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Exploring Plant-Derived Antimicrobials: Comparative Activity of *Andrographis paniculata* and *Jatropha curcas* Extracts against Resistant *Klebsiella* Species

Umezulora, B. I.[1], Okoye, E. L.[1] and Uba, B. O.[2*]

[1]Department of Applied Microbiology and Brewing, Nnamdi Azikiwe University, P.M.B. 5025, Awka, Anambra State, Nigeria.
[2]Department of Microbiology, Chukwuemeka Odumegwu Ojukwu University, P.M.B. 02 Uli, Anambra State, Nigeria.

*Corresponding author e-mail: bo.uba@coou.edu.ng

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ABSTRACT

The emergence of multidrug-resistant *Klebsiella pneumoniae* and *Klebsiella quasipneumoniae* necessitates the exploration of alternative antimicrobial agents. This study evaluated the antibacterial efficacy of aqueous, hexane, and methanol leaf extracts of *Andrographis paniculata* and *Jatropha curcas* against clinical *Klebsiella* isolates. Disk diffusion assays revealed concentration-dependent antibacterial activity across extracts, with selective resistance observed in certain strains. The aqueous extract of *A. paniculata* exhibited activity against most isolates, with a maximum inhibition zone of 19.00 ± 0.25 mm, though strains 41F and 72F were resistant. The hexane extract showed broader activity, including superior efficacy compared to Ciprofloxacin against strain 41F, with a peak inhibition of 21.00 ± 0.20 mm. Methanol extract demonstrated the highest antibacterial activity overall, yielding a maximum inhibition zone of 31.00 ± 0.23 mm. Similarly, *J. curcas* extracts displayed notable antibacterial effects. The aqueous, hexane, and methanol extracts produced maximum inhibition zones of 22.00 ± 0.19 mm, 22.00 ± 0.30 mm, and 21.00 ± 0.15 mm, respectively, with enhanced activity against strain 41F compared to ciprofloxacin. Against *K. quasipneumoniae*, activity was only observed at higher concentrations (200 – 400 mg/mL), with hexane extract of *A. paniculata* showing the highest inhibition (17.00 ± 0.10 mm). Minimum inhibitory concentrations (MICs) ranged from 50 – 200 mg/mL and minimum bactericidal concentrations (MBCs) from 100–400 mg/mL for *K. pneumoniae*, while *K. quasipneumoniae* showed MICs of 100 – 200 mg/mL and MBCs of 200 –400 mg/mL. The findings highlight the potent antibacterial potential of *A. paniculata* and *J. curcas*, particularly methanol and hexane extracts, as promising candidates for the development of alternative therapies against resistant *Klebsiella* infections.

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Keywords
Andrographis paniculata; Antibacterial activity; Antimicrobial resistance; *Jatropha curcas*; *Klebsiella pneumoniae; Klebsiella quasipneumoniae*

Introduction

The global escalation of antimicrobial resistance (AMR) has emerged as one of the most pressing threats to public health, compromising the effective treatment of infectious diseases and increasing morbidity and mortality worldwide (Egurefa *et al*. 2020a; 2020b; Obiefuna *et al.* 2025; Obiefuna *et al*. 2026; Okoye *et al*., 2026). Among the most problematic pathogens is *Klebsiella pneumoniae*, a member of the *Enterobacteriaceae* family, which is widely implicated in hospital- and community-acquired infections such as pneumonia, urinary tract infections, septicemia, and wound infections (Uba *et al*., 2019a; 2019b; Alisa *et al*., 2020; Okolo *et al*., 2025). The organism is particularly concerning due to its remarkable ability to acquire resistance determinants, including extended-spectrum β-lactamases (ESBLs) and carbapenemases, rendering many frontline antibiotics ineffective. Furthermore, closely related species such as *Klebsiella quasipneumoniae* are increasingly recognized in clinical settings and are often associated with misidentification and multidrug resistance, thereby complicating treatment strategies (Anichebe *et al*., 2019; Uba *et al*., 2020a; 2020b; Anukam *et al*., 2020a; 2020b).

The diminishing efficacy of conventional antibiotics has intensified the need for alternative antimicrobial agents, particularly those derived from natural sources (Okoye *et al*. 2020a; 2020b; 2020c). Medicinal plants have long served as a cornerstone in drug discovery, providing structurally diverse bioactive compounds with therapeutic potential (Uba, 2019a; 2019b; Umeh *et al.*, 2020; 2021; Okpalaunegbu *et al.*, 2025). In recent years, there has been renewed scientific interest in phytochemicals due to their broad-spectrum antimicrobial activity, reduced side effects, and lower propensity for resistance development compared to synthetic drugs (Uba, 2019c; Uba *et al*. 2019c; 2019d). Notably, plant-derived compounds such as alkaloids, flavonoids, terpenoids, tannins, and phenolics have been widely reported to exhibit significant antibacterial activity against both Gram-positive and Gram-negative bacteria (Uba and Chukwura, 2016; Uba *et al*. 2020d; 2020e; Uba *et al*. 2024; Mere *et al.* 2025).

One such plant of considerable pharmacological importance is *Andrographis paniculata*, commonly referred to as the "king of bitters." This plant has been extensively utilized in traditional medicine systems across Asia and Africa for the treatment of infections, inflammation, and fever. Its biological activity is largely attributed to diterpenoid lactones, particularly andrographolide, which has been shown to possess potent antimicrobial, antiviral, anti-inflammatory, and immunomodulatory properties (Enemchukwu *et al*. 2026a; 2026b; Ofunwa *et al*., 2026a; 2026b). Recent studies have demonstrated that extracts of *A. paniculata* can inhibit the growth and biofilm formation of multidrug-resistant *K. pneumoniae*, highlighting its potential as a novel antimicrobial agent (Okafor *et al*. 2021a; 2021b; Dokubo *et al*., 2024; Obiefoka *et al*., 2023; Ubajekwe *et al*., 2025; Uba *et al*., 2025). Additionally, systematic reviews have confirmed the broad-

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spectrum antimicrobial activity of andrographolide against diverse microbial pathogens, reinforcing its relevance in combating AMR (Uba *et al*. 2016; Uba *et al.*, 2017; Njoku *et al*. 2019a; 2019b; Nkamigbo *et al*. 2020a; 2020b).

