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ANTIBACTERIAL POTENTIAL OF *Adansonia digitata* L. (Baobab) LEAF EXTRACTS AGAINST MULTIDRUG-RESISTANT BACTERIA

AYODEJI SUNDAY ADEDEJI[1*], OLUWATOSIN OMOBOLANLE OYEBIYI[2], ABIODUN OMOLARA AKINKUGBE[1], EMMANUEL PRAISE IDOGBE[1], TAYE MICHAEL ONIWAYA[1], BABAJIDE ENIOLURUNDA BAYO-AFOLABI[1]

1. Department of Biological Sciences, Faculty of Natural And Applied Science, Achievers University, Owo, Ondo State, Nigeria.
2. Department of Biochemistry, College of Medicine, Achievers University, Owo, Ondo State, Nigeria

ABSTRACT

The increasing prevalence of multidrug-resistant bacteria have turned bacterial infections into a significant global healthcare challenge, necessitating immediate action. This study evaluated the antibacterial activity of *Adansonia digitata* L. of leaf extracts against multidrug-resistant isolates, including *Pseudomonas* sp., *Klebsiella* spp., *Escherichia* sp., *Staphylococcus* sp., and *Bacillus* sp. The bacterial isolates were confirmed through morphological, cultural and biochemical characterization. Subsequently, bacterial isolates were subjected to antibiotic susceptibility testing (AST), and multiple antibiotic resistant (MAR) indices were determined. The leaves of *A. digitata* L. were extracted using aqueous and ethanol-based solvents. The antibacterial activity of the resulting plant extracts was evaluated at varying concentrations (100, 50, 25, and 12.5 mg/L). Determination of Minimum inhibitory concentrations (MIC) were carried out by broth dilution method, and the minimum bactericidal concentrations (MBC) were established by plating aliquots from non-turbid wells. The AST results revealed that all isolates were multidrug-resistant with varying susceptibility to some antibiotics. Extracts of *Adansonia digitata* L. demonstrated notable antibacterial effects against the tested organisms, with ethanolic extract showing greater activity. Ethanolic extract further demonstrated bactericidal activity against all tested organisms at concentrations between 40– 80mg/mL except for *Escherichia coli*, which exhibited resistance at various concentrations. In contrast, the aqueous extract demonstrated only bacteriostatic activity with no bactericidal effect observed. The significant antibacterial activity of *A. digitata* L. leaves against MDR organisms, particularly the bactericidal effect of the ethanolic extract, highlights their potential as a source of therapeutic agents for MDR pathogen infections.

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INTRODUCTION

Antibiotics have long served as the cornerstone for the management and treatment of infectious diseases caused by bacteria or fungi. However, in recent years, treatments have become a significant issue owing to the rise of antibiotic resistance [1, 2]. Bacterial infections constitute a formidable concern given the propensity of bacteria for horizontal acquisition and dissemination of resistance genes harbored on mobile genetic elements, including transposons and plasmids [3]. This process has contributed to the increasing prevalence of multidrug-resistant (MDR) bacteria, which constitute a significant global health problem. The World Health Organization (WHO) estimates that approximately

***Corresponding author:** as.adedeji@achievers.edu.ng; +234 810 394 9106
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700,000 deaths occur annually due to drug-resistant infections, with projections indicating a potential rise to 10 million in the absence of effective intervention [4]. Multidrug resistance occurs when microbes evolve mechanisms (such as genetic mutation, reduced permeability, enzymatic inactivation, and efflux pump production) that protect them from the effects of antibiotics.
This phenomenon is attributed to indiscriminate and excessive use of antimicrobial agents in both human and veterinary practices [5]. Consequences include the spread of resistant strains, therapeutic failure as common infections become increasingly difficult to manage or treat, increased healthcare costs due to prolong hospitals stays, increased mortality, and a greater demand for intensive care [4] (WHO, 2019).
One of the principal strategies for combating antibiotic resistance is the development of new drugs. However, the WHO has reported that declining private-sector investment and limited innovation in antibiotics development are undermining global efforts to combat drug-resistant infections [6]. Furthermore, the continuous and indiscriminate use of antibiotics has accelerated the emergence of resistance among various strains of microorganisms [7]. Consequently, it has become imperative to seek out alternative sources of antimicrobial agents that are effective, less toxic, and economically viable.

Natural products have gained considerable attention as potential alternatives due to their relative safety, accessibility, and long-standing use in traditional or folk medicine for addressing various diseases condition [8]. Plants, in particular, are rich in diverse phytoconstituents, with potential antimicrobial properties, making them a promising candidate for drug discovery.

Over 35,000-70,000 (14%-28%) plant species worldwide have been utilized for medicinal purposes [9]. Secondary metabolites such as phenol, flavonoids, alkaloids, tannin, and other bioactive compounds present in these plants suggest their potential as antibacterial agents. However, the limited investigation of many plant species underscores the urgent need for continued research into diverse plants with potential antibacterial properties. Expanding this exploration is essential for identifying novel bioactive compounds that could serve as effective alternatives in combating antimicrobial resistance.

Adansonia digitata L. (baobab) is a drought-resistant, multipurpose tree widely distributed across sub-Saharan Africa and valued for its nutritional and medicinal importance [10-12]. The plant parts, such as the fruit pulp, seeds, leaves, and bark are utilized both as food sources and in traditional medicinal practices. The fruit pulp is especially rich in vitamin C, minerals, and dietary fiber, while the leaves are commonly consumed as vegetables [13,14].

Traditionally, this plant has been employed in the management of various ailments, including fever, diarrhea, malaria, and dysentery [15,16]. These therapeutic effects are attributed to its phytochemical constituents, which suggest its use as an antimicrobial agent. This study therefore examined the antibacterial potential of leaf extracts of *Adansonia digitata* L. against some selected multidrug-resistant bacteria.

MATERIALS AND METHODS

Plant Collection and Identification

Fresh leaves of *Adansonia digitata* L. were harvested and excised using a sterile knife from a mature tree in Owo, Ondo State, Nigeria. The tree was taxonomically identified by a Plant Scientist in the Department of Botany, Obafemi Awolowo University, Ile Ife, Nigeria, with voucher number of IFE 18520. The leaves were air dried, pulverized using a mechanical grinder and preserved in airtight containers prior to use.

