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Potentials of leaf extracts of *Anogeissus leiocarpa* (DC.) Guill. & Perr. Against two pathogenic microbes

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Abstract

Plants have long served as dependable sources of therapeutic agents, and their contribution to drug discovery continues to attract widespread global attention. Anogeissus leiocarpa (DC.) Guill. & Perr. possesses considerable ethnobotanical relevance for the management of various ailments. This study evaluated the phytochemical composition and antibacterial activity of the ethanolic leaf extract of A. leiocarpa against Salmonella typhimurium and Escherichia coli. Phytochemical screening and antibacterial assays were conducted using validated analytical procedures. The results revealed the presence of tannins, phenols, glycosides, steroids, alkaloids, flavonoids, saponins, terpenoids, carbohydrates, and resins in varying concentrations. Quantitative analysis showed alkaloids (39.00 mg/g) as the most abundant phytochemical, while phenols (12.90 mg/g) were the least abundant. The ethanolic leaf extract of A. leiocarpa (EtLEAL) exhibited significant ($p < 0.05$) zones of inhibition against the test organisms in a concentration-dependent manner. The minimum inhibitory concentrations (MICs) of EtLEAL were ≤ 6.25 mg/mL for S. typhimurium and 12.5 mg/mL for E. coli, while the minimum bactericidal concentrations (MBCs) were 100 mg/mL and > 100 mg/mL, respectively. The calculated MBC/MIC ratios of 16.0 and 8.0 indicated a comparatively weaker bacteriostatic effect of the extract against S. typhimurium than E. coli. These findings suggested the need for further studies employing higher extract concentrations, different plant parts, and alternative extraction solvents to enhance antibacterial activity.

Nomenclature and units

ZOI = Zone of Inhibition (mm)
MIC = Minimum inhibitory concentration (mg/mL)
MBC = Minimum bactericidal concentration (mg/mL)
Phytochemical composition (mg/g)

1.0 Introduction

Plants have constituted the foundation of traditional medicine from antiquity to the present day, with their continued relevance largely dependent on floristic diversity. The use of plant parts, either singly or in combination, as therapeutic or preventive agents remains fundamental to human health and well-being (Davis & Choisy, 2024). Globally, over 80% of the population relies on traditional medicine and plant-derived products for primary health-care needs (Saggar *et al.,* 2022; Asigbaase *et al.,* 2023). Consequently, herbal medicine continues to serve as a viable alternative to certain orthodox pharmaceuticals (Nyirenda & Chipuwa, 2024). Medicinal plants represent invaluable reservoirs for the discovery of novel drugs targeting a wide spectrum of diseases. In Asia, the long-standing practice of herbal medicine reflects deep-rooted ethnobotanical knowledge shaped by sustained human–environment interactions (Yuan *et al*., 2016). Medicinal applications of plants involve the use of roots, barks, stems, leaves, and seeds, as well as extracts and decoctions prepared from these parts (Ogbulie *et al*., 2007; Chinsembu *et al*., 2019). The antimicrobial efficacy of medicinal plants is largely attributed to the presence of diverse secondary metabolites, commonly referred to as phytochemicals (Arsalan *et al*., 2022). In recent years, increasing scientific attention has been directed toward these biologically active compounds derived from indigenous plant species (Anand *et al*., 2019).

Anogeissus leiocarpa, a deciduous tree belonging to the family Combretaceae (Arbab, 2014), is sparsely distributed and widely exploited by local communities for its medicinal value. The species grows to a maximum height of approximately 30 m, with an average height of 15–18 m, and is characterized by light-green foliage and a broad, occasionally striped basal trunk. It is commonly known as African birch and locally referred to as Marke (Hausa), Atara (Igbo), and Orin-odan (Yoruba) (Salifou et al., 2017; Onoja *et al.,* 2018). Various parts of the plant have been traditionally employed in the treatment of microbial infections. The roots, used as chewing sticks, have demonstrated antibacterial activity against *Lactobacillus* spp. (Ali *et al.,* 2018; Ajao *et al.,* 2022). Additionally, combined extracts of *A. leiocarpa* and *Xanthoxylum gillettii*, mixed with citrus juice, have shown efficacy against HIV-associated opportunistic infections (Kwame *et al.,* 2005). Several studies have also reported broad-spectrum antimicrobial activity of *A. leiocarpa* against pathogenic microorganisms, alongside its therapeutic application in the treatment of helminthiasis, schistosomiasis, leprosy, diarrhoea, and psoriasis in humans and livestock (Soro *et al.,* 2013; Ndjonka *et al.,* 2013; Aiyenale *et al.,* 2017; Ali *et al.,* 2017).

