

Phytochemical Screening and Antibacterial Potency of *Aspilia africana* Leaves and *Gardenia jasminoides* Root Extracts

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Abstract

The increasing resistance of pathogenic bacteria to conventional antibiotics has necessitated the search for alternative antimicrobial agents, particularly from plants. This study investigates the phytochemical composition and antibacterial activity of ethanol and n-hexane extracts of *Aspilia africana* leaves and *Gardenia jasminoides* roots. Phytochemical screening was carried out using standard methods. The extracts were tested against *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis* using the disc diffusion method and Minimum Inhibitory Concentration (MIC) determination. The phytochemical screening revealed the presence of alkaloids, flavonoids, tannins, saponins, and terpenoids in varying concentrations across both plant extracts. The ethanol extract of *Aspilia africana* exhibited the highest flavonoid content (12.38%), while *Gardenia jasminoides* roots had a significantly higher tannin concentration (23.68%) in the ethanol extract compared to *Aspilia africana* (0.12%). Alkaloids and saponins were also abundant in *Gardenia jasminoides* roots (5.37% and 9.85%, respectively), particularly in the ethanol extract. Both plant extracts demonstrated significant antibacterial activity, with the ethanol extracts showing higher potency. The results of the antimicrobial analysis showed that the ethanol extracts of both plants demonstrated greater antimicrobial efficacy, particularly at higher concentrations. *Gardenia jasminoides* root extract showed inhibitory activity against all tested bacteria at 50mg/l and lower concentrations. Similarly, the n-hexane extract exhibited antibacterial activity, albeit less potent than the ethanol extract. In conclusion, the results indicate that both plant species possess potent antibacterial properties, particularly in ethanol extracts, supporting their traditional use in treating bacterial infections and justifying further research into their medicinal applications.

Keywords: *Aspilia Africana*, *Gardenia Jasminoides*, Phytochemical Screening, Antibacterial Activity, Minimum Inhibitory Concentration.

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INTRODUCTION

Medicinal plants have long been utilized in traditional medicine due to their wide array of bioactive compounds, which possess significant therapeutic properties, including antibacterial, antiviral, antifungal, and anti-inflammatory activities [1]. The utilization of medicinal plants has been driven by the belief that they are effective with minimal side effects; in contrast to modern conventional medications derived from the same plant remedies [2]. The increasing prevalence of antimicrobial resistance has prompted a growing interest in the search for alternative sources of antimicrobial agents, particularly from plants. The exploration of plant-derived antimicrobial agents is particularly important given the declining efficacy of many synthetic antibiotics, and the global health challenge posed by multidrug-resistant bacterial strains.

Aspilia africana (Pers.) C.D. Adams, belonging to the Compositae (Asteraceae) family, is a fast-growing, perennial herbaceous plant native to tropical Africa. Commonly referred to as the “wild sunflower,” it thrives in open fields, roadsides, and disturbed habitats, making it a ubiquitous species across West and Central Africa. This plant holds significant importance in traditional medicine, where various parts—especially the leaves—are employed for multiple therapeutic purposes. The leaves of *A. africana* are known for their wound-healing properties, often used to stop bleeding and prevent infections when applied directly to cuts and wounds [3]. A decoction of the leaves is used to treat feverish headaches by washing the face and eyes [4]. In maternal health, an infusion of the leaves is consumed by women to aid childbirth and promote lactation by increasing milk flow in nursing mothers. Additionally, traditional healers

prepare a root decoction as an oral contraceptive, cough remedy for children, and as part of the treatment for tuberculosis [5]. Phytochemical investigations of *A. africana* have revealed the presence of bioactive compounds such as alkaloids, flavonoids, tannins, and saponins, which are known to contribute to its medicinal potential [4].

Gardenia jasminoides (Rubiaceae family), commonly known as Cape jasmine, is a small evergreen shrub native to parts of Asia. They are used as contraceptive, febrifuge, analgesic, diuretic, larvicide, antihypertensive, antibacterial, anxiolytic, antiplasmodial and for the treatment of headaches [6]. However, despite extensive research on *G. jasminoides*, the antibacterial potential of its root extracts remains largely unexplored.

