https://chemclassjournal.com/
ChemClass Journal Vol. 9 Issue 4 (2025); 181 - 193
e-ISSN:3092-8214 p-ISSN:3092-8206
doi.org/10.33003/chemclass-2025-0904/13

Phytochemical Profiling and In-vitro Antimicrobial Evaluation of *Euphorbia dendroides* Pax Leaf Extracts against Multidrug-Resistant Microorganisms

***Lawal Abubakar[1], Muhammad Sirajo[1], Umar Abubakar Birnin Yauri[2], Yunusa Buhari Idris[1]**

[1]*Faculty of Science, Department of Pure and Industrial Chemistry, Sokoto State University, Sokoto, Nigeria*
[2]*Faculty of Chemical and life Sciences, Department of Pure and Environmental Chemistry, Usmanu Danfodiyo University, Sokoto, Nigeria.*

***Corresponding author:** Email: abbakarlawal14@gmail.com.

Abstract

Antimicrobial resistance (AMR) poses a critical global health challenge, prompting the search for alternative sources of effective antimicrobial agents. This study evaluated the phytochemical constituents and antimicrobial activities of *Euphorbia dendroides* Pax leaf extracts using n-hexane, ethyl acetate, and methanol as solvents. The leaves were collected from Kasarawa, Sokoto State, Nigeria, authenticated, and subjected to extraction via maceration. Phytochemical screening revealed the presence of bioactive constituents such as flavonoids, alkaloids, saponins, tannins, steroids, and diterpenoids, particularly concentrated in the ethyl acetate and methanol extracts. Antimicrobial activity was assessed against a panel of bacteria and fungi using the agar diffusion method, with standard drugs (Ampicillin and Ketoconazole) as controls. The ethyl acetate extract exhibited the strongest and broadest antimicrobial activity, with notable inhibition against *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella typhi*, while fungi such as *Candida krusei* and *Aspergillus flavus* showed resistance. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) confirmed the potency of the ethyl acetate extract at low concentrations. These findings suggest that *E. dendroides* Pax is a promising source of natural antimicrobial compounds that could serve as leads in the development of plant-based therapeutics to combat resistant pathogens.

Keywords: AMR, Bioactive compounds, *Euphorbia dendroides* Pax, medicinal plant.

Introduction

Antimicrobial resistance (AMR) is a global health challenge, with the emergence of resistant strains outpacing the development of new antibiotics [1]. This crisis necessitates the discovery of new antimicrobial agents from alternative sources, such as plants, which have historically provided a wealth of bioactive compounds [2]. The increasing prevalence of antibiotic-resistant microorganisms necessitates the search for new antimicrobial agents from natural sources such as *Euphorbia dendroides* [3]. *E.dendroides* Pax, a species within the *Euphorbia* genus, is known for its medicinal properties but remains underexplored for its antimicrobial potential. Traditional uses of *E.*

© CSN Zaria Chapter

dendroides Pax in treating infections suggest that it may contain valuable bioactive constituents [4]. This study seeks to scientifically investigate the antimicrobial properties of the plant's leaves. The figure 1 shows a close-up of the leaves of plant, which appears to be from the *Euphorbia* genus, *E .dendroides* Pax. *E. dendroides*, commonly known as tree spurge in English language, *Aguwa* in Hausa language, *Bawo* in Fulfude, *Egun-oro* in Yoruba language and *Kaska* in Kanuri language. It is a perennial shrub known for its distinctive green foliage and potential medicinal properties. This plant is native to the Mediterranean region and has been used in traditional medicine for treating various ailments, including skin conditions and inflammation. The leaves of *E. dendroides* are narrow and elongated, with a slightly waxy texture. They are arranged alternately along the stems. The plant is known known for producing milky latex containing various bioactive compounds, including diterpenes and flavonoids, which have shown potential antimicrobial, anti-inflammatory, and antioxidant activities in preliminary studies. In the context of research, *E. dendroides* is of interest for its potential pharmacological applications, particularly in isolating and characterizing active antimicrobial constituents. The bioactive compounds derived from its leaves could contribute to the development of new natural products for medicinal use, including treatments for microbial infections [5].

