content (TFC) of the prepared bark extract of *A. latifolia*

S. No	Name of the plant extract	TFC (µg eq. of Quercetin/mg of extract) (Mean ± SD)
1	Acetone	85.7 ± 6.8
2	Chloroform	122.3 ± 9.8
3	Methanol	135.4 ± 10.5
4	Water	90.2 ± 8.6

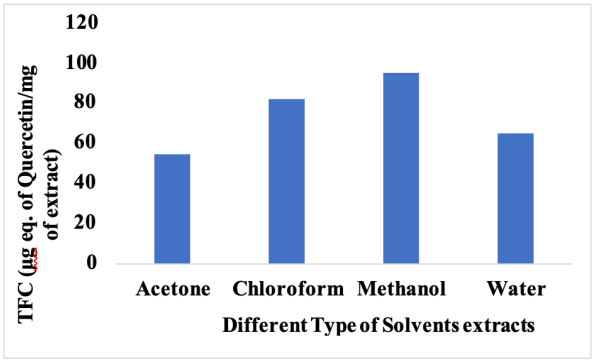

Fig. 2 **Flavonoid content (TFC) of the prepared bark extract of *A. latifolia***

Total Tannin Content

The total tannin content (TTC) of the bark extracts of *Anogeissus latifolia* was quantitatively analyzed and reflected notable variations based on the extraction solvent, as presented in Table 4. The results, expressed in mg tannic acid equivalents per gram of dry extract, indicated that methanol was the most efficient solvent, yielding the highest tannin concentration (95.4 ± 10.4), followed by chloroform (82.3 ± 8.6), water (65.2 ± 9.6), and acetone (54.7 ± 6.9). Tannins are well-recognized polyphenolic compounds noted for their broad spectrum of physiological activities, including anti-irritant, anti-secretolytic, anti- inflammatory, antimicrobial, and anti-parasitic effects. These bioactive compounds have attracted considerable focus due to their therapeutic potential in phytomedicine. Traditionally, tannin-rich plants such as *A. latifolia* have been widely employed in the management of non- specific ailments such as gastrointestinal disorders, skin irritations, and microbial infections. Their astringent nature facilitates reduction of inflammation and tissue irritation while their antimicrobial properties effectively combat pathogenic microbes. Additionally, tannins regulate excessive secretions supporting homeostatic balance in biological systems.

Table-4 Total tannin content of the prepared bark extract of *A. latifolia*

S. No	Name of the plant extract	Tannin (µg eq. of tannic acid/mg of extract) (Mean ± SD)
1	Acetone	54.7 ± 6.9
2	Chloroform	82.3 ± 8.6
3	Methanol	95.4 ± 10.4
4	Water	65.2 ± 9.6

\

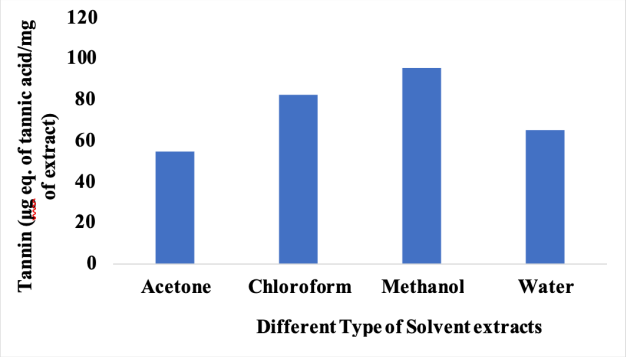

Fig.3 Tannin content (TNC) of the prepared bark extract of *A. latifolia*

Antibacterial activity of bark extract of *Anogeissus latifolia*

The antibacterial activity of the bark extracts of *Anogeissus latifolia* was assessed using carbinol, ether, and water as extraction solvents against four selected pathogenic bacterial strains (*Clostridium perfringens, Enterococcus faecalis, Pseudomonas aeruginosa,* and *Salmonella typhimurium*). The zone of inhibition values obtained at different concentrations (40, 60, and 80 µg/mL) are presented in Tables 5–7. Among the tested extracts, the carbinol extract exhibited the most pronounced antibacterial activity across all test organisms. At 80 µg/mL, the maximum inhibition zones were recorded against *Pseudomonas aeruginosa* (22.3 ± 0.36 mm) and *Clostridium perfringens* (21.4 ± 0.45 mm), which were comparable to the standard drug chloramphenicol (25.2 ± 0.15 mm and 23.1 ± 0.25 mm, respectively). Even at lower concentrations (40 and 60 µg/mL), the carbinol extract produced moderate activity against all tested bacteria, suggesting that carbinol efficiently extracted bioactive compounds with broad-spectrum antibacterial potential.