Therefore, this study aims to evaluate the phytochemical composition and antibacterial efficacy of ethanol and n-hexane extracts of *A. africana* leaves and *G. jasminoides* roots. The antibacterial activity is assessed against *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis*.

MATERIALS AND METHODS

Plant Collection and Identification

Fresh leaves of *A. africana* and roots of *G. jasminoides* were collected from around the vicinity of the River State University Port Harcourt, Nigeria. The plants were identified and authenticated at the Department of Microbiology, Rivers State University, Port-Harcourt.

Preparation of Plant Extracts

Fresh leaves of *A. africana* and roots of *G. jasminoides* were collected and washed thoroughly with distilled water to remove dirt and impurities. The cleaned plant materials were chopped into small pieces and air-dried at room temperature for three weeks until fully dried. After drying, the plant materials were ground into a fine powder using a mechanical grinder and stored in airtight containers for further use.

Extraction of Plant Material

For each plant material, 120 g of the powdered samples were separately soaked in 300 mL of ethanol and 300 mL of n-hexane, yielding two sets of extracts for each plant (ethanol extract and n-hexane extract). The mixtures were macerated for 72 hours at room temperature with occasional shaking to enhance extraction. After the extraction period, the solutions were filtered using Whatman No. 1 filter paper to remove plant debris. The filtrates were concentrated using a rotary evaporator under reduced pressure at 40°C to evaporate the solvents, leaving behind the crude extracts. The crude ethanol and n-hexane extracts were stored in sterile containers and kept at 4°C until further analysis.

Phytochemical Screening

Qualitative Phytochemical Screening

Qualitative phytochemical screening for saponins, alkaloids, tannins, flavonoids, terpenoids, cardiac glycosides, in the ethanol and n-hexane extracts of both plants were done following standard methods as described by [7, 8].

Quantitative Phytochemical Screening

In addition to the qualitative tests, quantitative phytochemical screening was conducted to determine the concentrations of key bioactive compounds in both ethanol and n-hexane extracts. The following methods were employed:

Total Alkaloid Content

5 g of each extracts were weighed and placed into a flask. Then, 200 ml of 10% acetic acid in ethanol was added to the sample. The mixture was shaken and left to stand for 4 hours to extract the alkaloids. Afterward, the mixture was filtered, and the filtrate was collected. The filtrate was then evaporated to approximately a quarter of its original volume. A few drops of concentrated ammonium hydroxide (NH₄OH) solution were added to the concentrated filtrate to precipitate the alkaloids. The precipitate was filtered through pre-weighed filter paper. The filter paper containing the alkaloid precipitate was placed in an oven and dried at 60°C until a constant weight was achieved, which took approximately 30 to 60 minutes. Finally, the filter paper was weighed again to record the final weight, which was used to determine the alkaloid content.

$$\text{Saponins (\%)} = \frac{(\text{Weight of dish + Solute}) - \text{Weight of dish}}{\text{Sample weight}} \times 100$$

Saponin Content

To determine the total saponins, 20 g of each extract was weighed and transferred into a conical flask, followed by the addition of 100 mL of 20% aqueous ethanol. The mixture was heated in a water bath at 50°C for 4 hours with continuous stirring to facilitate

extraction. After heating, the solution was filtered, and the filtrate was re-extracted with 200 mL of 20% ethanol. The combined extracts were concentrated by evaporating the solution to 40 mL in a water bath at 90°C. The concentrate was transferred into a 250 mL separating funnel, where 20 mL of petroleum ether was added,

followed by vigorous shaking. The ether layer, containing impurities, was discarded, and the aqueous layer was retained. Next, 60 mL of n-butanol was added to the aqueous layer, and the mixture was shaken. The butanol layer was collected and washed twice with 10

mL of 5% aqueous sodium chloride (NaCl) to remove residual water-soluble components. Finally, the butanol extract was transferred into a pre-weighed dish, evaporated to dryness, and the dish was weighed again to determine the saponin content.

$$\text{Saponins (\%)} = \frac{(\text{Weight of dish + Solute}) - \text{Weight of dish} \times 100}{\text{Sample weight}}$$

