

Antimicrobial effects of Utazi (*Gongronema latifolium* benth.) leaf extracts against selected microbial isolates

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Abstract

The growing problem of antibiotic resistance has made it increasingly difficult to treat common infections, prompting the search for new and effective alternatives. One promising source is *Gongronema latifolium*, a leafy plant widely used in traditional African medicine. This study is aimed at exploring the antimicrobial potential of its leaf extracts, specifically the aqueous, ethanol, and methanol extracts, against selected Gram-positive and Gram-negative bacterial isolates including *Staphylococcus aureus*, *Bacillus subtilis*, *Bacillus cereus*, *Enterococcus sp.*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Citrobacter freundii*, *Citrobacter koseri*, *Proteus mirabilis*, and *Acinetobacter baumannii*. The phytochemical screening of *G. latifolium* leaf extracts revealed the presence of bioactive compounds such as flavonoids, saponins, phenols, terpenoids, glycosides, and phytosterols. Antibiotic susceptibility with *G. latifolium* extracts using the agar well diffusion method, at 1g/ml concentration, all three extracts exhibited antibacterial activity, though at varying levels with the methanol extract showing the highest effect, with the highest inhibition zone of 32 mm against *Bacillus subtilis*. Other susceptible organisms included *Staphylococcus aureus* (16 mm) *Klebsiella pneumonia* (13 mm), *Pseudomonas aeruginosa* (12 mm), *Proteus mirabilis* (9 mm) and *Citrobacter sp.* (12 mm). However, *Acinetobacter baumannii* (0-8 mm) and *Enterococcus sp.* showed higher resistance to the plant extracts (0-10 mm). These results show the potential of *G. latifolium* as a new antibacterial agent and validate the traditional use of the plant in the treatment of infections. Particularly, the methanol extract shows potential for further development into treatments targeting drug-resistant bacteria.

Keywords: *Gongronema Latifolium*; Antimicrobial; Resistance; Phytochemical Composition; Antibiotics.

1. Introduction

The relationship between humans and microorganisms has been complex and multifaceted throughout history. Microorganisms, particularly bacteria, exist in virtually every habitat on Earth and have evolved intricate relationships with humans, ranging from beneficial symbiotic associations to pathogenic interactions that cause disease. The discovery of antibiotics in the early 20th century marked a turning point in medical research, ushering in what many refer to as the “antibiotic era” and drastically transforming the practice of medicine.

However, the widespread and indiscriminate use of antibiotics has led to one of the most pressing public health challenges of the 21st century: antimicrobial resistance (AMR). Due to natural selection and antibiotic exposure, bacteria have developed various mechanisms to resist the effects of antibiotic drugs [1]. The World Health Organization (WHO) has declared antimicrobial resistance one of humanity's top ten global public health threats. According to epidemiological data, approximately 700,000 people die annually due to drug-resistant infections, with projections suggesting this figure could rise to 10 million deaths per year by 2050 if effective interventions are not implemented [2].

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The growing incidence of antibiotic resistance among pathogenic bacteria has become a significant challenge in global health. Conventional antibiotics are becoming increasingly ineffective, leading to prolonged illnesses, higher medical costs, and increased mortality. As a result, there is an urgent need to discover new antimicrobial agents, especially from natural sources [3].

The African continent, particularly Nigeria, is home to a wide range of plant species, some of which have medicinal properties. This rich ethnopharmacological heritage represents a largely untapped reservoir of potential antimicrobial compounds. Among these plants, *Gongronema latifolium* stands out as a species with the potential to fight bacterial infections.

Gongronema latifolium, commonly known as “utazi” among the Igbos and “arokeke” among the Yorubas of Nigeria, is a perennial edible plant belonging to the family *Asclepiadaceae*. It is indigenous to tropical and sub-tropical regions, particularly in countries across West Africa, including Nigeria, Ghana, Senegal, and Sierra Leone [4]. The plant grows as a slender-stemmed climbing plant that can reach heights of up to 5 meters when properly supported with leaves that are broadly heart-shaped, with a characteristic bitter taste [5].