The plant is traditionally used in folk medicine, with its extracts being applied topically or consumed to treat various ailments. The focus on the leaves in the image emphasizes their importance as a source of these therapeutic compounds. The collection and study of such plant materials form a crucial part of natural product research aimed at discovering new drugs derived from plants. In the context of modern drug development, the isolation and characterization of active constituents from *E. dendroides,* leaves can lead to the discovery of novel antimicrobial agents. These agents are especially valuable in the face of rising antibiotic resistance, offering alternative solutions from nature to combat pathogenic microbes [5].



Figure 1: Picture of *Euphorbia dendroides* Pax

Ethodology

Sample Collection and Treatment

The leaves of *E. dendroides* Pax were collected in Kasarawa Area, Wamakko Local Government, Sokoto State, Nigeria. The plant material was identified and authenticated by experts at the Herbarium of the Botany Unit, Department of Biological Sciences, Usmanu Danfodio University, Sokoto, Sokoto State, Nigeria. A voucher specimen was preserved for future reference. To prepare the samples, the leaves underwent a thorough washing process: - 2-3

times with tap water to remove dirt and debris, - Once with sterile distilled water to eliminate any remaining impurities.

After washing, the leaves were shade-dried for 7 days to remove excess moisture. This careful process ensured the quality and integrity of the plant material for further analysis. The plant sample was chopped into small pieces using a mortar and pestle, then ground into powdered form.

Sample Extraction

Five hundred and sixty grams (560g) of the powdered sample was macerated with 2.5 liters of analytical grade n-hexane for 72 hours. Stirring was performed occasionally to enhance the extraction. The n-hexane solvent with its extracted components was decanted and filtered with Whatman-1 filter paper. The filtrate was air dried at room temperature to obtained n-Hexane extract. The same procedure was employed using ethyl acetate and methanol to obtain the ethyl acetate extract and the methanol extract, respectively. The percentage yield of the extract was calculated using the following formula in equation.

$$\% \text{ yield } = \frac{\text{weight of extract}}{\text{sample weight}} X\ 100 \quad ---\text{Eqn 1}$$

Preliminary Phytochemical Screening

Portion (1g) of the extract was subjected to phytochemical screening for the presence of secondary metabolites including, flavonoids, saponins, tannins, steroids/triterpenes and alkaloids, according to standard procedures [6].

Antimicrobial Studies

The antimicrobial activity of the methanolic stem bark extract of *E. dendroides* was first determined through susceptibility test using agar diffusion. 0.2g of the extract was weighed and dissolved in 10 ml of dimethyl sulfoxide (DMSO) to obtain a concentration of 20 mg/ml. Mueller Hinton agar would be the medium used as the growth medium for the microbes. The medium would be prepared according to the manufacturer's instructions and sterilized at 121oC for 15 minutes. It would be poured into sterile petri dishes, allowed to cool and solidify. The sterilized medium would be seeded with 0.1ml of standard inoculum of the test microbe; the inoculum would be spread evenly over the surface of the medium by the use of a sterile swab. A standard sterile cork borer of 6mm in diameter would be used to bore a well at the centre of each inoculated medium. The wells would be filled with 0.1ml of the solution of the extract (2mg) and allowed to diffuse for I hour. Incubation of the inoculated medium would be made at 37oC for 24hrs, after which the medium would be observed for the zone of inhibition of growth; the tests would be done in duplicates and the zone of inhibition would be measured with a transparent ruler. The mean of the results would be recorded in millimeters (mm) [7].

Test Organisms

Clinical isolates of *Methicillin Resistant Staphylococcus aureus* (MRSA), *Vancomycin Resistant Enterococci, Staphylococcus aureus, Streptococcus faecalis, Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Salmonella typhi, Cryptococcus neoformans, Candida albicans, Candida krusei, Aspergillus fumigatus, Aspergillus nigre,* and *Aspergillus flavus.*

would be obtained from the Department of Medical Microbiology, Ahmadu Bello University Teaching Hospital, Zaria, Nigeria. All the isolates would be checked for purity and maintained in slants of nutrient agar (for bacteria) and in slants of sabouraud dextrose agar for fungi [8].

Preparation of Test Organism

Test organisms were sub-cultured and grown on 10 mL Sabouraud dextrose agar slants and were eventually kept in the refrigerator.

Preparation of Extract for Antimicrobial Studies

The stock concentration of the extracts, 1000 µg/cm^3, 1.0 mg of the extracts was dissolved in 10 cm^3 of dimethyl sulfoxide (DMSO) and a two-fold serial dilution was carried out to obtain four more concentrations of 500, 250, 125 and 62.5 µg/cm^3. The growth medium and Mueller Hinton agar were prepared in accordance with the manufacturer's instructions and sterilized at 121 °C for 15 minutes.