The ether extract, in contrast, showed comparatively weaker activity. No inhibition zones were observed against *Clostridium perfringens* and *Salmonella typhimurium* at all tested concentrations. However, appreciable inhibition was noted against *Enterococcus faecalis* (18.3 ± 0.26 mm) and *Pseudomonas aeruginosa* (17.3 ± 0.36 mm) at 80 µg/mL. The limited antibacterial activity of the ether extract may be attributed to the solvent's non-polar nature, which might not have extracted sufficient quantities of polar or semi-polar phytoconstituents responsible for strong antibacterial activity.

The water extract exhibited selective antibacterial activity, particularly against *Enterococcus faecalis* and *Pseudomonas aeruginosa*. At 80 µg/mL, the inhibition zones reached 12.3 ± 0.18 mm and 12.2 ± 0.26 mm, respectively, which were lower than those observed for the carbinol extract but significant compared to the ether extract. Interestingly, *Salmonella typhimurium* was moderately inhibited (14.2 ± 0.16 mm at 80 µg/mL), while *Clostridium perfringens* showed no sensitivity to the water extract. These differences suggest that the water extract contains some polar antibacterial compounds, but possibly in lower concentrations or with weaker potency compared to those extracted in carbinol.

When comparing across extracts, a clear trend emerges: carbinol > water > ether in terms of overall antibacterial efficacy. The carbinol extract consistently demonstrated higher inhibition zones across all tested organisms and at all concentrations, indicating its superior ability to extract phytochemicals with potent antibacterial activity. The water extract exhibited moderate and selective activity, while the ether extract showed limited effectiveness, particularly failing against *Clostridium perfringens* and *Salmonella typhimurium*.

The results collectively highlight that the solvent system plays a crucial role in determining the antibacterial potential of *A. latifolia* extracts. Carbinol, being a polar protic solvent, may facilitate the extraction of a broader range of secondary metabolites such as tannins, flavonoids, and phenolic compounds, which are widely reported for their antibacterial properties. Ether, being non-polar, might have primarily extracted non-polar phytoconstituents with relatively weak antibacterial effects, whereas water extracted some polar compounds but possibly with lower efficiency than carbinol.

Overall, the findings demonstrate that the carbinol extract of *Anogeissus latifolia* bark is the most effective against the tested bacterial strains, showing broad-spectrum and dose-dependent activity that, in some cases, approached the efficacy of chloramphenicol. These observations support the potential of *A. latifolia* bark, particularly its carbinol-soluble components, as a promising source of antibacterial agents.

Table.5 Antibacterial activity of carbinol extract of *Anogeissus latifolia* against selected bacteria

S.No	Test Organism	Zone of inhibition (mm)			
		Carbinol extract of *Anogeissus latifolia* (µg/mL)			
		40	60	80	Standard (Chloramphenicol) 30µg/mL
1	*Clostridium perfringens* (MTCC-13124)	15.2±0.26	17.2±0.45	21.4±0.45	23.1±0.25
2	*Enterococcus faecalis* (MTCC-6845)	10.2±0.16	14.2±0.52	18.3±0.26	21.2±0.25

\

3	*Pseudomonas aeruginosa* (MTCC-6363)	-	7.2±0.52	22.3±0.36	25.2±0.15
4	*Salmonella typhimurium* (MTCC-443)	12.5±0.16	16.2±0.22	18.2±0.46	20.2±0.15