Escherichia coli is a Gram-negative, facultative anaerobic, rod-shaped bacterium of the genus *Escherichia* and a major cause of food-borne infections in humans. Infection with *E. coli* is commonly associated with diarrhoea, abdominal pain, and stomach cramps, and may progress to life-threatening conditions in severe cases (Amin, 2019). Similarly, *Salmonella typhimurium*, a Gram-negative rod-shaped bacterium belonging to the family Enterobacteriaceae, is a significant food- and water-borne pathogen responsible for gastroenteritis characterized by diarrhoea, fever, and abdominal pain (Rosenberger, 2000; Yasin *et al.,* 2018).

The rapid emergence of antimicrobial-resistant bacterial strains represents a growing global public health challenge (Uddin *et al.,* 2021), leading to diminished efficacy of existing antibiotics and increased treatment failure. This challenge is particularly acute in developing countries, where rising drug costs driven by inflation and foreign-exchange constraints restrict access to imported pharmaceuticals (WHO, 2020). Moreover, treatment options are further limited by drug-related toxicity, adverse effects, and the prevalence of multidrug-resistant pathogens (Serwecinska, 2020; Aderaw *et al.,* 2023). These concerns underscore the need for alternative antimicrobial agents and have motivated the present study to investigate the antibacterial potential of ethanolic leaf extracts of *Anogeissus leiocarpa* against *Escherichia coli* and *Salmonella typhimurium*.(recommended)

2.0 Materials and Methods

2.1 Collection, preparation and ethanolic extract of plant materials

Leaf samples of *Anogeissus leoicarpa* obtained from Toro LGA of Bauchi State, Nigeria were identified at the Federal College of Forestry, Jos Herbarium, thereafter washed with distilled water and dried under shade in the chemistry laboratory for four weeks, to keep its compositional integrity (Adekanmi *et al*., 2020). The dried samples were pulverized into powder using mortar and pestle, packed in an airtight sterilized bottle and kept until used (Alozie *et al.,* 2022). 120g of the leaf powder was mixed with 80mls of ethanol and kept for 24 hours before filtering with whatsman filter paper. The oven-dried filtrate (at 37^{o}C) was refrigerated until used at 4^{o}C.

2.2 Yield of ethanolic leaf extract of *Anogeissus leiocarpa*(EtLEAL)

Percentage yield of extract was calculated using the formula below:

$$\%\ yield = \frac{\text{Weight of dried extract}}{Weight\ of\ dried\ plant\ material}\ x\ 100\%$$

(Azwanida, 2015).

2.3 Phytochemical analysis of ethanolic leaf extract of *Anogeissus leiocarpa*(EtLEAL)

The phytochemical constituents of the leaf powder were assessed using the following procedures (Prashant *et al.,* 2011).

Test for Saponins 1.0g of the leaf extract was mixed in 10 ml of distilled water in a conical flask, swirled vigorously for 30

seconds and kept for 30 minutes. A honey comb-like froth formed for more than 30 minutes showed positive result for saponins.

Test for Steroids: Into a test tube containing 2 ml of the leaf extract was 2.0 ml of acetic anhydride added. This was followed by addition of 1ml of conc. H_2SO_4 gently down the side of the tube. Blue-green coloration gave a positive test for steroids (Adekanmi *et al.,* 2020).

Test for Terpenoids; Half gram (0.5 g) of the leaf extract was mixed in 2.0 ml of Chloroform, followed by addition of 3.0ml of conc. H_2SO_4. Reddish- brown coloration showed a positive test for terpenoids.

Test for Flavonoids; The emergence of a yellow coloration after addition of 5 drops of aqueous NaOH was to 5 ml of the leaf extract gave a positive test for flavonoids.

Test for Tannins

2.0ml of $Fec1_3$ was added to 1.0ml of the leaf extract in a conical flask. The test was positive for tannins due to appearance of a dark green coloration (Ikeyi *et al*., 2013).

Test for Alkaloids (Dragendorffs and Wagner's Tests**)**

Two drops of the Dragendorffs Reagent were added to 2.0ml of the leaf extract. A rose red precipitate indicated the presence of alkaloids. Wagner's Test was carried out with 2 drops of the Wagner's Reagent added to 2.0 ml of the extract in a separate conical flask. A brown/reddish brown precipitate indicates the presence of alkaloids.