Traditional healers across West Africa have utilized various parts of *G. latifolium* for centuries to treat a wide range of ailments including diabetes, hypertension, malaria, digestive disorders, inflammation, and notably, infectious diseases [6]. Recent scientific research has begun to validate the traditional uses of *G. latifolium*, particularly regarding its antimicrobial properties. Several studies have demonstrated inhibitory effects against some Gram-positive and Gram-negative bacteria, including *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Salmonella* sp. [7,8].

This study aims to investigate and compare the antimicrobial effects of different solvent extracts (aqueous, ethanol, and methanol) of *G. latifolium* leaf against selected Gram-positive and Gram-negative bacterial populations and identify promising extract formulations for developing new antimicrobial agents.

2. Materials and methods

2.1. Collection of plant samples and test organisms

Fresh samples of *G. latifolium* leaves were purchased from the local dealers at Oja-Oba market in Iwo, Osun State. All samples were transported to the Microbiology laboratory, Bowen University, Iwo, Osun state, Nigeria.

Pure cultures of bacterial isolates were obtained from Dr. Omowumi Akinola and the Microbiology Laboratory of the Microbiology Programme, Bowen University, Iwo, Osun State. Before being used, the purity of the bacterial isolates was confirmed by streaking them onto a sterile nutrient agar plate and incubating them for 24 hours at 37°C. The pure isolates, as shown by their colonial and microscopic features and biochemical tests carried out, were certified.

2.2. Antibiotic sensitivity test

After confirming the purity of the bacterial isolates, antibiotic sensitivity test was carried out using the Kirby-Bauer agar disc diffusion method. A loopful of each 24-hour pure bacterial culture was transferred into a test tube containing 2 mL of sterile water and mixed until the turbidity matched the McFarland standard. Sterile swab sticks were then used to evenly spread the bacterial suspension onto the surface of Muller-Hinton agar plates. For Gram-positive bacteria isolates, the antibiotic discs utilized were Clotrimazole (COT) 25µg, Ceftriaxone (CEF) 30µg, Amoxicillin (AMX) 25µg, Erythromycin (ERY) 5µg, Ciprofloxacin (CEF) 10µg, Ofloxacin (OFL) 5µg, Azithromycin (AZY) 12µg, Pefloxacin (PEF) 10µg, Levofloxacin (LEV) 5µg, and Tetracycline (TET) 10µg. For Gram-negative isolates, Ampiclox (AMX) 30µg, Levofloxacin (LEV) 5µg, Cefuroxime (CEF) 30µg, Ciprofloxacin (CPX) 30µg, Chloramphenicol (CHL) 30µg, Penicillin (PEN) 10µg, Tetracycline (TET) 30µg, Gentamycin (GEN) 10µg, Streptomycin (STR) 10µg, and Ofloxacin (OFL) 5µg were the antibiotics discs used. Before incubating the plates at 37°C for 24 hours, antibiotic-impregnated paper discs were gently but firmly placed on the surface of the inoculated agar. After incubation, the zones of inhibition around the discs were measured and assessed to determine the bacterial susceptibility to the antibiotics following the CLSI (Clinical and Laboratory Standards Institute) 2024 standard-[9].

2.3. Processing of the leaves

The leaves of the plant were carefully separated from the stalk and air dried for 15 days. All dried leaf samples were ground into fine powder using an electric blender. The powdered plant material was then placed in a clean airtight container before the time for extraction. The extraction was done using various solvents (ethanol, aqueous and methanol).