Preparation of Reference Antimicrobial Agents

The stock concentration of the Ampicillin and Ketoconazole (62.5 µg/cm^3) was prepared by dissolving 0.0625 mg of the powered in 10 mL of dimethyl sulfoxide (DMSO).

Antimicrobial Screening

An agar diffusion technique susceptibility test was used to evaluate the antibacterial activity of the extracts of the sample [8]. for the antibacterial and antifungal tests, nutrient broth and sabraud dextrose broth were utilized, respectively. For 24 hours for bacteria and 48 hours for fungi, the test organisms were inoculated and incubated. At 45 °C, 0.1 cm^3 of the test microbe's standard inoculum was added to the petri dish's solidified sterile media. A 6 mm cork borer was used to drill wells into the hardened inoculation nutrient agar plates. A 0.1 cm^3 dimethyl sulfoxide solution of the extract was added to each well. As a control, discs with blank extraction solutions were used. Positive controls were reference antibacterial medications (Ampicillin) and antifungal medications (Ketoconazole).

The standard drugs and extract were added to the agar and allowed to diffuse overnight. For bacteria and fungi, respectively, all incubations took taken place at 37°C and 25 °C. The diameter of the inhibitory zone was measured using a transparent ruler at the conclusion of the incubation period and documented. The zones of microbial growth inhibition were examined twice,

and the millimeter (mm) value of the mean result was recorded.

Determination of Minimum Inhibitory Concentration (MIC)

The Broth dilution method was used to determine the extract's minimum inhibitory concentration (MIC) [9]. In accordance with the manufacturer's instructions, Mueller Hinton broth was made; 2.0 cm^3 of the medium would be poured into test tubes with screw caps, sterilized at 121 °C for 15 minutes, and then allowed to cool. McFarland's standard turbidity scale number 0.5 was also prepared. Sterile normal saline was used to progressively dilute the organism suspension until, when compared visually, the turbidity matched McFarland's scale. The extract in the sterile broth was serially diluted twice to determine the concentrations. Then, 0.1 cm^3 of the test microbe's standard inoculum was added to the broth at various concentrations. The plates were then be checked for turbidity (growth) after the tubes were incubated for 24 hours at 37 °C for bacteria and 48 hours at 25 °C for fungi. The minimum extract concentration (MIC) was determined by measuring how much of each microorganism was unable to grow visibly.

Determination of Minimum Bactericidal/Fungicidal Concentrations (MBC/MFC)

The Mueller Hinton agar broth was made, sterilized for 15 minutes at 121 °C, and then cooled and solidified on sterile petri dishes. According to Ali [10]. The MBC/MFC plate was the one with the lowest concentration of the extract in the serial dilution without colony growth. The contents of the MIC in the serial dilution were sub-cultured into the prepared medium and incubated at 37 °C for 24 hours. The plates would then be observed for colony growth.

Results and Discussions

Percentage Yields: n-Hexane extract showed the highest yield (12.42%), followed by Ethylacetate (9.82%) and Methanol (8.24%).The result is presented in Table 1.

Table 1: The percentage yield of the extracts (%)

Solvent	Weight (g)	Yield (%)	Colour
n-Hexane	69.56	12.42	Green
Ethylacetate	55.04	9.82	Dark Green
Methanol	46.16	8.24	Pale Green

Phytochemical Screening: Flavoniods, Tannins, Saponins, Alkaloids, Cardiac glycoside, Steroids, Antharaquinones, Carbohydrates, Diterpenoids, Protein was detected in the ethylacetate extract. Flavonoids have the biological activity of Antidiabetic, anti-inflammatory, antioxidant, antimicrobial, anticancer [11].Tannins Antioxidant, antimicrobial, astringent, effects on gut health [12].Alkaloids Antimicrobial, anticancer, anti-inflammatory, cytotoxic [13].

The result was presented in Table 2.