Similarly, *Jatropha curcas* has gained attention for its diverse pharmacological properties, including antimicrobial, antifungal, antioxidant, and anti-inflammatory activities. The plant is rich in bioactive constituents such as flavonoids, saponins, tannins, and diterpenes, which contribute to its antimicrobial efficacy (Uba e*t al.,* 2021a; 2021b; Dokubo *et al*., 2022a; 2022b; Anidu *et al*., 2023). Extracts of *J. curcas* have been reported to exhibit inhibitory effects against several pathogenic bacteria, including Gram-negative organisms, suggesting its potential role as an alternative therapeutic agent in the management of resistant infections (Uba *et al*., 2026a; 2026b; 2026c; Okwonkwo *et al.* 2026).

An important factor influencing the antimicrobial activity of plant extracts is the solvent used during extraction (Uba *et al*. 2018a; Uba *et al*. 2018b Uba *et al*., 2018c). Solvent polarity determines the type and quantity of phytochemicals extracted, thereby affecting the biological activity of the resulting extract (Ubani *et al*., 2024a; 2024b; Ubani *et al.,* 2025; Ekwenze *et al*., 2025). Polar solvents such as methanol and water are known to extract hydrophilic compounds like phenolics and flavonoids, while non-polar solvents such as hexane preferentially extract lipophilic compounds such as terpenoids and essential oils (Ibo *et al*. 2020; Okafor *et al.*, 2023; Ele *et al*., 2025; Uba and Okonkwo *et al*. 2025; Dokubo and Uba, 2026). Consequently, comparative evaluation of aqueous, methanol, and hexane extracts is essential to identify the most effective extraction method and bioactive fraction (Ofunwa *et al*., 2024: Alfred *et al*., 2023; Alfred *et al*., 2025; Okeke *et al*. 2025a; 2025b; Uba and Udaba *et al*. 2026).

Despite increasing evidence on the antimicrobial potential of medicinal plants, there remains a need for comprehensive studies evaluating their efficacy against clinically relevant and resistant bacterial strains, particularly within the *Klebsiella* complex (Uba *et al*., 2020c; Dokubo and Uba, 2023; Uba and Obiefuna, 2023). Moreover, the determination of key antimicrobial parameters such as minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) is critical for assessing the therapeutic potential and practical applicability of plant-derived extracts (Idu *et al*., 2026a; 2026b; Ibe *et al*., 2023; Chukwura *et al*., 2025).

Therefore, the present study investigates the antibacterial activity of aqueous, hexane, and methanol leaf extracts of *A. paniculata* and *J. curcas* against clinical isolates of *K. pneumoniae* and *K. quasipneumoniae*. By integrating inhibition zone assays with MIC and MBC determinations, this research aims to provide a comprehensive evaluation of the antimicrobial potential of these plants and contribute to the ongoing search for effective alternatives to conventional antibiotics in the era of rising antimicrobial resistance.

Materials and Methods

Analysis of the Study Plants

Collection and identification of plant materials

Fresh leaves from *Jatropha curcas* (JC) and *Andrographis paniculata* (AP) were collected from a local garden in Anambra State. They were taxonomically identified and authenticated by Iroka Finian, an expert Botanist from the Department of Botany, Nnamdi Azikiwe University, Awka. The herbarium numbers given to the plants were: NAUH – 028B for *Jatropha curcas* and NAUH – 198A for *Andrographis paniculata*.

Preparation of plant extracts

Fresh leaves from *Jatropha curcas* and *Andrographis paniculata* were washed with clean water and then air-dried for 14 days at room temperature. The leaves were ground into coarse powder using an industrial blender sterilized with 70% alcohol. Twenty (20 g) of each of the powdered leaves of the test plants were weighed and placed into three different 500 ml conical flasks containing 100 ml distilled water, 300 ml n-hexane and 300 ml methanol, mixed and macerated for 24 h for distilled water and for 72 h for hexane and methanol. The aqueous, n-hexane, and methanol extracts were double filtered using a muslin cloth and then through a filter paper (Whatman no. 1). The filtrates were concentrated to dryness in a water bath at 45 °C. The extracts were stored in a desiccator until needed (Oso *et al*., 2018).

Evaluation of the antibacterial properties of the plant extracts against test bacterial species and determination of the Minimal inhibitory concentration (MIC) and Minimal bactericidal concentration (MBC) of the plant extracts

Evaluation of the antibacterial properties of the plant extracts against test bacterial species using disk diffusion technique

Preparation of plant disk

The extracts were tested using 6 mm sterilized filter paper disks. The plant disks were prepared by immersing sterile blank discs into the extract concentrations. After the immersion, the plant disks were placed in a petri dish but were not allowed to touch each other. The plates containing the disks were exposed under the lamina flow hood to evaporate. Filter paper discs dipped in pure DMSO were also prepared for use as a negative control.

Preparation of media

Mueller Hinton agar media was prepared according to the manufacturer's instructions and sterilized in an autoclave at 121ºC for 15 minutes. Thereafter, fifteen milliliters of sterilized Mueller-Hinton agar medium were poured into sterile Petri dishes and stored in the refrigerator at 4º C.

Preparation of Extract Concentrations

Stock solutions (400 mg) of the aqueous, methanolic, and n-hexane extracts of *Jatropha curcas* and *Andrographis paniculata* were prepared by weighing 12g of the extracts and dissolving them in 30 ml of pure dimethylsulfoxide (DMSO) and diluting it two times (50%). This stock extract was sterilized and filtered using filter paper (0.2 µm) and stored in Eppendorf tubes at 4°C. The sterile stock extract was diluted in two-folds to obtain 400, 200, 100, 50 and 25 mg/ml concentrations, respectively as described by Oso *et al*. (2018).

Collection of Clinical Isolates

Pure isolates of *K. pneumoniae* and *K. quasipneumoniae* were collected from apparently healthy individuals, which were kindly donated by Nwankwo, Uchechukwu (unpublished data) and authenticated in Perfect Glory Scientific and Environmental Research Laboratory Services, Nnewi and the identity was reconfirm using appropriate colonial characteristics, staining techniques and biochemical tests according to the method of Cheesbrough (2006). For the experiments, the bacterial isolates were first subcultured in nutrient broth and incubated according to standard procedure. Pure colonies of the bacterial isolates were characterized and identified using Bergey's manual of Determinative Bacteriology authored by (Holt et al., 1994) after carrying out the biochemical test (Gram staining, spore staining,

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motility test, catalase test, citrate test, indole test, Methyl red test, Voges Proskauer test and sugar fermentation test (Uba *et al*., 2020f; 2020g; Oghonim *et al*., 2026a; 2026b).