Extraction of Plant Materials

The maceration method was employed using two solvents of differing polarity, namely ethanol and distilled water. For the ethanolic extraction, 100 g of powdered leaves was soaked in 800 mL of ethanol in a sterile conical flask, while an equal quantity of the powdered leaves was macerated in 1500 mL of distilled water to obtain the aqueous extract. The mixtures were intermittently agitated and kept at ambient temperature for 72 h to enhance the extraction of phytochemical constituents. Both aqueous and ethanolic extracts were obtained using muslin cloth and Whatman No.1 filter respectively [17]. The resulting filtrates were concentrated by solvent evaporation in a thermostatically controlled water bath at 40°C, and the dried extracts obtained were subsequently preserved for further analysis.

Preparation of Stock Solution of Extracts
Stock solution (200mg/mL) of the crude extract was prepared in 10% v/v DMSO. The stock was serially diluted two-fold with sterile distilled water to obtain concentrations of 100, 50, 25, and 12.5 mg/mL. Dimethyl sulfoxide (DMSO) at the corresponding concentrations served as the negative control for antibacterial activity [18].

Source and Collection of Test Organisms
Test organisms were procured from the Department of Biological Sciences, Achievers University, Owo. The isolates comprised *Pseudomonas* sp.*, Klebsiella* spp., *Escherichia* sp.*, Staphylococcus* sp.*,* and *Bacillus* sp.

Confirmation of Test Organisms
Macroscopic and Microscopic Examination
Isolates were subcultured on nutrient agar plates and incubated at 37^0C for 24 h. Following incubation, the colonies were examined for morphological characteristics such as size, colour, texture, pigmentation, consistency, and odour. Thereafter, Gram staining was performed, and the staining smears were examined under the microscope [19].

Biochemical Identification of Isolates
The methods described by [19,20] were adopted for the biochemical re-characterization and identification of the procured organisms. The biochemical tests, including catalase, motility, sugar fermentation, triple sugar iron (TSI), methyl red, indole, Voges–Proskauer, urease, coagulase, oxidase, and citrate utilization were carried out.
Standardization of Inoculum
The 0.5 McFarland standard was obtained by mixing 0.5ml of 1% barium chloride with 9.5mL of 1% sulphuric acid [21]. For inocula standardization, bacterial isolates were subcultured on Muller-Hinton agar and incubated overnight at 36^0C. Fresh colonies were then inoculated on Muller-Hinton broth (HIMedia, India) and the inoculum density was standardized to correspond to McFarland standard (1.5×10^8 CFU/mL) [22].

Antibiotic Susceptibility Testing
The Kirby–Bauer disk diffusion technique was employed, following the standardized guidelines of the Clinical and Laboratory Standards Institute [23]. Briefly, standardized bacterial suspensions were inoculated onto Muller-Hinton agar (Ready MED, India) plate, after which commercially prepared antibiotic discs were aseptically placed on it. The antibiotics evaluated in this study against the bacterial isolates were manufactured by Celtec diagnostic (Belgium), comprising amoxicillin (30 µg), cefotaxime (25 µg), gentamicin (10 µg), nitrofurantoin (30 µg), nalidixic acid (30 µg), ofloxacin (5 µg), cefuroxime (30 µg), imipenem/cilastatin (10/10 µg), ceftriaxone/sulbactam (30 µg), cefixime (5 µg), ampiclox (10 µg), levofloxacin (5 µg), erythromycin (15 µg), and azithromycin (15 µg).

Determination of Multidrug Resistance and Multiple Antibiotic Resistance Index
Multidrug-resistant was defined as bacterial isolate that showed resistant to at least one agent in three or more antimicrobial categories [24]. Additionally, the multiple antibiotic resistance (MAR) index was calculated as MAR = a/b, where “a” denotes the number of antibiotics to which the isolate exhibited resistant, and “b” denotes the total number of antibiotics tested [25].

Antibacterial Susceptibility Assay
The antibacterial potential of the extracts was assessed using the agar well diffusion technique. Standardized inocula of confirmed multidrug-resistant (MDR) bacterial isolates were uniformly inoculated on Muller–Hinton agar plates. The plates were allowed to sit for a few minutes to allow inoculum absorption. Using a sterile cork borer, five wells, each approximately 6 mm in diameter, were aseptically punched into the agar. Aliquots of 50 µL of the different extract concentrations (100, 50, 25, and 12.5 mg/L) were carefully dispensed into the respective wells. Antibacterial activity was evaluated after incubation at 36^0C for 24 h by measuring the inhibition zone around the wells in millimeters (mm) [26].

Evaluation of the Minimum Inhibitory Concentration (MIC) of the Test Extract
The minimum inhibitory concentration (MIC) of the plant extract was determined using the broth dilution technique with minor modifications. A stock solution of 100 mg/mL was serially diluted with Mueller–Hinton broth to obtain desired concentrations. In brief, 4.0, 3.0,2.0,1.0 and 0.5mL of the stock solution were aseptically mixed with 1.0, 2.0, 3.0, 4.0, 4.5mL sterile broth respectively to achieve concentrations of 80, 60, 40, 20, and 10 mg/mL, with each test tube adjusted to a final volume of 5 mL. Each tube was inoculated with 0.5 mL of standardized bacterial suspension equivalent to the 0.5 McFarland standard. A tube containing only Mueller–Hinton broth served as the negative control. The tubes were examined for turbidity after

incubation at 36^0 C for 24 h. The MIC was recorded as the lowest concentration of the extract that showed no visible bacterial growth [27,28].

Evaluation of the Minimum Bactericidal Concentration (MBC)

The MBC of the extract was determined to evaluate its bactericidal activity. Briefly, 0.1 mL aliquots from MIC tubes showing no visible bacterial growth and were aseptically subcultured onto freshly prepared Mueller–Hinton agar plates. After incubation at 37°C for 24 h, the plates were observed for bacterial growth. The MBC was taken as the lowest concentration of the extract that produced no visible colony growth or resulted in at least 99% reduction of the initial bacterial population [27].