2.4. Extraction with solvent (Ethanol, Methanol and Aqueous)

A hundred grams (100 g) of the dried, ground plant material was weighed and placed into a clean, dry container. Then, 1000 mL of each solvent was added to completely cover the leaf materials. The container was sealed to prevent evaporation and contamination, and the mixture was left to soak at room temperature for 48 hours, with a gentle shaking each day. After soaking, the mixture was filtered through muslin cloth to separate the liquid extract from the solid residue. After filtration, the filtrate was transferred to a rotary evaporator and concentrated to remove the solvent and obtain the crude extract. The extracts were then preserved in the fridge until they were needed for use.

2.5. Phytochemical analysis of *Gongronema latifolium*

Qualitative analyses were carried out on the methanol, ethanol, and aqueous leaf extract as follows

- Test for Saponins: 5.0 mL of distilled water was mixed with each crude extract in a test tube, and it was mixed vigorously. The frothing was mixed with a few drops of olive oil and mixed vigorously. The appearance of foam indicates the presence of saponins.
- Test for Flavonoids: Following the alkaline reagent test method, 10% ammonium hydroxide solution was added to 1 mL of each plant extract. The appearance of a yellow fluorescence indicates the presence of flavonoids.
- Test for Tannins: Braymer's test was carried out to detect tannins. 3 mL of distilled water was added to 1 mL of the plant extract, followed by 3 drops of a 10% ferric chloride solution. A blue-green coloration indicates the presence of tannins.
- Test for Terpenoids: 0.5 mL of the extract was treated with 2 mL of chloroform and concentrated sulfuric acid. Formation of reddish brown colour at the interface indicates the presence of terpenoids.
- Test for Alkaloids: To 1 mL of each extract, 1 mL of Marquis reagent, 2 mL of concentrated sulfuric acid, and a few drops of 40% formaldehyde were added and mixed. Appearance of dark orange or purple color indicates the presence of alkaloids.
- Test of Phenols: To 2 mL of each extract, 2 mL of 5% aqueous ferric chloride was added; formation of a blue color indicates the presence of phenols in the sample extract.
- Test for Glycosides: To 1 mL of each extract, 0.5 mL of glacial acetic acid and 3 drops of 1% aqueous ferric chloride solution were added; formation of a brown ring at the interface indicates the presence of cardiac glycosides in the sample extract.
- Test for Phytosterols: A few drops of concentrated sulfuric acid (H₂SO₄) were added to 1 mL of each plant extract. The mixture was shaken well and then allowed to stand. A red color in the lower layer indicates the presence of phytosterols.

2.6. Susceptibility tests of leaf extracts of *G. latifolium*

Agar well diffusion technique was used to determine the antibacterial activity of the extracts. Each isolate was prepared by suspending a 24-hour pure bacterial culture in 2 mL of sterile water in test tubes and mixing thoroughly until a uniform suspension was obtained. The turbidity of each suspension was then adjusted to match the McFarland standard. A sterile swab stick was dipped into the bacterial suspension and used to evenly spread the organism over the entire surface of the Mueller-Hinton agar plate.

A sterile 3 mm cork borer was used to bore 3 holes in each of the inoculated Mueller-Hinton agar plates. The bored holes (wells) were labelled appropriately to represent each of the leaf extracts. For each extract, the concentration of 1 g/mL was used by weighing 1g of the crude extract and dissolving it in 1 mL of the solvent that was used for the extraction. Using a micropipette, 100 µl of each extract was placed into its designated well on the agar plates. The plates were then incubated at 37°C for 24 hours. After incubation, the plates were examined for the presence of zones of inhibition, which were measured and recorded accordingly.

3. Results

The bacterial isolates obtained from the Bowen University Microbiology laboratory were identified to include *Citrobacter freundii*, *Citrobacter koseri*, *Acinetobacter baumannii*, *Enterococcus sp.*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Bacillus subtilis*, *Bacillus cereus*, *Pseudomonas aeruginosa*, and *Proteus mirabilis* as shown in table 1.