2.4 Methods of quantitative phytochemical assessment

The quantitative phytochemical assessment was carried based on the methods as described in Table 1 below:

Table 1: Quantitative Phytochemical Analysis

S/N	Phytochemical	Test/Method	Reference
1	Tannins	Folin–Denis spectrophotometric method	Pearson (1976); Baba & Malik, (2019); Arsalan *et al*., (2022)
2	saponins	gravimetric method	Obadoni & Ochuko (2001); Makkar *et al*., (2018); Arsalan *et al*., (2022).
3	Flavonoids	gravimetric method	Boham & Kocipai-Abyazan (1974); Anand *et al*., (2019); Arsalan et al., (2022).
4	Phenols	Folin–Ciocalteu spectrophotometric method	Singleton & Rossi, (1965); Ainsworth & Gillespie, (2019); Zhang *et al*., (2018).
5	Glycosides	Baljet's method	El-Olemy *et al.* (1994); Babu *et al*., 2020; Arsalan *et al*., 2022)
6	Steroids	ethanol extraction and gravimetric	Sofowora (2008), Anand *et al.,* (2019); Arsalan *et al.,* (2022
7	Alkaloids	gravimetric method	Harborne (1973); Arsalan *et al*., (2022)
8	Carbohydrate	phenol–sulfuric acid colorimetric method	(Masuko *et al.*, 2018; Ainsworth & Gillespie, 2019)
9	Terpenoids	gravimetric method	Ferguson (1956); Edeoga *et al*., (2018); Arsalan *et al.,* (2022).
10	Resins	gravimetric method	Zhang *et al.,* 2018; Arsalan *et al.,* 2022
11	Anthraquinone	spectrophotometric method based on alkaline hydrolysis	Babu *et al.,* 2020; Oboh *et al.,* 2021)

:

Quantitative phytochemical contents were calculated from gravimetric measurements or standard calibration curves, as appropriate, and expressed as milligrams per gram (mg/g) of dry plant material.

General formula for expression in mg/g for gravimetric methods

$$\text{Phytochemical content (mg/g)} = \frac{\text{Weight of phytochemical residue (mg)}}{\text{Weight of dry plant sample (g)}}$$

For spectrophotometric methods (using a standard curve)

$$\text{Phytochemical content (mg/g)} = \frac{C \times V}{W}$$

Where:

C = concentration obtained from the calibration curve (mg/mL)

V = total volume of extract (mL)

W = weight of dry plant material extracted (g)(Dong Shui *et al*., 2023)

2.5 Preparation of ethanolic leaf extract solutions (100–6.25 mg/ml)

The plant leaves were extracted using ethanol following standard phytochemical extraction procedures, and the solvent was

evaporated to dryness to obtain the dried ethanolic leaf extract. The dried extract was reconstituted in dimethyl sulfoxide (DMSO) to prepare a stock solution of 100 mg/mL. Briefly, 1.0 g of the dried ethanolic extract was accurately weighed and dissolved in 10 mL of DMSO, followed by thorough vortexing to ensure complete dissolution.

From the stock solution, two-fold serial dilutions were prepared aseptically, using DMSO to obtain extract concentrations of 50, 25, 12.5, and 6.25 mg/mL. All prepared concentrations were freshly used for antimicrobial susceptibility assays. The final concentration of DMSO in all test solutions did not exceed **1% (v/v)** and showed no inhibitory effect on the test organisms (Andrews, 2001; Balouiri, *et al.*, 2016).

2.6 Standardization of test organism and tests for antimicrobial activities

Cultures of *Salmonella typhimurium* and *Escherichia coli* were obtained from National Veterinary Research Institute (NVRI), VOM, Jos South LGA, Plateau State, Nigeria. Cultural morphological and biochemical tests were performed to ascertain their identities. The microbial isolates were maintained on nutrient agar at 4ºC (Akimnibosun *et al*., 2009).

Pure cultures of the test organisms were inoculated into 10 mL of nutrient broth in sterile McCartney bottles and incubated at 37 °C for 18–24 h. The resulting broth cultures were adjusted to a turbidity equivalent to the 0.5 McFarland standard (Andrews, 2001), corresponding to a cell density of approximately 1.5×10^6 colony-forming units (CFU)/mL (Olajubu *et al.*, 2012).