Statistical Analysis

All experiments were carried out in triplicate, and the results obtained were expressed as the mean ± standard deviation. The data were graphically represented and statistically analyzed using analysis of variance (ANOVA), followed by Tukey's HSD post hoc test. Statistical significance was established at $p < 0.05$ using IBM SPSS Statistics Version 20.

RESULTS

Confirmation and Identification of Bacterial Isolates

Morphological Characteristic of Bacterial Isolates

The six (6) selected bacterial isolates investigated in this study showed different morphological characteristics when sub-cultured on Nutrient Agar as presented in Table 1. The differences are evident in terms of colony shape, surface texture, opacity, margin, elevation, and pigmentation, indicating the presence of phenotypically diverse bacterial populations.

Biochemical Characterization

The cellular morphology, Gram staining reaction, and biochemical characterization of the six bacterial isolates suggest their classification into five probable groups, namely *Pseudomonas* sp. (Ak1), *Escherichia* sp. (Ak2), *Klebsiella* spp. (Ak3 and Ak4), *Staphylococcus* sp. (Ak5), and *Bacillus* sp. (Ak6) (Table 2).

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Gram staining revealed that isolates Ak1–Ak4 were Gram-negative, appearing as pink-stained rods under the microscope. In contrast, isolates Ak5 and Ak6 were Gram-positive, retaining the crystal violet stain and appearing purple.

The biochemical profiles further supported these groupings. Isolate Ak1 displayed an oxidase-positive reaction, a feature commonly associated with *Pseudomonas* species, alongside metabolic versatility typical of this group. Isolate Ak2 demonstrated lactose fermentation and indole production, characteristics widely used in the presumptive identification of *Escherichia coli*. Isolates Ak3 and Ak4 were lactose fermenters and exhibited mucoid growth, suggestive of *Klebsiella* species. Isolate Ak6 was identified as a Gram-positive, rod-shaped bacterium that was catalase-positive, consistent with characteristics generally attributed to *Bacillus* species. In contrast, isolate Ak5 exhibited Gram-positive cocci morphology and tested positive for catalase, aligning with features commonly observed in *Staphylococcus* species.

Antibiotic Susceptibility Profile or Patterns

The antibiotic susceptibility and resistance profiles of the Gram-negative and Gram-positive isolates are presented in Tables 3 and 4, respectively. Classification of the isolates as resistant (R), susceptible (S) or intermediate (I) was based on criteria outlined by CLSI. The findings revealed that all isolates exhibited multidrug resistance, with MAR index values ≥ 0.2.

All Gram-negative isolates demonstrated complete resistance to amoxicillin, ceftriaxone, cefotaxime, nalidixic acid, imipenem, ampiclox, cefuroxime, and cefixime. However, they exhibited susceptibility to ofloxacin. The MAR indices of these isolates were generally high, with *Escherichia* sp. exhibiting the highest level of resistance (MAR index values of 0.83).

In contrast, the Gram-positive isolates demonstrated relatively lower MAR index values. *Staphylococcus* sp. and *Bacillus* sp. had MAR indices of 0.67 and 0.33, respectively. Both isolates were resistant to amoxicillin, cefoaxime, erythromycin, gentamycin, while remaining susceptible to azithromycin, ofloxacin, and levofloxacin

Table 1: Morphological Characteristics of Bacterial Isolates

Isolates Codes	Colony Shape	Elevation	Margin	Surface/Texture	Color/Pigmentation	Opacity
Ak1	Irregular	Flat	Undulate	Smooth, moist	Greenish/blue-green	Translucent
Ak2	Circular	Convex	Entire	Smooth, moist	Cream/off-white	Opaque
Ak3	Circular	Convex	Entire	Mucoid, smooth	Cream	Opaque
Ak4	Circular	Convex	Entire	Highly mucoid	Cream	Opaque
Ak5	Circular	Convex	Entire	Smooth, glistening	Golden yellow/cream	Opaque
Ak6	Irregular	Flat/raised	Lobate/undulate	Rough, dry	Whitish	Opaque

Table 2: Biochemical Characteristics of Bacterial Isolates

Isolates Codes	Gram reaction	Cellular Morphology	Indole	Methy Red (MR)	Voges Proskauer (VP)	Citrate	Motility	Urease	Catalase	Oxidase	Triple Sugar Iron		Lactose Fermentatio	H_2S	Gas	Probable organisms
											Slant	Butt				
Ak1	-ve	R	-	-	-	+	+	-	+	+	Alk	Alk	-	-	+	*Pseudomonas* sp
Ak2	-ve	R	+	+	-	-	+	-	+	-	Acid	Acid	+	-	+	*Echerichia* sp.
Ak3	-ve	R	-	-	+	+	-	+	+	-	Acid	Acid	+	-	+	*Klebsiella* sp
Ak4	-ve	R	-	-	+	+	-	+	+	-	Acid	Acid	+	-	+	*Klebsiella* sp
Ak5	+ve	C	-	+	+	+	-	+	-	-	N/A	N/A	+	-	-	*Staphylococcus* sp
Ak6	+ve	R	-	-	+	+	+	-	+	-	Acid	Acid	-	-	-	*Bacillus* sp

Key : N/A - Not Applicable

Table 3: Antibiotic Susceptibility and Resistance Profile of Gram-Negative Isolates

Isolates Codes	AUG	CTX	IMP	OFX	GN	NA	NF	CXN	CRO	ACX	ZEM	LBC	MAR Indices	Probable Organisms
Ak1	0R	0R	0R	18S	7R	0R	15I	0R	10R	0R	0R	15I	0.75	*Pseudomonas* sp.
Ak2	0R	0R	0R	16S	0R	0R	11R	0R	0R	0R	07R	0R	0.83	*Echerichia* sp.
Ak3	0R	0R	07R	18S	14I	0R	13R	0R	0R	0R	0R	20S	0.75	*Klebsiella* sp.
Ak4	0R	0R	0R	18S	7R	0R	15I	0R	10R	0R	0R	15I	0.75	*Klebsiella* sp.