Preparation of 0.5 McFarland standards

Solution A was prepared by adding 1.175 g of barium chloride ($BaCl_2$, $2H_2O$) to 100 mL distilled water. Solution B was prepared by adding 1ml of sulphuric acid (H_2S0_4) (0.36N) to 100 ml of distilled water. Then 0.5 mL of solution A was added to 99.5 mL of solution B, mixed well and distributed in test tubes with a screw cap. The cap is closed tightly to avoid evaporation. The mixture was stored in the dark. The solution was agitated vigorously before using it. After standardization of bacterial suspension, a sterile cotton swab was immersed in it and was rotated several times with firm pressure on the inside wall of the tube to remove excess fluid (Omwirhiren *et al*., 2017).

Test for antibacterial activities

The disk diffusion method was used to evaluate the antimicrobial activity of each plant extract. Using the Mueller Hinton media, Swab sticks inoculated with each bacterial suspension (viable cell count of 1.5 x 10^8 CFU) was used to inoculate the media and was appropriately labelled. After 30 minutes of inoculation, sterile filter paper discs loaded with plant extract concentrations of 400, 200, 100, 50, and 25 mg/ml concentrations were placed aseptically on the inoculated Mueller-Hinton media. Filter paper disks loaded with 1 mg of Ciprofloxacin was used as a positive control, while filter papers impregnated with pure DMSO was the negative control. The plates were kept in the fridge at 5 °C for 2 h to permit plant extract diffusion and then incubated at 37 °C for 24 h, respectively. The presence of inhibition zones around each disk was measured by millimeter calibrated meter rule, recorded, and considered an indication of antibacterial activity (Mostafa *et al*., 2018).

Determination of the Minimal Inhibitory Concentration (MIC) and Minimal Bactericidal Concentration (MBC) of the plant extracts

Determination of the Minimal Inhibitory Concentration (MIC)

MIC is defined as the lowest concentration of the antimicrobial agent that inhibits microbial growth after 24 h and 48 h of incubation. The MIC was performed according to the Clinical and Laboratory Standards Institute guidelines (CLSI, 2025) using extracts against bacterial pathogens in a 96-well U-bottomed microtitre plate. The plant extracts were serially diluted from a concentration of 400mg/ml to 0.78mg/ml, and were added to the final inoculum of 5 x 10^5 CFU/ml. Using the tube dilution technique, 12 g of *Jatropha curcas* and *Andrographis paniculata* extracts were dissolved in 30 ml sterile nutrient broths, giving 400 mg/ml. Thereafter, two-fold serial dilutions were made from the original stocks of 30 ml using Mueller-Hinton broth to achieve the following concentrations: 400, 200, 100, 50, 25, 12.5, 6.25, 3.125, 1.56 and 0.781 mg/ml (Adegoke *et al*., 2010). Having obtained different dilutions and concentrations, 0.1 ml of standardized test organisms was inoculated into each diluted tube and incubated at 37 °C for 24 h for the test bacterial isolates. The lowest concentration of the extracts that inhibited the growth of the test organism was recorded as the MIC.

Determination of the Minimal Bactericidal concentration (MBC)

Bacterial tubes showing no visible growth from the MIC test were sub-cultured into nutrient agar and incubated at 37 °C for 24 h. The lowest concentration of the extracts yielding no growth on sub-culturing was recorded as the minimum bactericidal concentration (MBC) for bacteria described by Adegoke *et al*. (2010).

Data Analysis

The results were expressed as mean ±standard deviation (Mean ± S.D.) of three different measurements. Statistical analysis was performed on data generated from the study using GraphPad Prism version 8.0.2 software. Statistical significance was determined using two-way analysis of variance (ANOVA). Values with $p \leq 0.05$ compared with the control groups were considered as being statistically significant (Uba *et al.*, 2020h; Afulukwe *et al.*, 2025; 2026).

Results

Tables 1A and 1B summarized the antibacterial activity of aqueous extract of *Andrographis paniculata* leaves at varying concentrations against *Klebsiella pneumoniae* strains. The aqueous extract of *Andrographis paniculata* leaves demonstrated antibacterial activity at different concentrations against all the *K. pneumoniae* strains tested except *K. pneumoniae* strains 41F and 72F, where inhibition zone diameter was 0.00, signifying no activity. Notably, *K. pneumoniae* strain 41F was also resistant to the standard control antibiotic. The aqueous extract of *Andrographis paniculata* exhibited antibacterial activity at all concentrations tested against *K. pneumoniae* strains 60FA, 193F and 344F only. On the overall, the highest inhibition zone diameter (mm) obtained from aqueous extract of *Andrographis paniculata* leaves was 19.00±0.25 against *K. pneumoniae* strains.

Tables 2A and 2B summarized the antibacterial activity of the hexane extract of *Andrographis paniculata* leaves at varying concentrations against *Klebsiella pneumoniae* strains. The hexane extract of *Andrographis paniculata* leaves demonstrated antibacterial activity at different concentrations against all the *K. pneumoniae* strains tested except *K. pneumoniae* strains 258F and 376F, where inhibition zone diameter was 0.00, signifying no antibacterial activity. Notably, the hexane extract of *Andrographis paniculata* leaves was more active than the standard control antibiotic, Ciprofloxacin against *K. pneumoniae* strain 41F. The hexane extract of *Andrographis paniculata* exhibited antibacterial activity at all concentrations tested against *K. pneumoniae* strains particularly; 60FA, 193F and 344F. On the overall, the highest inhibition zone diameter (mm) obtained from hexane extract of *Andrographis paniculata* leaves was 21.00±0.20 against *K. pneumoniae* strains.

Tables 3A and 3B summarized the antibacterial activity of methanol extract of *Andrographis paniculata* leaves at varying concentrations against *Klebsiella pneumoniae* strains. The methanol extract of *Andrographis paniculata* leaves demonstrated antibacterial activity at different concentrations against all the *K. pneumoniae* strains tested except *K. pneumoniae* strain 72F, where inhibition zone diameter of extract was 0.00, signifying no activity. Notably, the methanol extract of *Andrographis paniculata* leaves was more active than the standard control antibiotic, Ciprofloxacin against *K. pneumoniae* strain 41F. The methanol extract of *Andrographis paniculata* leaves exhibited maximum antibacterial activity at all concentrations tested against *K. pneumoniae* strains 60FA, 302M, 344F and 345F. On the overall, the highest inhibition zone diameter (mm) obtained from methanol extract of *Andrographis paniculata* leaves was 31.00±0.23 against *K. pneumoniae* strains