Table 2: Phytochemical Screening of *Euphorbia dendroides* Pax leaves extract

Constituents	**Test**	**Observation**	**N-Hexane Extract**	**Ethylacetate Extract**	**Methanol Extract**
Steroids/Triterpenes	Salkowski test	Reddish brown color was observed	+	+	+
	Lieberman-Buchard		+	+	+
Cardiac glycosides	Killerkilearni test	Green ring at the interface was observed	+	+	+
Anthraquinones	Borntrager's test	Emerald green color was formed	-	-	-
Carbohydrates	Molisch's test	Reddish browning at the interface was observed	-	-	-
	Fehling's test		-	+	+
Saponins	Frothing test	Red color was observed	-	+	+
Flavonoids	Ferric chloride test	Violet ring color was observed	-	+	+
	NaOH test		-	+	+
	Shinoida's test		-	+	+
Tannins	Ferric chloride test	Brick-red precipitates was observed	-	+	+
	Lead acetate test		-	+	+
Alkaloids	Mayers test	Honey comb froth was observed	-	+	
	Hager's test		-	+	+
	Wagner's test		-	+	+
Diterpenoids	Copperacetate test	Greenish black color was observed	-	+	+
Proteins	Xanthoprotics test	Yellow color was observed	-	-	+

Key: (+) = Detected (-) = Not Detected

Antimicrobial Activity

Table 3: The anti-microbial activities of plant extract. Table 3 shows the microbial activities of the *Euphorbia dendroides* Pax leaves extract

Test Organisms	EA	ME	HE
Methicillin Resist Staph aureus	S	S	S
Vancomycin resist enterococci	R	R	R
Staphylococcus aureus	S	S	S
Streptococcus feacalis	R	R	R
Escherichia coli	S	S	S
Klebsiella pneumoniae	S	S	S
Pseudomonas aeruginosa	S	S	S
Salmonella typhi	S	S	R
Cryptococcus neoformans	R	R	R
Candida albicans	S	S	S
Candida krusei	R	R	R
Aspergillus fumigatus	R	R	R
Aspergillus nigre	S	S	R
Aspergillus flavus	R	R	R

Key: S= Sensitive, R= Resistance

Ethyl Acetate extract had the broadest antimicrobial spectrum, making it the most promising; methanol extract was moderately effective; hexane extract had the weakest activity, with limited effectiveness; the most susceptible organisms were *Staph. aureus* strains and *E. coli*, which were sensitive to all three extracts, Fungi, *Candida krusei*, *Aspergillus fumigatus*, and *Aspergillus flavus* were consistently resistant.

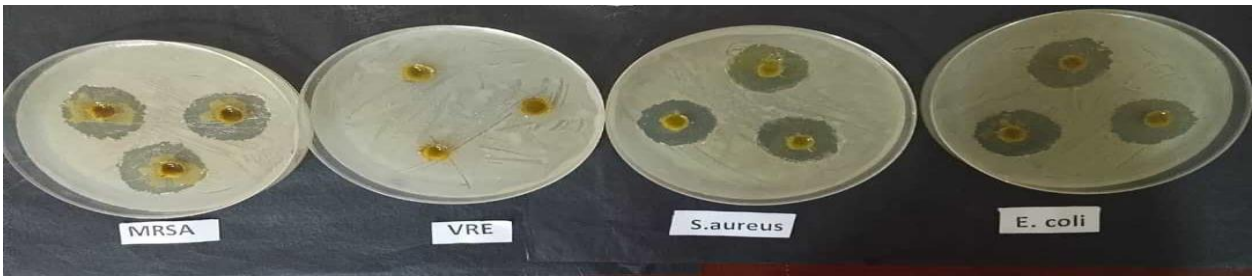


Plate 1: A Plate from antimicrobial screening

Table 4: Zone of inhibition of the extract against the test microorganisms

Test Organisms	EA	ME	HE
Methicillin Resist Staph aureus	22.67±0.58	20.33±0. 58	17.67±0.58
Vancomycin resist enterococci	0.00±0. 00	0.00±0. 00	0.00±0.00
Staphylococcus aureus	21.33±0. 58	17.67±0. 58	16.33±0.58
Streptococcus feacalis	0.00±0. 00	0.00±0. 00	0.00±0.00
Escherichia coli	21.33±1. 16	18.00±1. 63	17.33±058
Klebsiella pneumoniae	20.33±0. 58	18.67±0. 58	16.00±0.82
Pseudomonas aeruginosa	24.67±0. 58	19.33±0. 58	17.67±0.58
Salmonella typhi	21.33±0. 58	18.33±0. 58	0.00±0.00
Cryptococcus neoformans	0.00±0. 00	0.00±0. 00	0.00±0.00
Candida albicans	23.67±0. 58	21.33±0. 58	18.00±1.63
Candida krusei	0.00±0. 00	0.00±0. 00	0.00±0.00
Aspergillus fumigatus	0.00±0. 00	0.00±0. 00	0.00±0.00
Aspergillus nigre	21.00±1. 00	18.33±0. 58	0.00±0.00
Aspergillus flavus	0.00±0. 00	0.00±0. 58	0.00±0.00