Key: AUG- Amoxicillin, IMP- Imipenem/Cilastatin, CTX- Cefotaxime, OFX- Ofloxacin, GN- Gentamicin, NA- Nalidixic Acid, NF- Nitrofurantoin, CXN- Cefuroxime, CRO- Ceftriaxone-Sulbactarm, ACX- Ampiclox, ZEM -Cefexime, LBC- Levofloxacin

Table 4: Antibiotic Susceptibility and Resistant Profile of Gram-Positive Isolates

Isolates Codes	AUG	CTX	ZEM	CRO	LBC	IMP	CIP	CXM	OFX	ERY	GN	AZN	MAR Index	Probable Organisms
Ak5	0R	0R	0R	0R	18S	0R	11R	14I	18S	0R	0R	21S	0.67	*Staphylococcus* sp
Ak6	11R	13R	16I	19I	18S	17S	18S	15I	20S	0R	0R	18S	0.33	*Bacillus* sp

Key: AUG- Amoxicillin clavulanate, CTX- Cefoxime, CRO - Cefriaxone-Subtazosin, ZEM - Cefexime, LBC-Levofloxacin, CIP- Ciprofloxacin, MP- Imipenem/Cilastatin, CXM- Cefuroxime, OFX- Ofloxacin, ER - Erythromycin, GN- Gentamycin, AZN- Azithromycin

Antibacterial Activity of the Ethanolic Extract of Baobab

The ethanolic extract exhibited antibacterial activity against the test organisms, with inhibitory effects increasing progressively as the extract concentration increased (Figure 1). For instance, at the highest concentration (100 mg/mL), all isolates were susceptible, with zones of inhibition ranging from 14 ± 0.8 mm to 24 ± 1.4 mm, indicating the strong antibacterial potential of the extract. However, a progressive reduction in diameter of inhibition zones was observed across all bacterial isolate, as concentration decreases (50, 25, and 12.5mg/mL). However, as the concentration decreased (50, 25, and 12.5 mg/mL), a progressive reduction in the diameter of Among the tested isolates, *Staphylococcus* sp. exhibited the highest susceptibility at 100 mg/mL (24 ± 1.4 mm), followed closely by *Bacillus* sp. (22 ± 1.3 mm) and *Pseudomonas* sp. (22 ± 1.5 mm). Moderate activity was observed against *Klebsiella* sp. [AK3] (16 ± 0.6 mm), *Escherichia* sp. (15 ± 1.2 mm), and *Klebsiella* sp. [AK4] (14 ± 0.8 mm). At the least concentration (12.5 mg/mL), antibacterial activity was retained in some isolates such as *Pseudomonas* sp., *Klebsiella* sp. [AK3]. *Staphylococcus* sp., and *Bacillus* sp., although with reduced efficacy. In contrast, *Escherichia* sp. and *Klebsiella* sp. [AK4] showed no susceptibility, suggesting a higher resistance threshold.

Antibacterial Activity of Aqueous Extract of Baobab

The antibacterial activity of the aqueous extract against the tested bacterial isolates is presented in Figure 2. The results showed a concentration-dependent inhibitory effect across all susceptible isolates.

At the highest concentration (100 mg/mL), all isolates exhibited measurable inhibition zones. *Pseudomonas* sp. and *Bacillus* sp. showed similar levels of activity (14 ± 2.0 mm and 14 ± 1.4 mm, respectively), while *Staphylococcus* sp. exhibited the highest inhibition (15 ± 1.6 mm). Moderate activity was observed for *Klebsiella* sp. [AK4] (9 ± 1.5 mm) and *Klebsiella* sp. [AK3] (8 ± 0.8 mm), whereas *Escherichia* sp. showed the least susceptibility (6 ± 0.4 mm).

At 50 mg/mL, a reduction in antibacterial activity was observed. *Staphylococcus* sp. and *Bacillus* sp. maintained relatively higher inhibition (10 ± 1.2 mm and 10 ± 1.3 mm, respectively), followed by *Pseudomonas* sp. (9 ± 0.3 mm). Lower inhibition zones were recorded for *Klebsiella* sp. [AK4] (7 ± 1.7 mm) and *Klebsiella* sp. [AK3] (6 ± 1.2 mm), while *Escherichia* sp. exhibited no inhibition. At 25 mg/mL, only *Pseudomonas* sp. (6 ± 0.6 mm), *Staphylococcus* sp. (8 ± 1.6 mm), and *Bacillus* sp. (7 ± 1.7 mm) showed measurable inhibition, whereas *Klebsiella* species [AK3 and Ak4] and *Escherichia* sp. showed no inhibition. At 12.5 mg/mL, no inhibition was observed in any of the isolates.

Minimum Inhibitory Concentration

The minimum inhibitory concentrations (MICs) of both ethanolic and aqueous extracts of *Adansonia digitata* are presented in Table 5. The ethanolic extract demonstrated greater antibacterial activity compared to the aqueous extract. Gram positive bacteria, including *Bacillus* sp. and *Staphylococcus* sp. exhibited the same MIC value of 20 mg/mL, indicating higher susceptibility. Gram-negative bacteria, including *Klebsiella* species [AK3 and AK4], and *Pseudomonas* sp., recorded an MIC of 40 mg/mL, while *Escherichia coli* had MIC of 60mg/mL, reflecting moderate susceptibility.

Minimum Bactericidal Concentration

The ethanolic extract displayed a notable bactericidal effect against the test organisms at varying concentrations. *Staphylococcus* sp., *Bacillus* sp., and *Pseudomonas* sp. were completely killed at 40 mg/mL, indicating strong bactericidal efficacy at this concentration. In addition, *Klebsiella* sp. [AK3] and *Pseudomonas* sp. exhibited bactericidal activity at 60 mg/mL, while *Klebsiella* sp. [AK4] required a higher concentration of 80 mg/mL for a complete bactericidal effect. However, *Escherichia* sp. remained resistant to the bactericidal activity of the ethanolic extract across all tested concentrations. Conversely, the aqueous extract exhibited no bactericidal effect against any of the tested isolates, as all organisms were resistant at the concentrations evaluated (Table 6).