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Table 1A: Antibacterial Activity of Aqueous Extract of *Andrographis paniculata* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	41F	-	-	-	-	-	-	-	=0.0002	<0.0001
K. pneumoniae	57M	20.00±0.35	-	13.00±0.12	-	-	-	-	=0.0006	<0.0001
K. pneumoniae	60FA	20.00±0.45	-	19.00±0.25	18.00±0.20	18.00±0.15	10.00±0.15	8.00±0.10	=0.0004	<0.0001
K. pneumoniae	72F	23.00±0.40	-	-	-	-	-	-	=0.0377	<0.0001
K. pneumoniae	87F	25.30±0.30	-	13.00±0.10	11.00±0.20	-	-	-	=0.0620	<0.0001
K. pneumoniae	95F	29.00±0.50	-	15.00±0.09	15.00±0.10	-	-	-	<0.0001	<0.0001
K. pneumoniae	100F	20.00±0.22	-	12.00±0.12	-	-	-	-	=0.0909	<0.0001
K. pneumoniae	183M	33.00±0.33	-	10.00±0.10	8.00±0.10	-	-	-	=0.0007	<0.0001
K. pneumoniae	193F	22.00±0.22	-	15.00±0.22	13.00±0.23	12.00±0.10	10.00±0.10	9.00±0.11	=0.0005	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 1B: Antibacterial Activity of Aqueous Extract of *Andrographis paniculata* leaves against *Klebsiella pneumoniae* Strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	239M	36.00±0.44	-	12.00±0.14	-	-	-	-	= 0.0011	<0.0001
K. pneumoniae	258F	22.00±0.24	-	18.00±0.14	15.00±0.14	-	-	-	<0.0001	<0.0001
K. pneumoniae	302M	35.00±0.39	-	15.00±0.13	14.00±0.11	-	-	-	= 0.0006	<0.0001
K. pneumoniae	344F	40.00±0.35	-	19.00±0.14	19.00±0.14	18.00±0.12	10.00±0.11	10.00±0.10	= 0.0005	<0.0001
K. pneumoniae	345F	42.00±0.48	-	19.00±0.13	-	-	-	-	= 0.0001	<0.0001
K. pneumoniae	376F	56.00±0.65	-	14.00±0.15	11.00±0.15	-	-	-	= 0.0513	<0.0001
K. pneumoniae	418M	51.00±0.49	-	11.00±0.15	-	-	-	-	=0.3561	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 2A: Antibacterial Activity of Hexane Extract of *Andrographis paniculata* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	41F	-	-	16.00±0.15	12.00±0.20	-	-	-	= 0.0002	<0.0001
K. pneumoniae	57M	20.00±0.35	-	13.00±0.15	12.00±0.20	-	-	-	= 0.0006	<0.0001
K. pneumoniae	60FA	20.00±0.45	-	20.00±0.35	15.00±0.15	15.00±0.15	11.00±0.20	10.00±0.10	= 0.0004	<0.0001
K. pneumoniae	72F	23.00±0.40	-	14.00±0.40	11.00±0.40	-	-	-	= 0.0377	<0.0001
K. pneumoniae	87F	25.30±0.30	-	11.00±0.10	10.00±0.14	-	-	-	=0.0620	<0.0001
K. pneumoniae	95F	29.00±0.50	-	14.00±0.20	13.00±0.10	-	-	-	<0.0001	<0.0001
K. pneumoniae	100F	20.00±0.22	-	13.00±0.15	11.00±0.10	-	-	-	= 0.0909	<0.0001
K. pneumoniae	183M	33.00±0.33	-	14.00±0.15	-	-	-	-	= 0.0007	<0.0001
K. pneumoniae	193F	22.00±0.22	-	17.00±0.13	14.00±0.12	12.00±0.14	-	-	= 0.0005	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

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Table 2B: Antibacterial Activity of Hexane Extract of *Andrographis paniculata* Leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	239M	36.00±0.44	-	18.00±0.14	11.00±0.11	-	-	-	= 0.0011	<0.0001
K. pneumoniae	258F	22.00±0.24	-	-	-	-	-	-	<0.0001	<0.0001
K. pneumoniae	302M	35.00±0.39	-	21.00±0.12	19.00±0.11	15.00±0.12	11.00±0.11	11.00±0.10	= 0.0006	<0.0001
K. pneumoniae	344F	40.00±0.35	-	15.00±0.16	15.00±0.13	14.00±0.11	10.00±0.11	9.00±0.10	= 0.0005	<0.0001
K. pneumoniae	345F	42.00±0.48	-	21.00±0.20	21.00±0.21	20.00±0.16	19.00±0.16	18.00±0.13	= 0.0001	<0.0001
K. pneumoniae	376F	56.00±0.65	-	-	-	-	-	-	= 0.0513	<0.0001
K. pneumoniae	418M	51.00±0.49	-	12.00±0.14	-	-	-	-	=0.3561	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 3A: Antibacterial Activity of Methanol Extract of *Andrographis paniculata* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	41F	-	-	13.00±0.23	13.00±0.10	9.00±0.13	9.00±0.20	-	= 0.0002	<0.0001
K. pneumoniae	57M	20.00±0.35	-	12.00±0.20	-	-	-	-	= 0.0006	<0.0001
K. pneumoniae	60FA	20.00±0.45	-	16.00±0.12	16.00±0.10	15.00±0.10	15.00±0.20	10.00±0.15	= 0.0004	<0.0001
K. pneumoniae	72F	23.00±0.40	-	-	-	-	-	-	= 0.0377	<0.0001
K. pneumoniae	87F	25.30±0.30	-	11.00±0.20	10.00±0.15	-	-	-	=0.0620	<0.0001
K. pneumoniae	95F	29.00±0.50	-	17.00±0.20	13.00±0.10	-	-	-	<0.0001	<0.0001
K. pneumoniae	100F	20.00±0.22	-	20.00±0.13	18.00±0.14	-	-	-	= 0.0909	<0.0001
K. pneumoniae	183M	33.00±0.33	-	20.00±0.13	18.00±0.14	-	-	-	= 0.0909	<0.0001
K. pneumoniae	193F	22.00±0.22	-	15.00±0.15	12.00±0.13	11.00±0.10	11.00±0.11	-	= 0.0005	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 3B: Antibacterial Activity of Methanol Extract of *Andrographis paniculata* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	239M	36.00±0.44	-	13.00±0.15	13.00±0.13	12.00±0.10	12.00±0.11	-	= 0.0011	<0.0001
K. pneumoniae	258F	22.00±0.24	-	11.00±0.11	-	-	-	-	<0.0001	<0.0001
K. pneumoniae	302M	35.00±0.39	-	13.00±0.12	13.00±0.11	12.00±0.11	10.00±0.10	11.00±0.10	= 0.0006	<0.0001
K. pneumoniae	344F	40.00±0.35	-	31.00±0.23	31.00±0.20	30.00±0.21	15.00±0.12	13.00±0.12	= 0.0005	<0.0001
K. pneumoniae	345F	42.00±0.48	-	18.00±0.21	17.00±0.20	16.00±0.17	16.00±0.15	14.00±0.13	= 0.0001	<0.0001
K. pneumoniae	376F	56.00±0.65	-	16.00±0.15	11.00±0.13	-	-	-	= 0.0513	<0.0001
K. pneumoniae	418M	51.00±0.49	-	10.00±0.16	-	-	-	-	=0.3561	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