Tannin Content

To determine the tannin content, 1 g of each extract was accurately weighed into a conical flask, followed by the addition of 10 mL of distilled water. The mixture was thoroughly agitated and allowed to stand at room temperature for 30 minutes. Afterward, it was centrifuged at 2500 rpm for 15 minutes. From the resulting supernatant, 2 mL was transferred into a test tube. Next, 1 mL of Folin-Ciocalteu reagent was added, along with 2 mL of saturated sodium carbonate solution. The mixture was then diluted to 10 mL with distilled water. Finally, the solution was incubated at room temperature for 30 minutes to allow for color development, indicating the presence of tannins.

Total Flavonoid Content

Flavonoids in the extracts were extracted following a standardized protocol. A 0.5 g of each sample was weighed and transferred into conical flasks, to which 20 mL of 80% aqueous methanol was added at ambient temperature. The mixture was agitated for three hours to enhance flavonoid extraction. The resulting solution was filtered using Whatman filter paper to separate solid residues. The filtrate was then transferred into a pre-weighed dish and evaporated to dryness over one hour. After complete drying, the dish was weighed again to determine the yield of the extracted flavonoids.

$$\text{Flavonoids (\%)} = \frac{(\text{Weight of dish + Solute}) - \text{Weight of dish} \times 100}{\text{Sample weight}}$$

Test for Terpenoids

0.1 g of both plant extracts were weighed and placed into a conical flask. Ethanol (9 mL) was added to the sample, and the mixture was allowed to soak for 24 hours. After soaking, the mixture was filtered to separate the solution from the solid residue. The filtrate was then

extracted using 10 mL of petroleum ether in a separating funnel. The ether layer was carefully poured into a pre-weighed dish. The dish was placed in an air oven to evaporate the ether to dryness over the course of 1 hour. Once dried, the dish was weighed again, and the weight was recorded to determine the terpenoid.

$$\text{Terpenoidss (\%)} = \frac{(\text{Weight of dish + Solute}) - \text{Weight of empty dish} \times 100}{\text{Sample weight}}$$

Antibacterial Assay

Test Organisms

The antibacterial activity of the ethanol and n-hexane extracts of *A. africana* leaves and *G. jasminoides* roots was evaluated against three bacterial strains: *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis*. These organisms were chosen due to their clinical relevance and prevalence in bacterial infections.

Antibacterial Susceptibility and Sensitivity Test

The antibacterial susceptibility of the extracts was determined using the disc diffusion method. Sterile paper discs (6 mm in diameter) were impregnated with different concentrations of the extracts (200 mg/L, 100 mg/L, 50 mg/L, 25 mg/L, 12.5 mg/L, 6.25 mg/L, and

3.125 mg/L). These concentrations were prepared using serial dilution from a stock solution of the crude extracts. Gentacycline (10 µg/disc) was used as a positive control to compare the antibacterial potency of the extracts, while discs soaked in sterile distilled water served as the negative control. Afterward, Mueller-Hinton agar plates were inoculated with the bacterial strains by evenly spreading 100 µL of the bacterial suspension (1.0×10^8 CFU/mL) over the surface of the agar. The prepared extract-impregnated discs were placed on the inoculated agar plates. The plates were incubated at 37°C for 24 hours. After incubation, the zones of inhibition around each disc were measured in millimeters to assess the antibacterial activity of the extracts.

Minimum Inhibitory Concentration (MIC) Determination

To further evaluate the antibacterial potency of the extracts, the minimum inhibitory concentration (MIC) was determined by serial dilution. The concentrations of the extracts (starting from 200 mg/L) were diluted in nutrient broth in a two-fold serial dilution to obtain the following concentrations: 200 mg/L, 100 mg/L, 50 mg/L, 25 mg/L, 12.5 mg/L, 6.25 mg/L, and 3.125 mg/L. The MIC was defined as the lowest concentration of the extract that completely inhibited visible bacterial growth after 24 hours of incubation at 37°C.