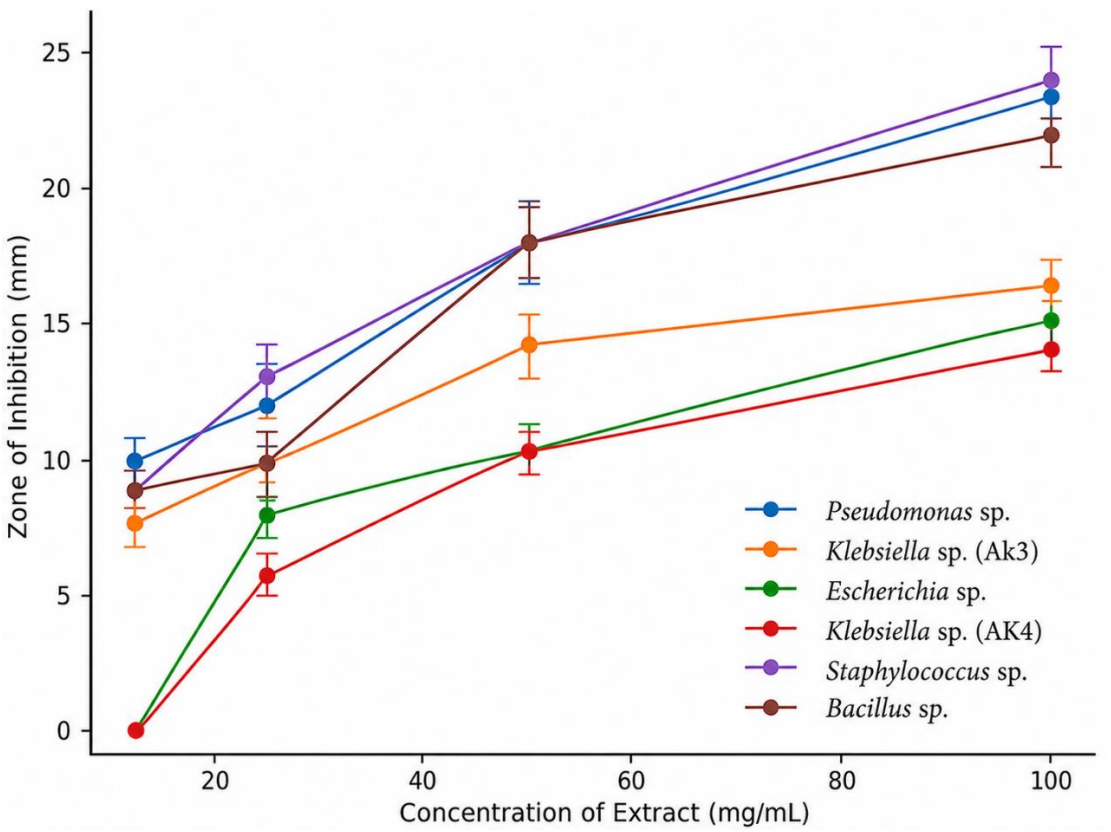


Figure 1: Antibacterial activity of ethanolic leaf extract at varying concentrations

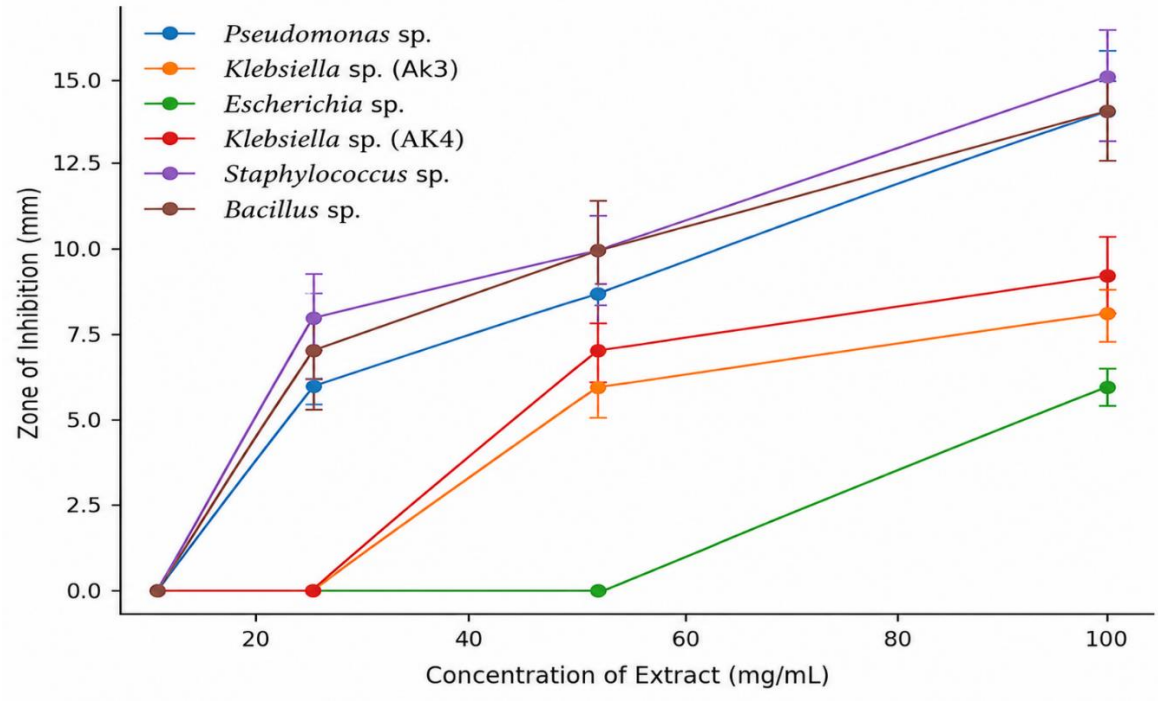


Figure 2: Antibacterial activity of aqueous leaf extract at varying concentrations

Table 5: Minimum Inhibitory Concentration for Baobab ethanolic extract

Bacterial Isolates	Minimum Inhibitory Concentration (mg/ml)	
	Ethanol	Aqueous
Pseudomonas sp.	40	60
Klebsiella sp. [AK3]	40	60
Escherichia sp.	60	-
Klebsiella sp. [AK4]	40	80
Staphylococcus sp.	20	40
Bacillus sp.	20	40

Key: no effect (-)