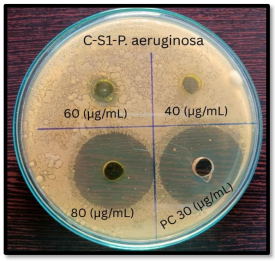

Fig. 4c: Antibacterial activity of carbinol against *P. aeruginosa*

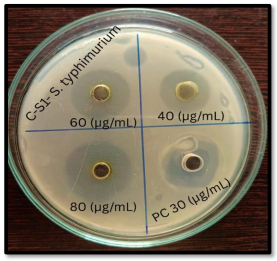

Fig. 4d: Antibacterial activity of carbinol plant extract plant extract against *S. typhimurium*

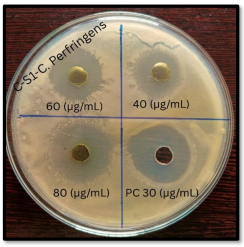

Fig. 4a: Antibacterial activity of carbinol against *C. perfringens*

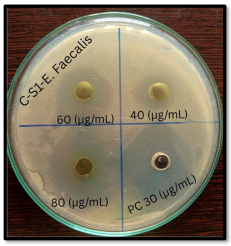

Fig. 4b:Antibacterial activity of carbinol plant extract plant extract against *E. faecalis*

Table .6 Antibacterial activity of ether extract of *Anogeissus latifolia* against selected bacteria

S.No	Test Organism	Zone of inhibition (mm) Ether extract of *Anogeissus latifolia* (µg/mL)			
		40	60	80	Standard (Chloramphenicol) 30µg/mL
1	*Clostridium perfringens* (MTCC-13124)	-	-	-	15.6±0.25
2	*Enterococcus faecalis* (MTCC-6845)	-	-	18.3±0.26	22.2±0.25
3	*Pseudomonas aeruginosa* (MTCC-6363)	-	-	17.3±0.36	23.2±0.15
4	*Salmonella typhimurium* (MTCC-443)	-	-	-	20.2±0.15

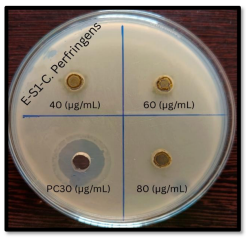

Fig. 5a: Antibacterial activity of carbinol *perfringens*

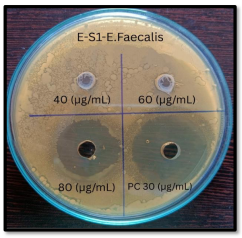

Fig. 5b: Antibacterial activity of carbinol extract against *C.* extract against *E. faecalis*/

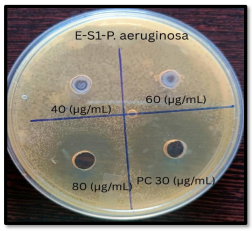

Fig. 5c:Antibacterial activity of carbinol extract against *P. aeruginosa*

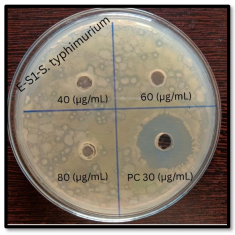

Fig. 5d: Antibacterial activity of carbinol extract against *S. typhimuriu*

Table .7 Antibacterial activity of water extract of *Anogeissus latifolia* against selected bacteria

S.No	Test Organism	Zone of inhibition (mm) Water extract of *Anogeissus latifolia* (µg/mL)			
		40	60	80	Standard (Chloramphenicol) 30µg/mL
1	*Clostridium perfringens* (MTCC-13124)	-	-	-	24.6±0.25
2	*Enterococcus faecalis* (MTCC-6845)	6.3±0.22	10.1±0.16	12.3±0.18	14.2±0.25
3	*Pseudomonas aeruginosa* (MTCC-6363)	-	9.1±0.14	12.2±0.26	19.1±0.15
4	*Salmonella typhimurium* (MTCC-443)	-	-	14.2±0.16	18.2±0.17