RESULTS AND DISCUSSION

Qualitative Phytochemical Screening

Qualitative tests are useful for determining the existence or lack of specific phytochemicals as well as their distribution in various plant parts [9]. The results are presented in Table 1, which shows the qualitative phytochemical screening of ethanol and n-hexane extracts of *A. africana* leaves and *G. jasminoides* roots. Alkaloids were identified in both plant extracts, with ethanol extracts showing a stronger presence compared to n-hexane extracts. Flavonoids were significantly present in the ethanol extract of *A. africana*, while the *G.*

jasminoides extracts exhibited moderate amounts in both solvents. Tannins and saponins were found to be more prominent in the ethanol extract of *G. jasminoides*, indicating the effectiveness of ethanol in extracting these compounds. Terpenoids, on the other hand, were detected in both ethanol and n-hexane extracts of both plants, although in lower concentrations compared to other phytochemicals. These phytochemicals play significant roles in the plant's antimicrobial, antioxidant, and anti-inflammatory activities. Flavonoids inhibit microbial enzymes and disrupt membrane integrity, making them useful against both Gram-positive and Gram-negative bacteria [10]. Tannins, which are more abundant in the ethanol extract of *G. jasminoides*, have antimicrobial effects by binding to microbial proteins and disrupting membrane functions. They also inhibit biofilm formation, a mechanism used by bacteria to resist antibiotics [11]. Research shows that alkaloid compounds are effective against a variety of pathogens, including *Staphylococcus aureus* and *Escherichia coli* [12].

This finding supports previous research that *A. africana* and *G. jasminoides* contains phytochemicals including saponins, flavonoids, terpenoids, alkaloids, and tannins [4-14].

Table 1: Qualitative Phytochemical Screening of Ethanol and n-Hexane Extracts of *Aspilia africana* Leaves and *Gardenia jasminoides* Roots

Phytochemical Component	Extract Type	<i>Aspilia Africana</i>	<i>Gardenia jasminoides</i>
Alkaloids	Ethanol	++	++
	n-Hexane	+	+
Flavonoids	Ethanol	++	+
	n-Hexane	+	+
Tannins	Ethanol	+	++
	n-Hexane	+	+
Saponins	Ethanol	+	++
	n-Hexane	+	+
Terpenoids	Ethanol	+	+
	n-Hexane	+	+

(+ = present, ++ = strongly present, - = absent)

Quantitative Phytochemical Evaluation

The quantitative phytochemical evaluation was conducted to determine the concentrations of specific phytochemicals in the extracts. The quantitative phytochemical analysis presented in Table 2 illustrates distinct variations in the concentration of bioactive compounds between the ethanol and n-hexane extracts of *A. africana* and *G. jasminoides*.

In terms of terpenoids, *A. africana* yielded a higher percentage in n-hexane extracts (2.31%)

compared to ethanol extracts (1.60%), whereas *G. jasminoides* demonstrated greater terpenoid levels in ethanol extracts (1.92%) than in n-hexane (1.75%). Flavonoid content was markedly higher in *A. africana*, with values of 12.38% for ethanol and 2.55% for n-hexane, in contrast to *G. jasminoides*, which had lower flavonoid concentrations (2.40% and 1.05%, respectively).

Table 2: Quantitative Phytochemical Evaluation of Ethanol and n-Hexane Extracts of *Aspilia africana* leaves and *Gardenia jasminoides* roots

Phytochemical Component	Extract Type	<i>Aspilia africana</i> (%)	<i>Gardenia jasminoides</i> (%)
Terpenoids	Ethanol	1.60	1.92
	n-Hexane	2.31	1.75
Flavonoids	Ethanol	12.38	2.40
	n-Hexane	2.55	1.05
Alkaloids	Ethanol	4.80	5.37
	n-Hexane	4.30	3.29
Saponins	Ethanol	2.69	9.85
	n-Hexane	2.46	7.32
Tannins	Ethanol	0.12	23.68
	n-Hexane	0.14	6.03

Alkaloid levels were higher in *G. jasminoides*, particularly in the ethanol extract (5.37%), compared to *A. africana* (4.80% for ethanol and 4.30% for n-hexane). Notably, saponin concentrations were significantly higher in *G. jasminoides* ethanol extract (9.85%) than in *A. africana* (2.69%). Tannin levels were exceptionally pronounced in *G. jasminoides* ethanol extract (23.68%), showcasing its rich tannin content compared to *A. africana* (0.12%).