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Tables 4A and 4B showed the antibacterial activity of the aqueous extract of *Jatropha curcas* leaves at varying concentrations against *Klebsiella pneumoniae* strains. The aqueous extract of *Jatropha curcas* leaves demonstrated antibacterial activity at different concentrations against all the *K. pneumoniae* strains tested except *K. pneumoniae* strains 87F, 100F and 183M, where inhibition zone diameter was 0.00, signifying no activity. Notably, the aqueous extract of *Jatropha curcas* leaves was more active than the standard control antibiotic, Ciprofloxacin against *K. pneumoniae* strain 41F. The aqueous extract of *Jatropha curcas* leaves exhibited antibacterial activity at all concentrations tested against *K. pneumoniae* strains particularly; 41F, 72F, 95F, 258F, 302M, 344F and 345F. On the overall, the highest inhibition zone diameter (mm) obtained from aqueous extract of *Jatropha curcas* leaves was 22.00±0.19 against *K. pneumoniae* strains.

Tables 5A and 5B showed the antibacterial activity of the hexane extract of *Jatropha curcas* leaves at varying concentrations against *Klebsiella pneumoniae* strains. The hexane extract of *Jatropha curcas* leaves demonstrated antibacterial activity at different concentrations against all the *K. pneumoniae* strains tested except *K. pneumoniae* strains 72F, 87F, 100F and 193F, where inhibition zone diameter was 0.00, signifying no activity. Notably, the hexane extract of *Jatropha curcas* leaves was more active than the standard control antibiotic, Ciprofloxacin against *K. pneumoniae* strain 41F. The hexane extract of *Jatropha curcas* leaves demonstrated high antibacterial activity at all concentrations tested against *K. pneumoniae* strains particularly; 41F, 95F, 183M, 302M, 344F and 345F. On the overall, the highest inhibition zone diameter (mm) obtained from hexane extract of *Jatropha curcas* leaves was 22.00±0.30 against *K. pneumoniae* strains.

Tables 6A and 6B showed the antibacterial activity of the methanol extract of *Jatropha curcas* leaves at varying concentrations against *Klebsiella pneumoniae* strains. The methanol extract of *Jatropha curcas* leaves demonstrated antibacterial activity at different concentrations against all the *K. pneumoniae* strains tested except *K. pneumoniae* strains 72F, 95F, 193F,258F and 376F, where inhibition zone diameter was 0.00, signifying no activity. Notably, the methanol extract of *Jatropha curcas* leaves was more active than the standard control antibiotic, Ciprofloxacin against *K. pneumoniae* strain 41F. The methanol extract of *Jatropha curcas* leaves exhibited antibacterial activity at all concentrations tested against *K. pneumoniae* strains particularly; 57M, 183M, 239M, 302M, 344F and 345F. On the overall, the highest inhibition zone diameter (mm) obtained from methanol extract of *Jatropha curcas* leaves was 21.00±0.15 against *K. pneumoniae* strains.

Table 7 presented the antibacterial activity of the aqueous, hexane and methanol extracts of *Andrographis paniculata* and *Jatropha curcas* leaves against *Klebsiella quasipneumoniae* at varying concentrations. Generally, the active plant extracts exhibited antibacterial activity against *Klebsiella quasipneumoniae* at 400 mg/ml and 200 mg/ml only. None of the extracts demonstrated antibacterial activity at lower therapeutic concentrations. Aqueous extract of *Andrographis paniculata* leaves demonstrated antibacterial activity against *Klebsiella quasipneumoniae* and gave the highest inhibition zone diameter (mm) of 14.00±0.20. The hexane extract of *Andrographis paniculata* leaves exhibited antibacterial activity against *Klebsiella quasipneumoniae* and produced the highest inhibition zone diameter (mm) of 17.00±0.10. The methanol extract of *Andrographis paniculata* leaves demonstrated antibacterial activity against *Klebsiella quasipneumoniae* and had the highest inhibition zone diameter (mm) of 12.00±0.10. The aqueous extract of *Jatropha curcas* leaves demonstrated antibacterial activity against *Klebsiella quasipneumoniae*, and presented the highest inhibition zone diameter (mm) of 12.00±0.10. The hexane extract of *Jatropha curcas* leaves exhibited antibacterial activity against *Klebsiella quasipneumoniae*, and produced the highest inhibition zone diameter (mm) of 14.00±0.20. The methanol extract of *Jatropha curcas* leaves showed antibacterial activity against *Klebsiella quasipneumoniae* and gave the highest inhibition zone diameter (mm) of 13.00±0.10.

Tables 8A - B showed the MIC (mg/ml) and MBC (mg/ml) of the aqueous, hexane and methanol extracts of *Andrographis paniculata* and *Jatropha curcas* leaves tested against *K. pneumoniae* strains. The plant extracts demonstrated varying MICs that ranged from 50 mg/ml to 200 mg/ml and MBCs that ranged from 100 mg/ml to 400 mg/ml. Hence, the highest MIC and MBC exhibited against *K. pneumoniae* strains were 50 mg/ml and 100 mg/ml, respectively.

Table 9 showed the MIC (mg/mL) and MBC (mg/mL) of the aqueous, hexane and methanol extracts of *Andrographis paniculata* and *Jatropha curcas* leaves tested against *K. quasipneumoniae*. The plant extracts demonstrated varying MICs that ranged from 100 mg/ml to 200 mg/ml and MBCs that ranged from 200 mg/ml to 400 mg/ml, hence, the highest MIC and MBC exhibited against *K. quasipneumoniae* were 100 mg/ml and 200 mg/ml, respectively.