Key: = The values are Mean, ±Standard deviation of triplicates, HE =hexane extract, EA= Ethyl acetate extract and ME= Methanol extract

Table 4: Zone of Inhibition results for three different plant extracts: Ethylacetate (EA), Methanol (ME), and Hexane (HE) against various microorganisms. This test measures the diameter (in mm) of the clear zone around a sample where bacterial or fungal growth is inhibited, showing the effectiveness of the extract

Minimum Inhibitory Concentration (MIC)

Table 5: Minimum inhibitory concentration of the extract against the test microorganism

	EDL (EA)					EDL (ME)					EDL (HE)				
	Zone of inhibition (mg/ml)														
Test organisms	**10**	**5**	**2.5**	**1.25**	**0.65**	**10**	**5**	**2.5**	**1.25**	**0.65**	**10**	**5**	**2.5**	**1.25**	**0.65**
Methicillin resist Staph aureus	-	-	0*	+	++	-	-	0*	+	++	-	0*	+	++	+++
Vancomycin resist enterococci															
Staphylococcus aureus	-	-	0*	+	++	-	0*	+	++	+++	-	0*	+	++	+++
Streptococcus feacalis															
Escherichia coli	-	-	0*	+	++	-	0*	+	++	+++	-	0*	+	++	+++
Klebsiella pneumonia	-	-	0*	+	++	-	0*	+	++	+++	-	0*	+	++	+++
Pseudomonas aeruginosa	-	-	0*	+	++	-	-	0*	+	++	-	0*	+	++	+++
Salmonella typhi	-	-	0*	+	++	-	0*	+	++	+++					
Cryptococcus neoformans															
Candida albicans	-	-	0*	+	++	-	-	0*	+	++	-	0*	+	++	+++
Candida krusei															

Aspergillus fumigates											
Aspergillus nigre	-	-	0*	+	++	-	0*	+	++	+++	
Aspergillus flavus											

Key=> - => No turbidity (no growth), 0*=>MIC, + => Turbid (light growth), ++ => Moderate turbidity.

The Minimum Inhibitory Concentration (MIC) of three different plant extracts EA (Ethyl Acetate), ME (Methanol), and HE (Hexane) tested against various bacterial and fungal organisms. The goal is to determine the lowest concentration of extract that can inhibit microbial growth. The results indicate that methanol and ethyl acetate extracts have stronger antimicrobial activity than the hexane extract. These findings suggest potential for developing plant-based antimicrobials, particularly against gram-positive bacteria and some fungi.

Minimum Bacterial Concentration (MBC)

Table 6 showed the Minimum Bactericidal/Fungicidal Concentration (MBC/MFC) of three different plant extracts (EA, ME, HE) against various microorganisms. It evaluates how increasing concentrations of each extract (from 0.63 mg/ml to 10 mg/ml) affect microbial growth. EA (Ethyl Acetate) extract is the most potent across bacteria and fungi, followed by ME (Methanol) and then HE (Hexane), higher concentrations (≥10 mg/ml) are required for significant antimicrobial effects, these extracts show promise as broad-spectrum antimicrobials, especially EA against resistant bacteria and yeasts.

Table 6: Minimum bactericidal/fungicidal concentration of the extract against the test microorganism

	EDL (E.A)					EDL (Me)					EDL (Hex)				
	Zone of inhibition (mg/ml)														
Test organisms	**10**	**5**	**2.5**	**1.25**	**0.65**	**10**	**5**	**2.5**	**1.25**	**0.65**	**10**	**5**	**2.5**	**1.25**	**0.65**
Methicillin resist Staph aureus	0*	+	++	+++	+++	0*	+	++	+++	+++	0*	+	++	+++	+++
Vancomycin resist enterococci															
Staphylococcus aureus	0*	+	++	+++	+++	0*	+	++	+++	+++	0*	+	++	+++	+++
Streptococcus feacalis															
Escherichia coli	0*	+	++	+++	+++	0*	+	++	+++	+++	0*	+	++	+++	+++
Klebsiella pneumonia	0*	+	++	+++	+++	0*	+	++	+++	+++	0*	+	++	+++	+++
Pseudomonas aeruginosa	-	0*	+	++	+++	0*	+	++	+++	+++	0*	+	++	+++	+++
Salmonella typhi	0*	+	++	+++	+++	0*	+	++	+++	+++					
Cryptococcus neoformans															
Candida albicans	-	0*	+	++	+++	0*	+	++	+++	+++	0*	+	++	+++	+++
Candida krusei															
Aspergillus fumigates															
Aspergillus nigre	0*	+	++	+++	+++	0*	+	++	+++	+++					
Aspergillus flavus															