These findings highlight the diverse phytochemical profiles of *A. africana* and *G. jasminoides*, indicating that *A. africana* is particularly rich in flavonoids, while *G. jasminoides* exhibits elevated levels of tannins and saponins. Such variations in phytochemical content suggest differing pharmacological potentials and underscore the significance of these plants in traditional medicine and

their potential applications in pharmaceutical formulations.

Antibacterial Activity of *Gardenia jasminoides* Ellis Root Extracts and *Aspilia africana* Leaf Extracts

The ethanol extract of *G. jasminoides* root displayed strong antibacterial activity, particularly against *S. aureus*, which exhibited the highest inhibition zone of 28 mm at 200 mg/l (Table 3). *E. coli* and *B. subtilis* were also susceptible, with inhibition zones of 24 mm and 22 mm, respectively, at the same concentration. The activity diminished at lower concentrations, with no inhibition observed at 3.125 mg/l. These findings suggest that *G. jasminoides* ethanol extract has potent antibacterial properties, comparable to gentamicin, a standard antibiotic, especially against gram-positive bacteria (*S. aureus*).

Table 3: Antibacterial Activity of Ethanol Extract of *Gardenia jasminoides* Ellis Root against *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis*

Organism	200 mg/l (mm)	100 mg/l (mm)	50 mg/l (mm)	25 mg/l (mm)	12.5 mg/l (mm)	6.25 mg/l (mm)	3.125 mg/l (mm)	Gentamicin (10ug)
<i>E. coli</i>	24	20	18	14	12	10	0	24
<i>S. aureus</i>	28	22	18	16	12	10	0	22
<i>B. subtilis</i>	22	20	18	16	14	10	0	22

Similarly, the n-hexane extract of *G. jasminoides* root exhibited considerable antibacterial activity, although slightly lower than the ethanol extract. At 200 mg/l, the n-hexane extract produced inhibition zones of 26 mm for *S. aureus*, 24 mm for *E. coli*, and 22

mm for *B. subtilis* (Table 4). This suggests that the non-polar constituents extracted by n-hexane also contribute to the antibacterial properties of *G. jasminoides*, although to a lesser extent than the ethanol extract.

Table 4: Antibacterial Activity of n-hexane Extract of *Gardenia jasminoides* Ellis Root against *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis*

Organism	200mg/l (mm)	100mg/l (mm)	50mg/l (mm)	25mg/l (mm)	12.5mg/l (mm)	6.25mg/l (mm)	3.125mg/l (mm)	Gentamicin (10ug)
<i>E. coli</i>	24	18	16	14	12	8	6	24
<i>S. aureus</i>	26	20	18	16	12	10	8	22
<i>B. subtilis</i>	22	18	16	14	10	6	0	22

Table 5: Antibacterial susceptibility test of Ethanol extract of *Aspilia africana* (leave extracts) on *Escherichia coli*, *Staphylococcus aureus* and *Bacillus subtilis*

Organism	200mg/l (mm)	100mg/l (mm)	50mg/l (mm)	25mg/l (mm)	12.5mg/l (mm)	6.25mg/l (mm)	3.125mg/l (mm)	Gentamycine (10ug)
<i>E. coli</i>	20	18	16	12	11	10	0	24
<i>S. aureus</i>	22	18	16	14	12	8	0	22
<i>B. subtilis</i>	18	16	14	11	8	6	0	22

Key: -: Clear tubes, +: Turbidity in tubes

The ethanol extract of *A. africana* leaves exhibited moderate antibacterial activity, with inhibition zones of 22 mm, 20 mm, and 18 mm for *S. aureus*, *E. coli*, and *B. subtilis*, respectively, at 200 mg/l (Table 5). The activity reduced at lower concentrations, with no inhibition observed at 3.125 mg/l.