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Table 4A: Antibacterial Activity of Aqueous Extract of *Jatropha curcas* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	41F	-	-	13.00±0.15	13.00±0.12	12.00±0.20	12.00±0.18	11.00±0.25	= 0.0002	<0.0001
K. pneumoniae	57M	20.00±0.35	-	12.00±0.25	-	-	-	-	= 0.0006	<0.0001
K. pneumoniae	60FA	20.00±0.45	-	14.00±0.15	13.00±0.18	-	-	-	= 0.0004	<0.0001
K. pneumoniae	72F	23.00±0.40	-	16.00±0.10	15.00±0.15	13.00±0.20	12.00±0.14	11.00±0.10	= 0.0377	<0.0001
K. pneumoniae	87F	25.30±0.30	-	-	-	-	-	-	=0.0620	<0.0001
K. pneumoniae	95F	29.00±0.50	-	17.00±0.30	15.00±0.10	13.00±0.10	12.00±0.20	12.00±0.15	<0.0001	<0.0001
K. pneumoniae	100F	20.00±0.22	-	-	-	-	-	-	= 0.0909	<0.0001
K. pneumoniae	183M	33.00±0.33	-	-	-	-	-	-	= 0.0007	<0.0001
K. pneumoniae	193F	22.00±0.22	-	14.00±0.13	-	-	-	-	= 0.0005	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 4B: Antibacterial Activity of Aqueous Extract of *Jatropha curcas* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	239M	36.00±0.44	-	17.00±0.16	16.00±0.15	-	-	-	= 0.0011	<0.0001
K. pneumoniae	258F	22.00±0.24	-	22.00±0.19	21.00±0.18	20.00±0.18	16.00±0.16	16.00±0.15	<0.0001	<0.0001
K. pneumoniae	302M	35.00±0.39	-	15.00±0.16	13.00±0.13	12.00±0.12	10.00±0.13	10.00±0.11	= 0.0006	<0.0001
K. pneumoniae	344F	40.00±0.35	-	18.00±0.15	18.00±0.15	17.00±0.14	14.00±0.12	9.00±0.11	= 0.0005	<0.0001
K. pneumoniae	345F	42.00±0.48	-	15.00±0.13	15.00±0.20	14.00±0.15	14.00±0.14	11.00±0.13	= 0.0001	<0.0001
K. pneumoniae	376F	56.00±0.65	-	13.00±0.12	12.00±0.13	12.00±0.15	-	-	= 0.0513	<0.0001
K. pneumoniae	418M	51.00±0.49	-	13.00±0.15	11.00±0.15	-	-	-	=0.3561	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 5A: Antibacterial Activity of Hexane Extract of *Jatropha curcas* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	41F	-	-	21.00±0.35	20.00±0.25	20.00±0.20	16.00±0.15	15.00±0.10	= 0.0002	<0.0001
K. pneumoniae	57M	20.00±0.35	-	13.00±0.10	11.00±0.35	-	-	-	= 0.0006	<0.0001
K. pneumoniae	60FA	20.00±0.45	-	15.00±0.20	15.00±0.10	-	-	-	= 0.0004	<0.0001
K. pneumoniae	72F	23.00±0.40	-	-	-	-	-	-	= 0.0377	<0.0001
K. pneumoniae	87F	25.30±0.30	-	-	-	-	-	-	=0.0620	<0.0001
K. pneumoniae	95F	29.00±0.50	-	22.00±0.30	22.00±0.20	21.00±0.10	15.00±0.10	13.00±0.10	<0.0001	<0.0001
K. pneumoniae	100F	20.00±0.22	-	-	-	-	-	-	= 0.0909	<0.0001
K. pneumoniae	183M	33.00±0.33	-	14.00±0.20	13.00±0.10	10.00±0.10	10.00±0.12	9.00±0.11	= 0.0007	<0.0001
K. pneumoniae	193F	22.00±0.22	-	-	-	-	-	-	= 0.0005	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

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Table 5B: Antibacterial Activity of Hexane Extract of *Jatropha curcas* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	239M	36.00±0.44	-	12.00±0.13	-	-	-	-	= 0.0011	<0.0001
K. pneumoniae	258F	22.00±0.24	-	15.00±0.13	13.00±0.11	-	-	-	<0.0001	<0.0001
K. pneumoniae	302M	35.00±0.39	-	20.00±0.19	19.00±0.16	15.00±0.13	13.00±0.11	10.00±0.10	= 0.0006	<0.0001
K. pneumoniae	344F	40.00±0.35	-	15.00±0.15	15.00±0.13	14.00±0.15	12.00±0.11	10.00±0.12	= 0.0005	<0.0001
K. pneumoniae	345F	42.00±0.48	-	15.00±0.14	14.00±0.13	13.00±0.11	11.00±0.12	9.00±0.10	= 0.0001	<0.0001
K. pneumoniae	376F	56.00±0.65	-	11.00±0.15	-	-	-	-	= 0.0513	<0.0001
K. pneumoniae	418M	51.00±0.49	-	13.00±0.15	-	-	-	-	=0.3561	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 6A: Antibacterial Activity of Methanol Extract of *Jatropha curcas* leaves against *Klebsiella pneumoniae* strains

Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	41F	-	-	18.00±0.15	12.00±0.10	-	-	-	=0.0002	<0.0001
K. pneumoniae	57M	20.00±0.35	-	16.00±0.25	15.00±0.10	15.00±0.15	10.00±0.10	9.00 ±0.12	= 0.0006	<0.0001
K. pneumoniae	60FA	20.00±0.45	-	14.00±0.10	13.00±0.15	-	-	-	= 0.0004	<0.0001
K. pneumoniae	72F	23.00±0.40	-	-	-	-	-	-	= 0.0377	<0.0001
K. pneumoniae	87F	25.30±0.30	-	11.00±0.30	11.00±0.20	-	-	-	=0.0620	<0.0001
K. pneumoniae	95F	29.00±0.50	-	-	-	-	-	-	<0.0001	<0.0001
K. pneumoniae	100F	20.00±0.22	-	12.00±0.17	12.00±0.20	-	-	-	= 0.0909	<0.0001
K. pneumoniae	183M	33.00±0.33	-	15.00±0.20	15.00±0.10	10.00±0.11	10.00±0.11	10.00±0.10	= 0.0007	<0.0001
K. pneumoniae	193F	22.00±0.22	-	-	-	-	-	-	= 0.0005	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

Table 6B: Antibacterial Activity of Methanol Extract of *Jatropha curcas* leaves against *Klebsiella pneumoniae* strains

Test Organism	Strain codes	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
K. pneumoniae	239M	36.00±0.44	-	21.00±0.15	18.00±0.15	13.00±0.12	12.00±0.10	12.00±0.11	= 0.0011	<0.0001
K. pneumoniae	258F	22.00±0.24	-	-	-	-	-	-	<0.0001	<0.0001
K. pneumoniae	302M	35.00±0.39	-	17.00±0.15	16.00±0.12	16.00±0.13	15.00±0.10	15.00±0.12	= 0.0006	<0.0001
K. pneumoniae	344F	40.00±0.35	-	18.00±0.18	17.00±0.16	16.00±0.13	16.00±0.14	15.00±0.11	= 0.0005	<0.0001
K. pneumoniae	345F	42.00±0.48	-	14.00±0.13	14.00±0.12	13.00±0.11	12.00±0.13	8.00±0.10	= 0.0001	<0.0001
K. pneumoniae	376F	56.00±0.65	-	-	-	-	-	-	= 0.0513	<0.0001
K. pneumoniae	418M	51.00±0.49	-	15.00±0.14	-	-	-	-	=0.3561	<0.0001

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), (-) = no activity, ± = Standard deviation values of the mean activity, p<0.05 – significant; p>0.05 – not significant.