Table 2 shows the result of the phytochemical analysis carried out on the three plant extracts (aqueous, ethanol, and methanol). Saponins, flavonoids, phenols, phytosterols, and terpenoids were present in all the extracts, but in different

concentrations. Tannins were absent in the aqueous extract, glycosides were absent in the ethanol and methanol extracts, and were present only in the aqueous extract. Alkaloids were absent in all three extracts. Flavonoids and terpenoids were highly present in the aqueous extracts.

Table 1 Biochemical Tests and Gram Staining Results Showing the Most Probable Organisms Identified of Sample Bacterial Isolates

S/N	ISOLATE	CITRATE	INDOLE	MR	VP	MOTILITY	CATALASE	STARCH	GRAM	GLUCOSE	LACTOSE	SUCROSE	MANNITOL	ISOLATE NAME
1.	AY1	+	-	+	-	+	+	-	-	AG	+	AG	AG	<i>Citrobacter freundii</i>
2.	AY1A	+	-	+	-	-	+	-	-	+	-	+	+	<i>Acinetobacter baumannii</i>
3.	AY1B	+	+	+	-	+	+	-	-	AG	+	AG	AG	<i>Citrobacter koseri</i>
4.	119	-	+	+	-	-	-	-	+	+	-	+	-	<i>Enterococcus sp.</i>
5.	115	-	-	+	+	-	+	-	+	+	-	+	+	<i>Staphylococcus aureus</i>
6.	G57	-	+	+	-	+	+	+	+	-	-	-	-	<i>Bacillus subtilis</i>
7.	P10	+	-	+	-	-	+	+	+	+	-	-	+	<i>Bacillus cereus</i>
8.	IR2A	+	-	-	-	+	+	+	-	-	-	-	-	<i>Pseudomonas aeruginosa</i>
9.	AY2	+	-	+	-	+	+	-	-	AG	-	AG	+	<i>Proteus mirabilis</i>
10.	EJ2	+	-	+	+	-	+	-	-	+	+	+	AG	<i>Klebsiella sp.</i>

KEY: MR=Methyl Red test, VP= Voges-Proskauer test, - =Negative, + = Positive, AG = acid with gas production, Gram = Gram's reaction

Table 2 Phytochemical qualitative analysis of *G. latifolium* leaf extracts

EXTRACT	SAPONINS	FLAVONIDS	TANNINS	TERPENIDS	ALKALOIDS	PHENOLS	GLYCOSIDES	PHYTOSTEROLS
Aqueous	++	+++	-	+++	-	+	+	+
Ethanol	+	+	+	+	-	++	-	++
Methanol	+	+	+	++	-	++	-	++

KEY: Highly present = (+++), moderately present = (++), lightly present = (+), Absent = (-)

Antibiotic susceptibility tests were performed on four Gram-positive bacterial isolates using ten different conventional antibiotics. The results are shown in Table 3. Among the four isolates tested, all were resistant to at least one antibiotic, with varying patterns of susceptibility. *Bacillus subtilis* (G57) and *Bacillus cereus* (P10) exhibited moderate resistance to certain antibiotics but remained susceptible to others, such as ofloxacin (21 – 22 mm) and tetracycline (20 – 21 mm). The highest zone of inhibition (22 mm) was observed with ofloxacin against *Bacillus cereus* (P10), while the lowest zone (7 mm) was recorded for the isolate with cotrimoxazole and tetracycline. Figure1 presents the percentage resistance and susceptibility patterns of the Gram-positive isolates to the tested antibiotics following the CLSI 2023 standard (Clinical and Laboratory Standards Institute).

Six (6) Gram-negative isolates were tested against ten conventional antibiotics. The results are summarized in Table 4. Following the CSLI standard, several isolates displayed multi-drug resistance, with *Pseudomonas aeruginosa* (IR2A) showing complete resistance to some antibiotics, particularly to penicillin (0 mm), tetracycline (0 mm), and gentamycin (0 – 14 mm). The highest zone of inhibition (22 mm) was recorded for ofloxacin against *Pseudomonas aeruginosa* (IR2A), while the lowest was 0 mm for penicillin against multiple isolates.