Table 6: Minimum Bactericidal Concentration of Baobab Extract

Bacterial Isolates	Minimum Bactericidal Concentration (mg/ml)	
	Ethanol	Aqueous
Pseudomonas sp.	60	-
Klebsiella sp. [AK3]	60	-
Escherichia sp.	-	-
Klebsiella sp. [AK4]	80	-
Staphylococcus sp.	40	-
Bacillus sp.	40	-

Key: no effect (-)

DISCUSSION

The increasing prevalence of multidrug-resistant (MDR) bacteria has turned out to be a serious and growing global challenge. Although these pathogens were once considered rare, they have now become widespread across many regions of the world, contributing significantly to rise in disease burden and treatment failure. In the present study, antibiotic susceptibility testing (AST) confirmed that the selected isolates (Gram-negative and Gram-positive) were multidrug-resistant (MAR indices ≥ 0.2), with Gram-negative bacteria exhibiting a significantly greater propensity for resistance. This result aligns with the study of [29], who observed that Gram-positive bacteria exhibit higher susceptibility to antibiotics compared with Gram-negative bacteria. Similarly, hospital-based studies have documented a greater prevalence of multidrug resistance among Gram-negative pathogens compared to Gram-positive isolates [30,31].

The resistance observed among Enterobacteriaceae isolates in this study agrees with the findings of [32], who documented complete (100%) resistance of Enterobacteriaceae to several commonly used antibiotics, including vancomycin, ampicillin, carbenicillin, methicillin, clindamycin, erythromycin, tetracycline, and trimethoprim/sulfamethoxazole. Likewise, the resistance profile of Gram-positive isolates corelates with [33], who documented multidrug resistance in 34.5% of *Staphylococcus* species, including resistance to aminoglycosides, fluoroquinolones, and β-lactam antibiotics.

The increasing trend of antibiotic resistance is predominantly driven by the overuse and misuse of antibiotics, resulting in treatment failure, increased morbidity, and mortality. This situation underscores the urgent need for alternative therapeutic options. Plant-derived compounds have therefore gained increasing attention due to their diverse phytochemical constituents and reported antimicrobial properties [34-36].

In the present study, baobab (*Adansonia digitata*) leaf extracts demonstrated notable antibacterial activity against the tested organisms, with both ethanolic and aqueous extracts showing inhibitory effects, although the ethanolic extract was consistently more effective. This finding is in agreement with [37], who reported similar activity against *Staphylococcus aureus* and *Escherichia coli*, and with [38] Magashi *et al*. (2018), who observed inhibition of several clinical isolates, including *Klebsiella pneumoniae*, *Staphylococcus aureus, Proteus mirabilis*, *Pseudomonas aeruginosa*, and *Streptococcus pneumoniae*, except for *E. coli*, which showed resistance. Furthermore, [39] reported that the leaf and pulp extract extracts of baobab exhibited strong antibacterial activity against *Enterococcus faecalis*, and in some cases, surpassed conventional antibiotics.

The observed antibacterial effects may be linked to its bioactive constituents such as phenolics, flavonoids, alkaloids, tannins, saponins, and terpenoids, which are widely recognized for their antimicrobial properties [40,35]. In addition, the higher efficacy of the ethanolic extract compared to the aqueous extract was observed. This may be due to the efficiency of ethanol in extracting a wider range of bioactive compounds, particularly non-polar constituents responsible for antimicrobial activity [41]. Statistical analysis further confirmed that the antibacterial activity was significantly affected by both extract type and the concentration used ($p < 0.05$).

In addition, the extract exhibited strong activity against Gram-positive bacteria including *Bacillus* sp. and *Staphylococcus* sp. than against Gram-negative bacteria, probably because of variation in their cell wall architecture. Gram-negative bacteria have an outer lipopolysaccharide membrane that functions as a barrier, limiting the penetration of antimicrobial agents and thereby contributing to their higher level of resistance [42]. Among the Gram-negative isolates, *Escherichia* sp. and *Klebsiella* sp. [AK4] exhibited higher resistance, particularly at lower concentrations, while *Pseudomonas* sp. showed comparatively moderate susceptibility, suggesting partial effectiveness of the extract against its intrinsic resistance mechanisms. The absence of activity at lower concentrations, especially in the aqueous extract, indicates sub-inhibitory levels insufficient to suppress bacterial growth.

Minimum inhibitory concentration (MIC) results further confirmed the superior efficacy of the ethanolic extract, as evidenced by generally lower MIC values across isolates. Gram-positive bacteria recorded lower MIC values (20-40 mg/mL), indicating higher susceptibility, in contrast, higher MIC values were observed in Gram-negative bacteria, which may be due to permeability barriers and efflux mechanisms [43,44]. *Escherichia* sp. showed the highest level of resistance, with no observable inhibition in the aqueous extract, suggesting an MIC greater than 80 mg/mL, consistent with reports of efflux pump activity and membrane impermeability in this organism [45]. Similarly, *Klebsiella* species [AK3 and AK4] exhibited elevated MIC values, likely due to capsule

formation and biofilm production [46]. Minimum bactericidal concentration (MBC) results showed that the ethanolic extract possessed appreciable bactericidal activity, particularly against Gram-positive bacteria, with MBC values as low as 40 mg/mL for *Staphylococcus* sp. and *Bacillus* sp. In contrast, Gram-negative bacteria exhibited higher MBC values or no bactericidal response within the tested range, highlighting their greater intrinsic resistance. The aqueous extract exhibited no bactericidal activity against the tested isolates, suggesting a predominantly bacteriostatic effect. This might be explained by the limited solubility of active compounds in polar solvents like water [17].

The observed differences between MIC and MBC values indicate that higher concentrations are required for bactericidal effects than for growth inhibition, which is consistent with established antimicrobial principles [18,47].

The ability of baobab leaf extracts, particularly the ethanolic fraction, to inhibit multidrug-resistant isolates is noteworthy, and suggest their potential application in drug development

CONCLUSION

The results of this study provide scientific evidence on the antibacterial activity of the leaf extracts of Baobab (*Adansonia digitata* L.) against multidrug-resistant bacteria, indicating its potential use as an alternative antibacterial agent. Future studies should therefore focus on evaluating *Adansonia digitata* leaf extracts on broader range of bacterial strains, analyzing their phytochemical composition, and determining their toxicity and safety profile.