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Table 7: The Antibacterial Activity of the Aqueous, Hexane, and Methanol Extracts of *Andrographis paniculata* and *Jatropha curcas* Leaves against *Klebsiella quasipneumoniae*

Test Organism/ strain code	ET	CIP (mm)	DMSO (mm)	400 mg/ml (mm)	200 mg/ml (mm)	100 mg/ml (mm)	50 mg/ml (mm)	25 mg/ml (mm)	P-value extract effect	P-value concentration effect
***K. quasipneumoniae* 194M**	APA	36.00±0.42	-	14.00±0.20	-	-	-	-	<0.0001	= 0.3765
	APH	36.00±0.42	-	17.00±0.10	14.00±0.20	-	-	-	<0.0001	= 0.3765
	APM	36.00±0.42	-	12.00±0.10	12.00±0.20	-	-	-	<0.0001	= 0.3765
	JCA	36.00±0.42	-	12.00±0.10	12.00±0.12	-	-	-	<0.0001	= 0.3765
	JCH	36.00±0.42	-	14.00±0.20	14.00±0.20	-	-	-	<0.0001	= 0.3765
	JCM	36.00±0.42	-	13.00±0.10	-	-	-	-	<0.0001	= 0.3765

CIP = Ciprofloxacin (positive control), DMSO = Dimethylsulphoxide (negative control), APA =*Andrographis paniculata* aqueous, APH = *Andrographis paniculata* hexane, APM = *Andrographis paniculata* methanol, JCA = *Jatropha curcas* aqueous, JCH = *Jatropha curcas* hexane, JCM = *Jatropha curcas* methanol, (-) = no activity, ± = Standard deviation values of the mean activity, $p<0.05$ – significant; $p>0.05$ – not significant; ET = Extract Type

Table 8A: The Minimal Inhibitory Concentrations (MICs) and Minimal Bactericidal Concentrations (MBCs) of the Aqueous, Hexane, and Methanol Extracts of *Andrographis paniculata* and *Jatropha curcas* Leaves against *Klebsiella pneumoniae*

Test Organisms	Strain codes	MICs of plant extracts (mg/ml)						MBCs of plant extracts (mg/ml)					
		APA	APH	APM	JCA	JCH	JCM	APA	APH	APM	JCA	JCH	JCM
K. pneumonia	41F	NT	100	200	200	50	100	NT	200	400	400	100	200
K. pneumonia	57M	200	200	200	200	200	100	400	400	400	400	400	200
K. pneumonia	60FA	100	50	100	200	200	200	200	100	200	400	400	400
K. pneumoniae	72F	NT	200	NT	100	NT	NT	NT	400	NT	200	NT	NT
K. pneumoniae	87F	200	200	200	NT	NT	200	400	400	400	NT	NT	400
K. pneumoniae	95F	100	200	100	100	50	NT	200	400	200	200	100	NT
K. pneumoniae	100F	200	200	50	NT	NT	200	400	400	100	NT	NT	400
K. pneumoniae	183M	200	200	50	NT	200	200	400	400	100	NT	400	400
K. pneumoniae	193F	100	100	200	200	NT	NT	200	200	400	400	NT	NT

APH - *Andrographis paniculata* hexane; APM - *Andrographis paniculata* methanol; APA -*Andrographis paniculata* aqueous; JCH - *Jatropha curcas* hexane; JCM= *Jatropha curcas* methanol; JCA= *Jatropha curcas* aqueous; NT = Not tested.

Table 8B: The Minimal Inhibitory Concentrations (MICs) and Minimal Bactericidal Concentrations (MICs) of the Aqueous, Hexane and Methanol Extracts of *Andrographis paniculata* and *Jatropha curcas* leaves against *Klebsiella pneumoniae*

Test Organisms	Strain codes	MICs of plant extracts (mg/ml)						MBCs of plant extracts (mg/ml)					
		APA	**APH**	**APM**	**JCA**	**JCH**	**JCM**	**APA**	**APH**	**APM**	**JCA**	**JCH**	**JCM**
K. pneumoniae	239M	400	100	200	100	200	50	400	200	400	200	400	100
K. pneumoniae	258F	100	NT	200	50	200	NT	200	NT	400	100	400	NT
K. pneumoniae	302M	100	50	200	200	50	100	200	100	400	400	100	200
K. pneumoniae	344F	100	200	50	100	200	100	200	400	100	200	400	200
K. pneumoniae	345F	100	50	100	200	200	200	200	100	200	400	400	400
K. pneumoniae	376F	200	NT	100	200	200	NT	400	NT	200	400	400	NT
K. pneumoniae	418M	200	200	200	200	200	100	400	400	400	400	400	200

APH - *Andrographis paniculata* hexane; APM - *Andrographis paniculata* methanol; APA -*Andrographis paniculata* aqueous; JCH - *Jatropha curcas* hexane; JCM= *Jatropha curcas* methanol; JCA= *Jatropha curcas* aqueous; NT = Not tested.

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Table 9: The Minimal Inhibitory Concentrations (MICs) and Minimal Bactericidal Concentrations (MBCs) of the Aqueous, Hexane and Methanol Extracts of *Andrographis paniculata* and *Jatropha curcas* Leaves against *Klebsiella quasipneumoniae*

Test Organisms/strain code	Extract types	MIC (mg/ml)	MBC (mg/ml)
***K. quasipneumoniae* 194M**	APA	200	400
	APH	100	200
	APM	200	400
	JCA	200	400
	JCH	100	200
	JCM	200	400

APH. = *Andrographis paniculata* hexane; APM=*Andrographis paniculata* methanol; APA =*Andrographis paniculata* aqueous; JCH= *Jatropha curcas* hexane; JCM= *Jatropha curcas* methanol; JCA= *Jatropha curcas* aqueous; NT = Not tested.