Figure 2 shows the percentage susceptibility and resistance of the Gram-negative isolates. As shown in the figure, ofloxacin demonstrated the highest levels of effectiveness (90%), while penicillin showed the highest level of resistance (100%) across the isolates.

Table 3 Antibiotic Sensitivity Tests for Gram-positive Bacterial Samples

S/N	ISOLATE CODE	ISOLATE NAME	ZONE OF INHIBITION OF GRAM-POSITIVE ANTIBIOTIC DISCS (mm)									
			COT (25µg)	CEF (30µg)	AMX (25µg)	ERY (5µg)	CPX (10µg)	OFL (5µg)	AZY (12µg)	PEF (10µg)	LEV (5µg)	TET (10µg)
1.	119	<i>Enterococcus sp.</i>	21	19	21	15	19	20	7	18	17	8
2.	115	<i>Staphylococcus aureus</i>	20	19	15	10	17	21	9	18	19	18
3.	G57	<i>Bacillus subtilis</i>	14	11	7	20	18	21	16	16	19	21
4.	P10	<i>Bacillus cereus</i>	20	8	15	15	17	22	17	20	20	20

KEY: AMX = Amoxicillin, LEV = Levofloxacin, CEF = Ceftriazone, CPX = Ciprofloxacin, COT = Clotrimazole, PEF = Pefloxacin, ERY = Erythromycin, TET= Tetracycline, AZY= Azithromycin, OFL = Ofloxacin.

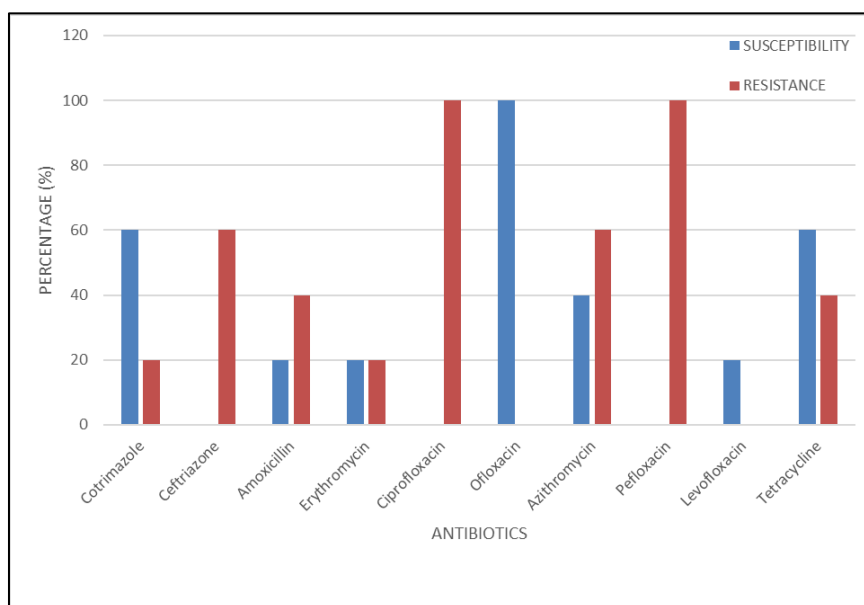


Figure 1 Percentage resistance and susceptibility of Gram-positive isolates to antibiotics