Nutrient agar (NA) was prepared according to the manufacturer's instructions (Oxoid CM003; 28 g L^{-1}). Briefly, 28 g of nutrient agar powder was weighed into a conical flask, and 1,000 mL of distilled water was added. The mixture was shaken using an orbital laboratory shaker set at 150 rpm to ensure complete dissolution, loosely capped, and sterilized by autoclaving at 121 °C for 15 min (Ardzard *et al.*, 2009).

Agar well diffusion methods were adopted to assessed the antimicrobial effects of ethanolic leaf extract *of Anogeissus leiocarpa* (EtLEAL) (Masood-ur *at al*,2017). Set sterile NA in plates were swabbed with the broth culture of the test bacteria isolates using sterile wire loop. Six equidistant wells were made through 6 mm diameter sterile cork borer with one at the center of the plate. Nutrient agar was added to seal the bottom. 100µl (0.1ml) of each concentration of the leaf extract (100, 50, 25, 12.5, 6.25 mg/ml) were respectively introduced into the well using sterile Pasteur pipette and 0.1ml the control (ciprofloxacin, 1.7mg/mL) was introduced into the well at the centre. This was done in triplicates and allowed to diffuse at ambient condition for 2 hours before incubation at 37ºc for 24hours. The antibacterial activities of the extracts were expressed as the mean diameter zone of inhibition (in mm) measured using a transparent ruler. This measured the degree of sensitivity.

2.7 Determination of minimum inhibitory concentration (MIC)

The effects of test concentrations of EtLEAL on the test organisms based on Inhibitory potential was assessed, using the broth dilution methods (Vollekova *et al*., 2001). Equal volume of nutrient broth was dispensed in to sterile test tubes, each containing different concentrations (6.25, 12.5, 25, 50, 100 mg/ml) of the leaf extract. Each tube was thereafter inoculated with 0.1ml of previously standardized suspension of *Escherichia coli and* S*almonella typhimurium* according to 0.5 Mcfarland (1.5×10^6 CFU/ml). The tubes were incubated at 37°C for 24 hours. After incubation, the tubes were examined for turbidity across the test concentrations, as an indication of microbial growth and on the contrary, inhibition. The MIC was obtained as the least extract concentration that inhibited the growth of the test microbes after 24 hours of incubation (Roger *et al*., 2015).

2.8 Determination of minimum bactericidal concentration (MBC)

Following the Minimum Inhibitory Concentration (MIC), tests, Minimum Bactericidal Concentration was assessed by sub-culturing aliquote portions from all the test tubes showing no turbidity or visible growth for MIC tests, after 1-10 dilution and inoculated with respective test concentrations of plant extract. The test tubes were incubated at 37ºC for 24hours. The MBC was measured as the least concentration that showing no turbidity or absence of growth after sub-culturing (Venkateswarulu *et al*.,2019).

2.9 Statistical analysis

Data gotten from the study were subjected to analysis of variance, using SPSS version 21. Significant means were tested with least significant difference.

3.0 Results and Discussion

3.1 Yield and phytochemical screening of *Anogeissus leiocarpa* leaf extracts

The yield percent of the ethanolic leaf extract of *Anogeissus leiocarpa* was 38.3% (Table 2). The phytochemical screening of EtLEAL indicated variations in tannins, phenols, glycosides, steroids, alkaloids, flavonoids, saponins, terpenoids, carbohydrates and resins, while anthraquinones was absent (Table 2).

Table 2: Showing the Percentage yield of plant extracts

Plant part	Solvent	Percentage yield (%)
Leaf	Ethanol	38.3

Table 3: Phytochemical Composition of EtLEAL

S/N	Phytochemical	Observation
1	Tannins	+

2	Phenols	++
3	Glycosides	++
4	Steroids	++
5	Alkaloids	++
6	Flavonoids	++
7	Saponins	++
8	Carbohydrate	+
9	Terpenoids	+
10	Resins	+
11	Anthraquinone	-

Key: − = Absence,+ = present, ++ = highly

Analysis based on retention factor (Rf) range of 12 ($\times 10^{-2}$) - 38($\times 10^{-2}$), indicated alkaloids (39.00mg/g) and phenols (12.90 mg/g) as the highest and lowest quantitative constituents respectively (Figure 1).