Discussion

The present study demonstrates that leaf extracts of *Andrographis paniculata* and *Jatropha curcas* possess measurable antibacterial activity against clinical isolates of *Klebsiella pneumoniae* and *Klebsiella quasipneumoniae*, with clear dependence on extraction solvent, concentration, and strain-specific susceptibility. Overall, methanol and hexane extracts exhibited stronger activity than aqueous extracts, and several extracts outperformed the standard antibiotic Ciprofloxacin against selected isolates (notably strain 41F), underscoring the therapeutic promise of these botanicals against resistant *Klebsiella* spp.

The superior antibacterial activity observed for methanol extracts—particularly *A. paniculata* with a peak inhibition zone of 31.00 ± 0.23 mm—is consistent with the ability of polar organic solvents to extract high yields of bioactive phytochemicals such as flavonoids, phenolics, and diterpenoids (e.g., andrographolide). These compounds disrupt bacterial membranes, inhibit nucleic acid synthesis, and interfere with quorum sensing and biofilm formation (Iheukwumere *et al*., 2012a; 2012b; Banerjee *et al*., 2021; Cushnie *et al*., 2020). The notable activity of hexane extracts (maximum 21.00 ± 0.20 mm for *A. paniculata* and 22.00 ± 0.30 mm for *J. curcas*) further suggests that non-polar constituents—such as terpenoids and essential oils—also contribute significantly to antibacterial action by targeting membrane integrity and permeability. Similar solvent-dependent differences have been widely reported, with methanol often yielding the highest antimicrobial potency due to its broad extraction spectrum (Azwanida, 2015; Do *et al.*, 2014; Mundi *et al*., 2013; 2014).

Across extracts, *A. paniculata* generally exhibited stronger antibacterial activity than *J. curcas*, particularly in methanol fractions. This aligns with reports attributing potent antimicrobial effects to andrographolide and related diterpenes, which have shown activity against multidrug-resistant Gram-negative bacteria, including *Klebsiella* spp. (Banerjee *et al.*, 2021; Gupta *et al.*, 2022). Nonetheless, *J. curcas* extracts demonstrated substantial inhibition (up to 22.00 ± 0.19 mm in aqueous and 21.00 ± 0.15 mm in methanol extracts), indicating that its phytochemical profile—rich in tannins, saponins, and flavonoids—also confers meaningful antibacterial activity. Prior studies have similarly documented the broad-spectrum antimicrobial properties of *J. curcas* leaf extracts, particularly against Gram-negative pathogens (Igbinosa *et al*., 2009; Devappa *et al*., 2020; Uba and Umennadi, 2026).

A striking finding in this study is the ability of certain plant extracts—especially hexane and methanol fractions of both plants—to exhibit greater activity than ciprofloxacin against *K. pneumoniae* strain 41F. This observation is particularly relevant given the global increase in fluoroquinolone resistance among *Klebsiella* spp., often mediated by chromosomal mutations and plasmid-borne resistance genes (WHO, 2023; Rodríguez-Medina *et al*., 2022). The enhanced activity of plant extracts may reflect their multi-target mechanisms of action, which reduce the likelihood of resistance development compared to single-target antibiotics. Additionally, phytochemicals may act synergistically, enhancing antibacterial efficacy through combined mechanisms such as membrane disruption and enzyme inhibition (Anameze *et al*. 2023; Ezeamama *et al*. 2025a; 2025b; Umezulora *et al.*, 2026).

Despite broad activity, several *K. pneumoniae* strains (e.g., 72F, 87F, 100F, 193F, 258F, 376F) showed resistance to specific extracts, as indicated by zero inhibition zones. This variability highlights the heterogeneity of clinical isolates and the influence of genetic determinants such as efflux pumps, porin loss, and enzyme-mediated resistance. The observed resistance even to certain plant extracts suggests that while phytochemicals are promising, they are not universally effective and require targeted optimization. Similar strain-dependent variability has been reported in studies evaluating plant extracts against multidrug-resistant bacteria (Okoye *et al*. 2013; Okoye *et al.* 2014; Okoye *et al*., 2016a; 2016b; O'Neill, 2022).

The antibacterial activity against *K. quasipneumoniae* was restricted to higher concentrations (200 – 400 mg/mL), with no activity observed at lower doses. The maximum inhibition zones—17.00 ± 0.10 mm for hexane extract of *A. paniculata* and 14.00 ± 0.20 mm for hexane extract of *J. curcas*—suggest moderate susceptibility relative to *K. pneumoniae*. This reduced sensitivity may reflect intrinsic resistance mechanisms or differences in cell envelope composition. The requirement for higher concentrations indicates a bacteriostatic threshold that must be exceeded for observable activity, emphasizing the importance of dosage optimization in therapeutic applications (Umennadi *et al*. 2026a; 2026b).

The MIC values (50 – 200 mg/mL for *K. pneumoniae*; 100–200 mg/mL for *K. quasipneumoniae*) and MBC values (100 – 400 mg/mL and 200 – 400 mg/mL, respectively) confirm the antibacterial potency of the extracts, although relatively high concentrations are required compared to conventional antibiotics. The MIC/MBC ratios suggest that many extracts exhibit bactericidal effects at higher concentrations, an

important criterion for therapeutic relevance. These findings are consistent with previous reports that plant extracts often require higher doses due to their crude nature but can achieve bactericidal activity when sufficiently concentrated (Cos *et al*., 2006; Kuete, 2010; Umeh *et al*., 2020; 2021).

The antibacterial activity observed in this study likely results from a combination of mechanisms, including disruption of bacterial membranes, inhibition of protein and nucleic acid synthesis, and interference with metabolic pathways. Importantly, the multi-component nature of plant extracts may reduce the risk of resistance development and enhance efficacy through synergistic interactions. Given the demonstrated activity against ciprofloxacin-resistant strains, these extracts hold promise as templates for the development of novel antimicrobial agents or as adjuvants to existing antibiotics.

Conclusion

Leaf extracts of *Andrographis paniculata* and *Jatropha curcas* demonstrated notable antibacterial activity against *Klebsiella pneumoniae* and *Klebsiella quasipneumoniae*, with methanol extracts showing the highest efficacy. Some extracts outperformed Ciprofloxacin against resistant strains, highlighting their potential as alternative antimicrobial agents. However, their activity was concentration-dependent, indicating the need for further purification and optimization for clinical application.

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