Table 4 Antibiotic sensitivity tests for Gram-negative organisms

S/N	ISOLATE CODE	ISOLATE NAME	ZONE OF INHIBITION OF GRAM-NEGATIVE ANTIBIOTIC DISCS (mm)									
			AMX (30µg)	LEV (5µg)	CEF (30µg)	CPX (30µg)	CHL (30µg)	PEN (10µg)	TET (30µg)	GEN (10µg)	STR (10µg)	OFL (5µg)
1.	AY1	<i>Citrobacter freundii</i>	15	16	17	17	16	0	18	17	9	19
2.	AY1A	<i>Acinetobacter baumannii</i>	20	18	18	17	10	0	16	21	8	18
3.	AY1B	<i>Citrobacter koseri</i>	19	19	18	19	21	10	16	14	9	17
4.	IR2A	<i>Pseudomonas aeruginosa</i>	9	18	18	17	18	0	0	0	0	22
5.	AY2	<i>Proteus mirabilis</i>	19	20	16	18	9	0	20	15	8	18
6.	EJ2	<i>Klebsiella sp.</i>	18	18	18	17	9	0	20	17	12	19

KEY: AMX = Ampiclox, LEV = Levofloxacin, CEF = Cefuroxime, CPX = Ciprofloxacin, CHL = Chloramphenicol, PEN = Penicillin, GEN = Gentamycin, TET= Tetracycline, STR= Streptomycin, OFL = Ofloxacin.

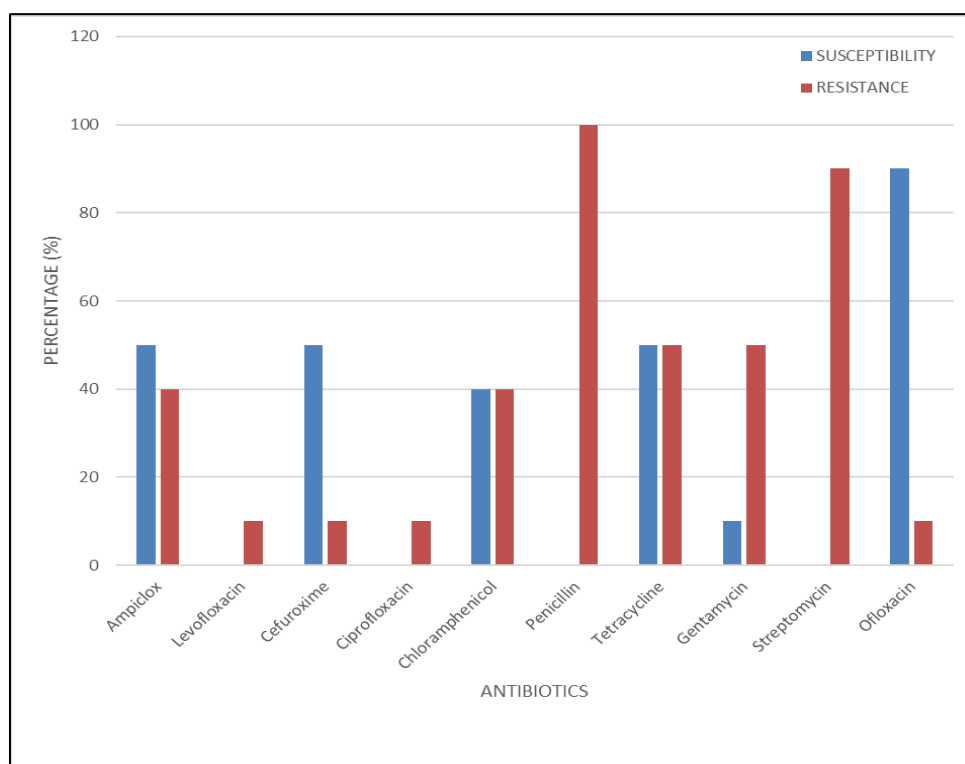
**Figure 2** Percentage resistance and susceptibility of Gram-negative isolates to antibiotics

Table 5 shows the result of the antibacterial activities of the three extracts, aqueous, ethanol and methanol leaf extracts, against ten selected bacterial isolates, which included both Gram-positive and Gram-negative organisms. The three extracts were tested in duplicates.