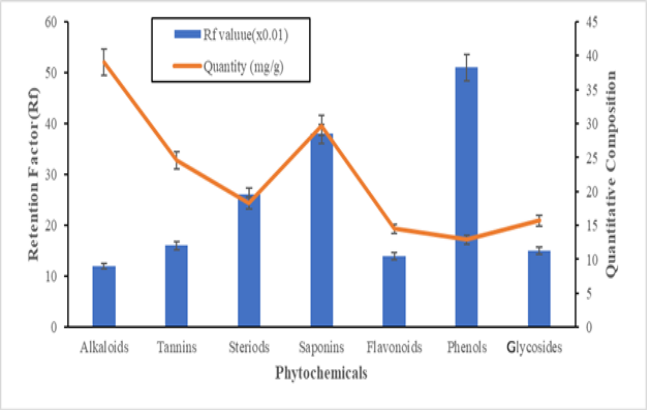

Figure 1: Quantitative Phytochemical Composition of Ethanolic Leaf Extract of *Anageisus leiocarpa*
R_f = Retention Factor

Overall, the extract demonstrated a clear dose-dependent antibacterial effect against both test organisms. *S. typhimurium* was more susceptible to the extract than *E. coli*, whereas ciprofloxacin showed superior inhibitory activity, particularly against *E. coli*.

3.2 Effects of EtLEAL on tests Microorganisms

The antibacterial activity of the extract against *Salmonella typhimurium* and *Escherichia coli* is presented as zones of inhibition (mm) expressed as mean ± standard deviation (SD) (Figure 2). The EtLEAL exhibited notable inhibitory activity against *S. typhimurium*, across all concentrations tested. The highest zone of inhibition (ZOI) was recorded at 100 mg/mL (22.7 ± 0.6 mm), followed by 50 mg/mL (20.3 ± 0.5 mm), 25 mg/mL (18.0 ± 0.4 mm), and 12.5 mg/mL (17.7 ± 0.4 mm), while the lowest activity was observed at 6.25 mg/mL (14.0 ± 0.3 mm). The positive control, ciprofloxacin (1.7mg/mL), produced a ZOI of 21.0 ± 0.5 mm. Statistical analysis indicated significant differences ($p < 0.05$) among the extract concentrations, as denoted by different superscript letters (Figure 2). Similarly, a concentration-dependent inhibitory pattern was observed for *E. coli*. The ZOI decreased progressively from 19.0 ± 0.5 mm at 100 mg/mL to 16.0 ± 0.4 mm at 50 mg/mL, 15.0 ± 0.4 mm at 25 mg/mL, 13.0 ± 0.3 mm at 12.5 mg/mL, and 12.7 ± 0.3 mm at 6.25 mg/mL. Ciprofloxacin exhibited the highest antibacterial activity against *E. coli* with a ZOI of 23.7 ± 0.6 mm, which was significantly greater ($p < 0.05$) than those produced by the extract.