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AUTHORS' CONTRIBUTION

ASA and OOO conceptualized the study. OOO and BEB handled plant identification and preparation. ASA, EPI, and TMO carried out the laboratory analysis. ASA wrote the manuscript, AOA reviewed it, and all authors proofread the final article.

CONFLICT OF INTEREST

The authors declared that there was no conflict of interest associated with this study.

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REFERENCES

1. Rice LB. The clinical consequences of antimicrobial resistance. Current Opinion in Microbiology, 12(5), 2009: 476–81.
2. Yadav S, Rawal G, Baxi M. An overview of infectious diseases. Journal of Community Health Management, 3(1), 2016: 41–3.
3. Partridge SR, Kwong SM, Firth N, Jensen SO. Mobile genetic elements associated with antimicrobial resistance. Clinical Microbiology Reviews, 31(4), 2018: e00088-17. DOI: 10.1128/CMR.00088-17.
4. World Health Organization. New multi-partner trust fund launched to combat antimicrobial resistance globally. [Accessed: 15 June, 2026]. Available: https://www.who.int/news/item/19-06-2019-new-multi-partner-trust-fund-launched-to-combat-antimicrobial-resistance-globally.
5. World Health Organization, 2023. Antimicrobial resistance. [Accessed: 15 June, 2026]. Available: https://www.who.int/news-room/fact-sheets/detail/antimicrobial-resistance.
6. World Health Organization, 2020. Lack of new antibiotics threatens global efforts to contain drug-resistant infections, World Health Organization, Geneva. [Accessed: 15 June, 2026]. Available: https://www.who.int/news/item/17-01-2020-lack-of-new-antibiotics-threatens-global-efforts-to-contain-drug-resistant-infections..
7. Gelband H, Miller-Petrie M, Pant S, Gandra S, Levinson J, Barter D, et al. The state of the world's antibiotics 2015. Center for Disease Dynamics, Economics & Policy, Washington, DC, 2015. [Accessed: 15 June, 2026]. Available: https://onehealthtrust.org/publications/reports/state-of-worlds-antibiotics-2015-2/
8. Caballero-Gallardo K, Quintero-Rincón P, Olivero Verbel J. Aromatherapy and essential oils: holistic strategies in complementary and alternative medicine for integral wellbeing. Plants, 14(3), 2025: 400. DOI: 10.3390/plants14030400.
9. Ullah I, Akhtar S, Adnan M, et al. Ethnobotanical knowledge of plants in Pakistan. Journal of Ethnobiology and Ethnomedicine, 21(1), 2025: 13.
10. De Caluwé E, Halamová K, Van Damme P. *Adansonia digitata* L. – A review of traditional uses, phytochemistry and

pharmacology. Afrika Focus, 23(1), 2010: 11–51. DOI: 10.1163/2031356X-02301005.
11. Gebauer J, El-Siddig K, Ebert G. Baobab (*Adansonia digitata* L.): A review on a multipurpose tree. Gartenbauwissenschaft, 67, 2002: 155–60.
12. Kamatou GPP, Vermaak I, Viljoen AM. Review of *Adansonia digitata*. South African Journal of Botany, 77(4), 2011: 908–19.
13. Chadare FJ, Linnemann AR, Hounhouigan JD, Nago MC, Van Boekel MAJS. Baobab food products: A review on their composition and nutritional value. Critical Reviews in Food Science and Nutrition, 49(3), 2009: 254–74. DOI: 10.1080/10408390701856330.
14. Rahul J, Jain MK, Singh SP, Kamal RK, Anuradha, Naz A, Gupta AK, Mrityunjay SK. *Adansonia digitata* L. (Baobab): A review. Asian Pacific Journal of Tropical Biomedicine, 5(1), 2015: 79–84.
15. Braca A, Sinisgalli C, De Leo M, Muscatello B, Cioni PL, Milella L, Ostuni A, Giani S, Sanogo R. Phytochemical profile, antioxidant and antidiabetic activities of *Adansonia digitata* L. (Baobab) from Mali, as a source of health-promoting compounds. Molecules, 23(12), 2018: 3104. DOI: 10.3390/molecules23123104.
16. Iyamah PC, Idu M. Ethnomedicinal survey of plants used in malaria treatment. Journal of Ethnopharmacology, 173, 2015: 287–302.
17. Tiwari P, Kumar B, Kaur M, Kaur G, Kaur H. Phytochemical screening and extraction: A review. International Pharmaceutical Sciences, 1(1), 2011: 98–106.
18. Clinical and Laboratory Standards Institute. Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically, 11th Ed., CLSI Standard M07-A11, Clinical and Laboratory Standards Institute, Wayne, PA, 2018.
19. Cheesbrough M. District Laboratory Practice in Tropical Countries, 2nd Ed., Cambridge University Press, Cambridge, 2010.
20. Negera E, Geritu N, Mulugeta K. Microbiological assessment of drinking water. African Journal of Microbiology Research, 11(6), 2017: 254–63.
21. Cheesbrough M. District laboratory practice in tropical countries. International Journal of Medical Sciences, 1(1), 2018: 65–8.
22. Oyeleke SB, Manga SB. Essentials of Laboratory Practicals in Microbiology, Tobest Publishers, Minna, 2008.
23. Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing, 33rd Ed., Clinical and Laboratory Standards Institute, Wayne, PA, 2023.
24. Magiorakos AP, Srinivasan A, Carey RB, Carmeli Y, Falagas ME, Giske CG, Harbarth S, Hindler JF, Kahlmeter G, Olsson-Liljequist B, Paterson DL, Rice LB, Stelling J, Struelens MJ, Vatopoulos A, Weber JT, Monnet DL. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: An international expert proposal for interim standard definitions for acquired resistance. Clinical Microbiology and Infection, 18(3), 2012: 268–81. DOI: 10.1111/j.1469-0691.2011.03570.
25. Osundiya OO, Oladele RO, Oduyebo OO. Multiple antibiotic resistance (MAR) indices of *Pseudomonas* and *Klebsiella* species isolates in Lagos University Teaching Hospital. African Journal of Clinical and Experimental Microbiology, 14(3), 2013: 164–8.
26. Ajijolakewu KA, Awarun FJ. Comparative antibacterial efficacy of *Vitellaria paradoxa* (Shea Butter Tree) extracts against some clinical bacterial isolates. Notulae Scientia Biologicae, 7(3), 2015: 264–8. DOI: 10.15835/nsb739617.
27. Ali M, Yahaya A, Zage AU, Yusuf ZM. In-vitro antibacterial activity and phytochemical screening of *Psidium guajava* on some enteric bacterial isolates of public health importance. Journal of Advances in Medicine and Pharmaceutical Sciences, 12, 2017: 1–7.
28. Yusha'u M, Hamza MM, Abdullahi N. Antibacterial activity of *Adansonia digitata* stem bark. International Journal of Biomedical and Health Sciences, 6(3), 2010: 129–35.
29. Alhumaid S, Al Mutair A, Al Alawi Z, Alzahrani AJ, Tobaiqy M, Alresasi AM, Bu-Shehab I, Al-Hadary I, Alhmeed N, Alismail M, Aldera AH, AlHbabi F, Al-Shammari H, Rabaan AA, Al-Omari A. Antimicrobial susceptibility of Gram-