The n-hexane extract of *A. africana* also displayed antibacterial activity, though less potent than the ethanol extract. At 200 mg/l, inhibition zones of 18

mm for both *S. aureus* and *E. coli*, and 16 mm for *B. subtilis* were recorded (Table 6). Similar to the ethanol extract, no activity was detected at 3.125 mg/l, highlighting the concentration-dependent nature of the extract's antibacterial efficacy.

These results align with previous studies that reported antibacterial properties of *A. africana* and *G. jasminoides* [14, 15].

Table 6: Antibacterial susceptibility test of n-Hexane extract of *Aspilia africana* leaves on *Escherichia coli*, *Staphylococcus aureus* and *Bacillus subtilis*

Organism	200mg/l (mm)	100mg/l (mm)	50mg/l (mm)	25mg/l (mm)	12.5mg/l (mm)	6.25mg/l (mm)	3.125mg/l (mm)	Gentamycine (10ug)
<i>E. coli</i>	18	16	14	12	10	6	0	24
<i>S. aureus</i>	18	16	14	10	8	0	0	22
<i>B. subtilis</i>	16	14	12	10	8	6	0	22

Key: -: Clear tubes, +: Turbidity in tubes

Antimicrobial Minimum Inhibitory Concentration

The minimum inhibitory concentration (MIC) test assesses the lowest concentration of a substance that inhibits visible bacterial growth. The results, as presented in Tables 7–10, highlight varying degrees of inhibitory activity depending on the extract type and concentration.

MIC of *Gardenia Jasminoides* Root Extracts

The ethanol extract of *G. jasminoides* root showed a strong inhibitory effect on all three bacteria,

though with differing MICs (Table 7). The MIC for both *E. coli* and *S. aureus* was determined to be 50 mg/l, as growth was absent at higher concentrations (100 mg/l and 200 mg/l). However, growth was observed at 50 mg/l and below, indicating this as the threshold for inhibition. For *B. subtilis*, the MIC was lower, at 25 mg/l, demonstrating greater sensitivity to the extract. This pattern suggests that *B. subtilis* is more susceptible to the ethanol extract of *G. jasminoides* compared to *E. coli* and *S. aureus*.

Table 7: Minimum inhibitory test of Ethanol extract of *Gardenia jasminoides* Ellis Root on *Escherichia coli*, *Staphylococcus aureus* and *Bacillus subtilis*

Organism	200 mg/l	100 mg/l	50 mg/l	25 mg/l	12.5 mg/l	6.25 mg/l	3.125 mg/l
<i>E. coli</i>	-	-	+	+	+	+	+
<i>S. aureus</i>	-	-	+	+	+	+	+
<i>B. subtilis</i>	-	-	-	+	+	+	+

Key: -: Clear tubes, +: Turbidity in tubes

The n-hexane extract exhibited lower antibacterial efficacy compared to the ethanol extract (Table 8). The MIC for *E. coli*, *S. aureus*, and *B. subtilis* was observed at 25 mg/l, as no growth was observed at 50 mg/l and above. However, growth occurred at

concentrations of 25 mg/l and below. This indicates that while the n-hexane extract of *G. jasminoides* is effective at inhibiting bacterial growth, it requires slightly higher concentrations to achieve the same inhibitory effects as the ethanol extract.

Table 8: Minimum inhibitory test of n-hexane extract of *Gardenia jasminoides* Root on *Escherichia coli*, *Staphylococcus aureus* and *Bacillus subtilis*

Organism	200 mg/l	100 mg/l	50 mg/l	25 mg/l	12.5 mg/l	6.25 mg/l	3.125 mg/l
<i>E.coli</i>	-	-	-	+	+	+	+
<i>S.aureus</i>	-	-	-	+	+	+	+
<i>B.subtillis</i>	-	-	-	+	+	+	+

Key: -: Clear tubes, +: Turbidity in tubes

MIC of *Aspilia Africana* Leaf Extracts

The ethanol extract of *A. africana* leaves demonstrated moderate antibacterial activity across all three bacterial species (Table 9). For *E. coli*, *S. aureus*, and *B. subtilis*, the MIC was consistently observed at 50 mg/l, with no bacterial growth at 100 mg/l and above.