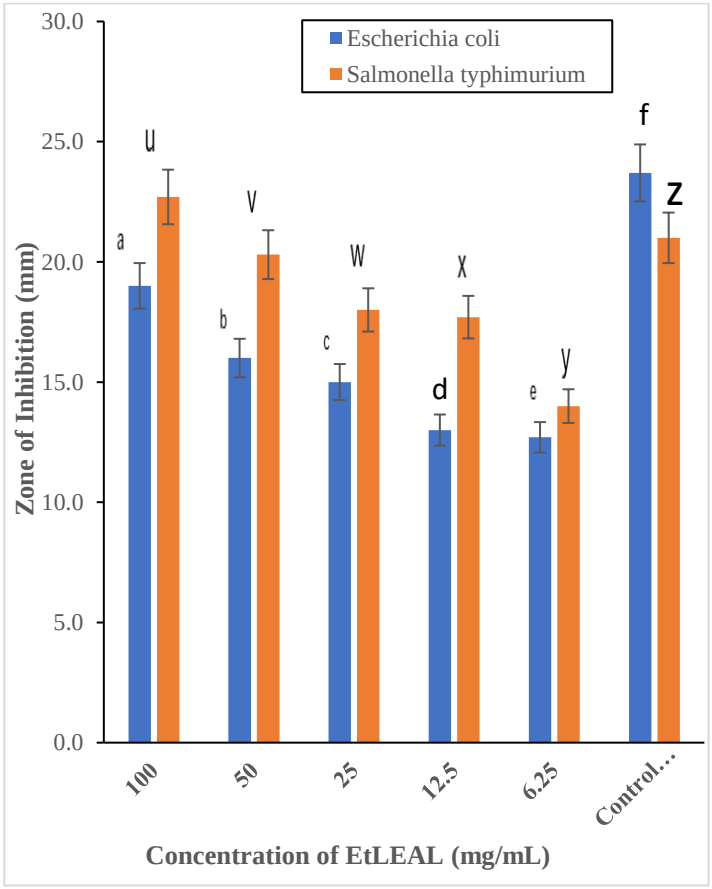

Figure 2: Mean Zone of Inhibition (mm) of Ethanolic Leaf Extract of *Anogeissus leiocarpa* on the test organisms

Values are expressed as mean ± SD (n = 3). Bars with different superscript indicated significant difference ($p < 0.05$) as determined by one-way ANOVA followed by Tukey's post-hoc test. Ciprofloxacin served as the positive control.
The effects of concentration of ethanolic leaf extract of *A. leiocarpa* were significant ($P < 0.05$) on the ZOI against *E. coli* and *Salmonella typhimurium*.

3.3 Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration of EtLEAL

The EtLEAL gave 12.5mg/ml and ≤6.25mg/ml as the minimum inhibitory concentration (MIC) against *Escherichia coli* and *Salmonella typhimurium* respectively, after incubating for 24 hours. Subsequently tests gave >100mg/ml and 100mg/ml as the

minimum bactericidal concentration (MBC) against *Escherichia coli* and *Salmonella typhimurium* respectively (Table 3).

Table 3: Antibacterial Effects of EtLEAL on Test Organisms

Antimicrobial Test								
			Extract Concentration(mg/mL)					Remark
S/N	**Test Org**	**IT (Hr)**	**100**	**50**	**25**	**12.5**	**6.25**	**MIC**
1	ST	24	-	-	-	-	-	≤6.25
2	EC	24	-	-	-	-	+	12.5
								MBC
3	ST	24	-	+	+	+	+	100
4	EC	24	+	+	+	+	+	>100
				MBC/MIC				**AP**
5	ST			16.0				Bacteriostatic
6	EC			8.0				Bacteriostatic

AP = Antibiotic Power, MIC = Minimum Inhibitory Concentration, MBC = Minimum Bactericidal Concentration, ST = *Salmonella typhimurium*, EC = *Escherichia coli*,IT = Incubation Time (Hour). Test Org = Test organism

4.0 Discussion

The detection of bioactive compounds (phytochemicals) in the ethanolic leaf extract of *Anogeissus leiocarpa* (EtLEAL) is consistent with the findings of Dayok *et al.* (2018), who reported the presence of biologically active phytochemical constituents in the leaf and stem bark extracts of *A. leiocarpa* with potential antimicrobial properties. These secondary metabolites are widely recognized for their role in inhibiting microbial growth. Wang *et al.* (2015) further emphasized that variations in the abundance and composition of phytochemicals influence the antibacterial potency of plant extracts, supporting their contribution to the observed activity of EtLEAL.

The antibacterial activity of EtLEAL against the test organisms demonstrated a concentration-dependent pattern, comparable in some cases to that of the standard antibiotic, ciprofloxacin. Notably, the zones of inhibition produced by EtLEAL against *Salmonella typhimurium* were relatively comparable to those of the positive control, indicating substantial antibacterial efficacy. This observation corroborates the report of Muhammad *et al.* (2022), who attributed microbial growth inhibition to the synergistic effects and strength of secondary metabolites present in plant extracts. The statistically significant differences ($p < 0.05$) observed across extract concentrations further suggest that EtLEAL possesses varying antibacterial activities against different pathogenic microorganisms.

The minimum inhibitory concentration (MIC) of EtLEAL, which ranged from 6.25 to 12.5 mg/mL, was lower than values previously reported by Timothy *et al.* (2015), indicating improved antibacterial effectiveness in the present study. This enhanced activity may be attributed to differences in extraction solvent, plant part used, or phytochemical composition. Previous studies have shown that plant-derived bioactive compounds exert inhibitory effects on microbial growth through mechanisms such as disruption of cell membranes, enzyme inhibition, and interference with cellular metabolism (Ikram *et al.*, 2015).