Among the three solvents, the methanol extract exhibited the highest overall antibacterial activity, producing notable zones of inhibition against multiple isolates, particularly for *Bacillus subtilis*, with an inhibition zone of 32 mm. The aqueous and ethanol extracts exhibited high inhibition zones (23 mm and 31 mm, respectively) for the same isolate. There was moderate activity across several other isolates (9 mm – 16 mm).

The lowest inhibition zone of the aqueous extract (0 mm) was seen in isolate AY1A (*Acinetobacter baumannii*) and 119 (*Enterococcus sp.*-0 mm). Low inhibition zone of the ethanol extract was observed in the isolates AY1A (*Acinetobacter baumannii*), 0 mm, and AY1 (*Citrobacter freundii*), 6 mm. A moderate level of inhibition (10 mm) each was observed against isolates AY1B (*Citrobacter koseri*), AY2 (*Proteus mirabilis*), 119 (*Enterococcus sp.*), and EJ2 (*Klebsiella sp.*) for the ethanol plant extract.

Among all the isolates tested, isolate 119 (*Acinetobacter baumannii*) exhibited the highest level of resistance (0-8 mm), as it showed the least susceptibility to all three extracts of *Gongronema latifolium* leaves. This suggests that the plant extracts had a minimal inhibitory effect on this particular organism compared to others.

Table 5 Result of Antimicrobial Susceptibility Testing of *G. latifolium* Plant Extracts Using the Agar-Well Diffusion Method.

S/n	Isolate code	Isolate name	Zone of inhibition of plant extract (mm)		
			AQUEOUS	ETHANOL	METHANOL
1.	AY1	<i>Citrobacter freundii</i>	11	6	10
2.	AY1A	<i>Acinetobacter baumannii</i>	0	0	8
3.	AY1B	<i>Citrobacter koseri</i>	8	10	12
4.	119	<i>Enterococcus sp.</i>	0	10	8
5.	115	<i>Staphylococcus aureus</i>	11	10	16
6.	G57	<i>Bacillus subtilis</i>	23	22	32
7.	P10	<i>Bacillus cereus</i>	10	11	12
8.	IR2A	<i>Pseudomonas aeruginosa</i>	16	12	12
9.	AY2	<i>Proteus mirabilis</i>	10	10	9
10.	EJ2	<i>Klebsiella sp.</i>	13	10	13

4. Discussion

The rapid emergence of antibiotic-resistant pathogenic bacteria is a serious threat to the management of infectious diseases. This has drawn the attention of the pharmaceutical and scientific communities towards researching into potential antimicrobial agents from natural sources such as plants. This study was therefore carried out to evaluate the antibacterial activity of the aqueous, ethanol, and methanol leaf extracts of *Gongronema latifolium* against selected Gram-positive isolates, including *Enterococcus sp.*, *Staphylococcus aureus*, *Bacillus subtilis*, and *Bacillus cereus*, and Gram-negative isolates, including *Citrobacter freundii*, *Citrobacter koseri*, *Acinetobacter baumannii*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Proteus mirabilis*.

Phytochemical analysis was conducted on all three extracts, revealing that *G. latifolium* leaves contain saponins, flavonoids, tannins, terpenoids, phenols, phytosterols, and glycosides. These phytochemicals have been reported to be responsible for the antibacterial activity of the plant [6, 10].

The antibacterial activity testing of *G. latifolium* proved effective against all test isolates provided by the laboratory, particularly *Bacillus subtilis*, which showed the highest inhibition zone (22 mm - 32 mm) for the extracts. Although *Bacillus subtilis* is considered relatively non-pathogenic and is often used as a probiotic in various dietary supplements due to its ability to promote intestinal health [11], its high sensitivity suggests that consumption of *G. latifolium* alongside *Bacillus subtilis*-based products may lead to antagonistic effects, thereby reducing the efficacy of either agent.