Growth occurred at 50 mg/l and lower, indicating that concentrations higher than 50 mg/l are necessary to inhibit these bacterial species. This uniformity in MIC values across the bacteria suggests that the ethanol extract of *A. africana* possesses broad-spectrum antibacterial properties.

Table 9: Minimum inhibitory test of Ethanol extract of *Aspilia africana* (leave extracts) on *Escherichia coli*, *Staphylococcus aureus* and *Bacillus subtilis*

Organism	200 mg/l	100mg/l	50 mg/l	25 mg/l	12.5 mg/l	6.25 mg/l	3.125 mg/l
<i>E. coli</i>	-	-	-	+	+	+	+
<i>S. aureus</i>	-	-	-	+	+	+	+
<i>B. subtilis</i>	-	-	-	+	+	+	+

Key: -: Clear tubes, +: Turbidity in tubes

Similar to the ethanol extract, the n-hexane extract of *A. africana* leaves exhibited antibacterial activity, though with slightly lower MIC values (Table 10). For all three bacterial strains, the MIC was found to be 25 mg/l. Turbidity was observed at concentrations of

25 mg/l and below, while higher concentrations resulted in clear tubes. This suggests that the n-hexane extract of *A. africana* is more potent than its ethanol counterpart, inhibiting bacterial growth at lower concentrations.

Table 10: Minimum inhibitory test of n-Hexane extract of *Aspilia africana* (leave extracts) on *Escherichia coli*, *Staphylococcus aureus* and *Bacillus subtilis*

Organism	200 mg/l	100 mg/l	50 mg/l	25 mg/l	12.5 mg/l	6.25 mg/l	3.125 mg/l
<i>E. coli</i>	-	-	-	+	+	+	+
<i>S. aureus</i>	-	-	-	+	+	+	+
<i>B. subtilis</i>	-	-	-	+	+	+	+

Key: -: Clear tubes, +: Turbidity in tubes

The results indicate that both *G. jasminoides* root and *A. africana* leaf extracts demonstrated significant antibacterial properties, with the ethanol extracts generally showing higher efficacy compared to the n-hexane extracts. The ethanol extract of *G. jasminoides* root had a lower MIC for *B. subtilis* compared to the other bacteria, indicating its stronger activity against this strain. Meanwhile, the n-hexane extract of *A. africana* leaves displayed more potent inhibition across all strains at a lower concentration (25 mg/l). The differences in MIC between the ethanol and n-hexane extracts can be attributed to the varying solubility of bioactive compounds in the respective solvents. Ethanol, being a polar solvent, is likely to extract polar compounds such as phenolics and flavonoids, which are known for their antimicrobial properties [12-16]. On the other hand, n-hexane, a non-polar solvent, extracts non-polar compounds like terpenoids and fatty acids, which also exhibit antimicrobial effects but may require higher concentrations to achieve inhibition [17].

Overall, both *G. jasminoides* and *A. africana* extracts demonstrated significant antibacterial properties, with the ethanol extracts generally showing higher efficacy compared to the n-hexane extracts. The ethanol extract of *G. jasminoides* root had a lower MIC for *B. subtilis* compared to the other bacteria, indicating its stronger activity against this strain. Meanwhile, the n-hexane extract of *A. africana* leaves displayed more potent inhibition across all strains at a lower concentration (25 mg/l).

CONCLUSION

The results presented in this study demonstrate that the ethanol and n-hexane extracts of *A. africana* leaves and *G. jasminoides* roots contain significant phytochemicals, including alkaloids, flavonoids, tannins, saponins, and terpenoids. Furthermore, the antibacterial analysis revealed that both extracts displayed inhibitory activity against *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis*, with ethanol extracts showing lower minimum inhibitory concentrations (MICs) and thus stronger antimicrobial activity. This suggests the

greater potential of ethanol extracts for therapeutic applications in combating bacterial infections. The findings highlight the pharmacological relevance of *A. africana* leaves and *G. jasminoides* roots, particularly their ethanol extracts, in developing plant-based antimicrobial agents. Further research is recommended to isolate and characterize the active compounds responsible for these effects.

Competing Interest: None declared

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