Furthermore, EtLEAL exhibited a bactericidal effect against *S. typhimurium* at 100 mg/mL, whereas a reduced effect was observed against *Escherichia coli* at the same concentration. This difference in susceptibility may be associated with the intrinsic resistance mechanisms of *E. coli*, including its outer membrane permeability barrier. The pronounced activity against *S. typhimurium* is, however, consistent with previous reports on the antibacterial potential of *A. leiocarpa*, highlighting its therapeutic relevance. These findings suggest that higher concentrations of EtLEAL or further purification of active compounds may be required to achieve comparable efficacy against *E. coli*.

The pronounced antibacterial activity of the ethanolic leaf extract of *Anogeissus leiocarpa* (EtLEAL), particularly against *Salmonella typhimurium*, can be attributed to the presence and relative abundance of multiple bioactive phytochemicals, including alkaloids, flavonoids, tannins, phenolic compounds, glycosides, steroids, saponins, carbohydrates, terpenoids, and resins as obtained from this study. Alkaloids are widely recognized for their antimicrobial efficacy and exert their effects by interfering with bacterial DNA replication, transcription, and protein synthesis, thereby inhibiting cell growth and division (Cowan, 1999; Othman *et al.*, 2019). The strong inhibitory activity observed at higher extract concentrations suggests a dose-dependent interaction between these compounds and bacterial cellular targets.

Flavonoids and phenolic compounds further contribute to antibacterial activity through disruption of bacterial cell membranes, inhibition of key metabolic enzymes, and chelation of metal ions essential for microbial survival (Cushnie & Lamb, 2011). Tannins enhance this effect by precipitating microbial proteins, inactivating adhesins, and forming complexes with bacterial cell wall components, mechanisms that are particularly effective against enteric pathogens such as *S. typhimurium* (Cowan, 1999). The comparatively lower susceptibility of *Escherichia coli* may be related to its outer membrane barrier and efficient efflux systems, which restrict phytochemical penetration.

In addition, glycosides and steroids present in EtLEAL contribute to antibacterial activity through membrane destabilization following enzymatic hydrolysis of glycosides and lipid–membrane interactions by steroidal compounds, resulting in increased permeability and cellular leakage (Doughari, 2012; Mahato *et al.*, 2020). Saponins, owing to their surface-active properties, form complexes with membrane sterols, leading to pore formation and cell lysis, which may explain the bactericidal effect observed against *S. typhimurium* at higher concentrations (Cheok *et al.*, 2014; Moghimipour *et al.*, 2015).

Although carbohydrates are not primary antimicrobial agents, certain polysaccharides inhibit microbial adhesion and biofilm formation, thereby indirectly enhancing antibacterial efficacy (Xu *et al.*, 2018). Terpenoids and resins are also known to exhibit strong antimicrobial activity by disrupting membrane integrity,

inhibiting respiration, and interfering with ion transport and enzymatic systems (Burt, 2004; Guimarães *et al.*, 2019; Sforcin & Bankova, 2011).

Collectively, the synergistic action of these phytochemicals provides a mechanistic explanation for the significant zones of inhibition and low MIC values recorded in this study. The enhanced susceptibility of *S. typhimurium* relative to *E. coli* further underscores the therapeutic potential of *A. leiocarpa* as a promising source of plant-derived antibacterial agents and supports further isolation and characterization of its active constituents.

5.0 Conclusion and Recommendations

This study demonstrated that the ethanolic leaf extract of *Anogeissus leiocarpa* possesses significant antibacterial activity against *Salmonella typhimurium* and *Escherichia coli*, with a clear concentration-dependent inhibitory effect. The extract showed greater efficacy against *S. typhimurium* than *E. coli*, while ciprofloxacin remained the most effective treatment, particularly against *E. coli*. The observed antibacterial activity can be attributed to the synergistic action of multiple phytochemicals, including alkaloids, flavonoids, tannins, phenolic compounds, glycosides, steroids, saponins, terpenoids, resins, and carbohydrates, which act through diverse mechanisms such as membrane disruption, enzyme inhibition, and interference with microbial metabolism. The low minimum inhibitory concentration values further support the antibacterial potency of the extract. Overall, the findings highlight the therapeutic potential of *A. leiocarpa* as a promising source of plant-derived antibacterial agents and justify further studies aimed at isolating and characterizing its bioactive compounds, as well as evaluating their efficacy and safety in vivo.

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Declaration of conflict of interest

The author hereby declared no conflict of interests associated with the study, manuscript and any organization in connection with the content of this manuscript.

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