\

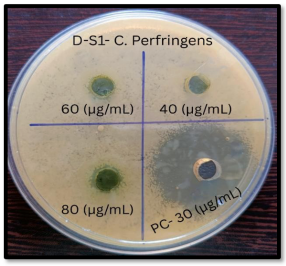

Fig. 6a: Antibacterial activity of carbinol extract against *C. perfringens*

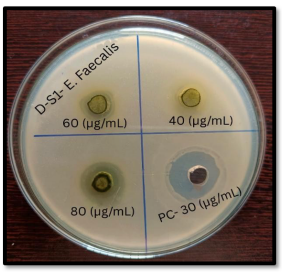

Fig. 6b: Antibacterial activity of carbinol extract against *E. faecalis*

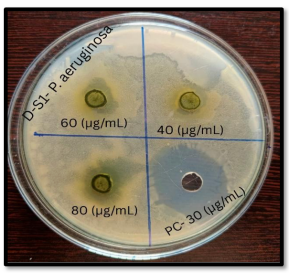

Fig. 6c: Antibacterial activity of carbinol extract against *P. aeruginosa*

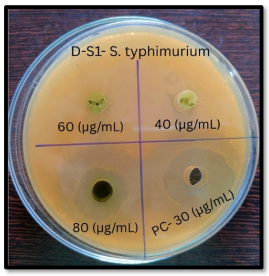

Fig. 6d: Antibacterial activity of carbinol extract against *S. typhimurium*

Conclusion

The present investigation provides comprehensive insight into the phytochemical composition and antibacterial potential of *Anogeissus latifolia* bark extracts, validating its ethnomedicinal relevance. Qualitative screening revealed the presence of diverse bioactive metabolites, including phenolics, flavonoids, tannins, terpenoids, steroids, glycosides, proteins, and quinones, which are key contributors to the plant's pharmacological activities. The solvent- dependent variability observed in both qualitative and quantitative analyses underscore the importance of extraction method optimization for maximizing bioactive yield. Methanol (carbinol) proved to be the most effective solvent, exhibiting the highest total phenolic (326.8 ± 22.6 mg GAE/g), flavonoid (135.4 ± 10.5 µg QE/mg), and tannin (95.4 ± 10.4 mg TAE/g) contents, followed by chloroform, water, and acetone. The antibacterial assay demonstrated strong inhibitory effects of the methanol extract against *Pseudomonas aeruginosa* and *Clostridium perfringens*, with inhibition zones comparable to the standard antibiotic chloramphenicol. This suggests that polar solvents efficiently extract phenolic and flavonoid- rich fractions responsible for the observed bioactivity. The comparative trend in antibacterial efficacy (carbinol > water > ether) further confirms the relationship between phytochemical abundance and antimicrobial potency.

References

Abreu, A.C., et al. (2020). Plant natural products for antimicrobial use: From traditional medicine to modern applications. *Phytochemistry Reviews*, 19(5), 1255–1280.

Bassetti, M., et al. (2023). Antimicrobial resistance: Challenges and opportunities in the post-antibiotic era. *Nature Reviews Microbiology*, 21(3), 143–158.

Garba, A.A., et al. (2025). Comparative phytochemical and antioxidant evaluation of *Anogeissus* species. *Journal of Ethnopharmacology*, 318, 116925.

Jain, S.K., et al. (2018). Ethnobotanical and pharmacological studies on *Anogeissus latifolia*: A review. *Indian Journal of Traditional Knowledge*, 17(2), 326–334.

Kumar, S., et al. (2021). Tannin-rich bark extracts as potential antimicrobial and wound- healing agents. *Journal of Medicinal Plants Research*, 15(7), 322–331.

Lal, D., et al. (2022). Phytochemical and pharmacological properties of *Anogeissus acuminata* and *A. latifolia*: Comparative evaluation. *Asian Pacific Journal of Tropical Biomedicine*, 12(8), 358–366.

Mishra, P., et al. (2023). Influence of solvent polarity on phytochemical yield and bioactivity in medicinal plants. *Frontiers in Plant Science*, 14, 1109321.

Olagbemide, P.T., et al. (2021). Bioactive plant secondary metabolites and their roles in health promotion. *Pharmacognosy Reviews*, 15(30), 101–113.

Panghal, M., et al. (2022). Phytochemicals: A promising arsenal against infectious diseases. *Journal of Applied Microbiology*, 133(4), 1723–1740.

Stray, F. (1998). *The Natural Guide to Medicinal Herbs and Plants.* Tiger Books International, London.

World Health Organization (2019). *WHO Global Report on Traditional and Complementary Medicine 2019.* Geneva: WHO Press.

\