Staphylococcus aureus, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa* also showed sensitivity to all three extracts, agreeing with a study reported by Ndubueze *et al.* [12], who reported that the aqueous and ethanolic extracts of *Gongronema latifolium* have concentration-dependent inhibitory effects on the test organisms, *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*.

Proteus mirabilis showed susceptibility to all three extracts: aqueous, ethanol, and methanol, with the inhibition zone ranging from 8 mm to 10 mm for a 1 g/mL concentration of the plant extracts. This conforms to a study conducted by Omodamiro and Ekeleme [13], who reported that the ethanolic leaf extract showed significant dose-dependent inhibition of *Proteus mirabilis*, *Staphylococcus aureus*, *Streptococcus pneumoniae*, *E. coli*, and *Pseudomonas aeruginosa*.

In this study, the results obtained from the aqueous extracts showed that most bacterial isolates, except *Acinetobacter baumannii* and *Enterococcus sp.*, were sensitive to the extract at varying levels, with the highest inhibition zone of 32 mm observed for the extract against *Bacillus subtilis*. This agrees with the work of Chinyere *et al.* [14], which also reported that both aqueous and ethanolic extracts of *G. latifolium* possess antibacterial activity. The aqueous extract proved to be effective and does not agree with the report of Oshodi *et al.* [15], which states that the aqueous extract possesses no antimicrobial activity.

The antibacterial activity of the methanol extract of *G. latifolium* leaves had a greater effect on *Bacillus cereus* compared to other extracts, with the inhibition zones of 12 mm and above. This agrees with the study reported by Morebise *et al.* [16] that the saponins fraction of the methanolic extracts of *G. latifolium* strongly inhibited human pathogens including *Bacillus cereus*, *Staphylococcus aureus*, *Candida albicans* and *Aspergillus niger*.

The aqueous, ethanol, and methanol extracts had effect on isolate IR2A (*Pseudomonas aeruginosa*), with the inhibition zone ranging from 12 mm to 16 mm compared to the penicillin, gentamycin, Streptomycin, and tetracycline antibiotics discs, which had 0 mm inhibition zones. It was less effective compared to ofloxacin, levofloxacin, and cefuroxime, with susceptibility ranging from 18 mm to 22 mm in diameter.

Citrobacter freundii and *Citrobacter koseri* showed moderate to low susceptibility, with inhibition zones of 6 mm to 11 mm across the various *G. latifolium* extracts. There is limited research specifically on the effect of *G. latifolium* extracts on *Citrobacter sp.* This study thus provides new insight into the usefulness of *G. latifolium* extracts against *Citrobacter species*.

The aqueous, ethanol, and methanol extracts of *G. latifolium* were effective against Gram-positive and Gram-negative lab isolates with varying inhibition zones, which suggests that the plant possesses the potential for broad-spectrum antibacterial properties. These extracts were also effective against several antibiotic-resistant isolates, including penicillin-resistant bacterial isolates. This indicates that *G. latifolium* also displays therapeutic potential and may be considered in the one-health approach to combat drug-resistant microbes.

5. Conclusion

In conclusion, this study revealed that the aqueous, ethanol, and methanol leaf extracts of *Gongronema latifolium* have antibacterial activity against varieties of bacterial isolates. Therefore, the extracts can be used to treat/eradicate clinical infections caused by these isolates (*Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Proteus mirabilis*, and *Citrobacter sp.*). There was a substantial difference between the methanolic extract and the ethanol or aqueous extract, as the methanolic extract proved to be the most effective solvent used against most bacterial isolates.

5.1. Recommendations

This work suggests further research in the use of *G. latifolium* extract against *Citrobacter species* and also in the treatment of infections caused by different Gram-positive and Gram-negative bacteria. Pharmaceutical industries are encouraged to consider extracting and purifying active ingredients of *G. latifolium* in the production of antibiotics effective on drug-resistant bacteria.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors declare no conflict of interest.

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