positive and Gram-negative bacteria: A 5-year retrospective analysis at a multi-hospital healthcare system in Saudi Arabia. Annals of Clinical Microbiology and Antimicrobials, 20(1), 2021: 43. DOI: 10.1186/s12941-021-00450-x.

30. Leal HF, Azevedo J, Silva GEO, Amorim AML, de Roma LRC, Arraes ACP, Gouveia EL, Reis MG, Mendes AV, de Oliveira Silva M, Barberino MG, Martins IS, Reis JN. Bloodstream infections caused by multidrug-resistant Gram-negative bacteria: epidemiological, clinical and microbiological features. BMC Infectious Diseases. 2019;19(1):609. https://doi.org/10.1186/s12879-019-4265-z
31. Zhu X, Tong A, Wang D, Sun H, Chen L, Dong M. Antibiotic resistance patterns of Gram-negative and Gram-positive strains isolated from inpatients with nosocomial infections in a tertiary hospital in Beijing, China from 2011 to 2014. Journal of Chemotherapy, 29, 2016: 317–20. DOI: 10.1080/1120009X.2016.1157946.
32. Abo-State MA, Mahdy HM, Ezzat SM, Abd El Shakour EH, El-Bahnasawy MA. Antimicrobial resistance profiles of *Enterobacteriaceae* isolated from Rosetta branch of River Nile, Egypt. World Applied Sciences Journal, 19(9), 2012: 1234–43.
33. Sperandeo P, Martorana AM, Polissi A. Lipopolysaccharide biogenesis and transport at the outer membrane of Gram-negative bacteria. Biochimica et Biophysica Acta: Molecular and Cell Biology of Lipids. 2017;1862(11):1451–1460. https://doi.org/10.1016/j.bbalip.2016.10.006
34. Kamanula M, Munthali CR, Dziwapo A, Kamanula JF. Mineral and phytochemical composition of baobab root tubers. Malawi Medical Journal, 30(4), 2018: 250–5.
35. Philip TT, Boamah VE, Badu M. Antioxidant and antimicrobial properties of *Adansonia digitata*. Heliyon, 10(8), 2024: e29660.
36. Wasihun AA, Sbhatu DB, Berhe GG, Abay KH, Gebreyohannes G. Phytochemical constituents of *Adansonia digitata* L. (Baobab) fruit pulp from Tekeze Valley, Tigrai, Ethiopia. International Journal of Analytical Chemistry, 2023, 2023: 5591059. DOI: 10.1155/2023/5591059
37. Muhammad J, Muaz M, Bako GD, Musa B, Zubair MI. Antibacterial activity of *Adansonia digitata* leaf extract. UMYU Journal of Microbiology Research, 10(3), 2025: 341–9.
38. Magashi AM, Abdulmalik U. Antibacterial activity and phytochemical screening of stem bark extracts of *Adansonia digitata* on some clinical isolates. UMYU Journal of Microbiology Research, 3(1), 2018: 1–7. DOI: 10.47430/ujmr.1831.001.
39. El Yahyaoui O, Bouabid B, Ait Ouaaziz N, El Bakkali M, El Harche H, Lrhorfi LA, Nakari K, Bengueddour R. The antibacterial activity and biochemical composition of *Adansonia digitata* edible parts. Arab Gulf Journal of Scientific Research, 41(1), 2023: 91–106. DOI: 10.1108/AGJSR-07-2022-0101.
40. Ajiboye AE, Sadiq SO, Adedayo MR. Antimicrobial activity and phytochemical screening of *Adansonia digitata* stem bark extract on some clinical isolates. Ife Journal of Science, 22(2), 2020.
41. Azwanida N. A review on extraction methods used in medicinal plants: Principle, strength and limitation. Medicinal and Aromatic Plants, 4, 2015: 1000196.
42. Sperandeo P, Martorana AM, Polissi A. Lipopolysaccharide biogenesis in Gram-negative bacteria. Biochimica et Biophysica Acta, 1862(11), 2017: 1451–60.
43. Silhavy TJ, Kahne D, Walker S. The bacterial cell envelope. Cold Spring Harbor Perspectives in Biology, 2(5), 2010: a000414.
44. Sun J, Rutherford ST, Silhavy TJ, Huang KC. Physical properties of the bacterial outer membrane. Nature Reviews Microbiology, 20, 2022: 236–48.
45. Lotfian Z, Nave H, Khalili R. Contribution of efflux activity in resistance to antibiotics in *Escherichia coli* clinical isolates. Journal of Infection and Chemotherapy, 31(3), 2025: 102725. DOI: 10.1016/j.jiac.2025.102725.
46. Liang J, Lin S, He L. Formation ability and drug resistance mechanism of *Klebsiella pneumoniae* biofilm and capsule for multidrug-resistant strains. Cellular and Molecular Biology (Noisy-le-grand), 69(10), 2023: 88–93. DOI: 10.14715/cmb/2023.69.10.12.

47. Pankey GA, Sabath LD. Clinical relevance of bacteriostatic vs bactericidal mechanisms. Clinical Infectious Diseases, 38(6), 2004: 864–70.