KEY: - =>No Colony Growth, 0* =>MBC/MFC, + =>Scanty colonies growth, ++ =>Moderate colonies growth, +++ =>Heavy colonies growth

Table 7 shows that sensitivity (S) or resistance (R) of various test micro-organisms to four drugs: Ciprofloxacin, Sparfloxacin, Fluconazole, and Fulcin. This is a qualitative assessment, complementing Table 8's quantitative zone of inhibition. Each drug shows organism-specific effectiveness.

Table 7: Control against the test micro-organisms

Test Organisms	Ciprofloxacin	Sparfloxacin	Fluconazole	Fulcin
Methicillin Resist Staph aureus	R	S	R	R
Vancomycin resist enterococci	S	R	R	R
Staphylococcus aureus	R	S	R	R
Streptococcus feacalis	S	R	R	R
Escherichia coli	S	R	R	R
Klebsiella pneumoniae	R	S	R	R
	R	S	R	R
Pseudomonas aeruginosa	S	R	R	R
Salmonella typhi	R	R	S	R
Cryptococcus neoformans	R	R	S	R
Candida albicans	R	R	S	R
Candida krusei	R	R	R	S
Aspergillus fumigatus	R	R	R	S
Aspergillus nigre	R	R	R	S
Aspergillus flavus				

Key: S= Sensitive, R= Resistance

- Ciprofloxacin and Sparfloxacin are useful for bacterial infections.
- Fluconazole is ideal for yeast infections.
- Fulcin is specific for **mold infections** (especially *Aspergillus* species)

Table 8 showed that the zone of inhibition (a measure of antimicrobial activity) of four drugs Ciprofloxacin, Sparfloxacin, Fluconazole, and Fulcin against various test micro-organisms, including both bacteria and fungi. A larger zone (in millimeters) indicates a stronger inhibitory effect of the drug on the microorganism. Ciprofloxacin and Sparfloxacin are effective against bacteria (but with different target ranges). Fluconazole is effective against yeast-like fungi (e.g., *Candida*, *Cryptococcus*). Fulcin is effective against mold fungi (*Aspergillus* species). No single drug is effective against all organisms; activity is pathogen-specific.

The ethyl-acetate extract of *Euphorbia dendroides* leaves exhibited the broadest and strongest in vitro antimicrobial activity among tested extracts, with meaningful inhibition of *S. aureus* (including MRSA), *E. coli*, *P. aeruginosa*

and *S. typhi* (zones ≈17–25 mm and low mg/mL MICs). These findings align with ethnomedicinal uses of *E. dendroides* for treating skin and infection-related ailments and are concordant with recent literature Smeriglio [14] and Benjamaa [15] that identifies diterpenes, phenolics and flavonoids in *Euphorbia* species as probable antimicrobial agents. While encouraging and comparable to published Euphorbia data, translation toward therapeutics requires isolation of active constituents, toxicity evaluation (given known Euphorbia latex irritancy), and mechanism-of-action studies.

Table 8: Zone of inhibition of the drug against the test micro-organisms

Test Organisms	Ciprofloxacin	Sparfloxacin	Fluconazole	Fulcin
Methicillin Resist Staph aureus	0	32	0	0
Vancomycin resist enterococci	32	0	0	0
Staphylococcus aureus	0	32	0	0
Streptococcus feacalis	31	0	0	0
Escherichia coli	35	0	0	0
Klebsiella pneumoniae	0	32	0	0
Pseudomonas aeruginosa	0	30	0	0
Salmonella typhi	40	0	0	0
Cryptococcus neoformans	0	0	32	0
Candida albicans	0	0	34	0
Candida krusei	0	0	33	0
Aspergillus fumigatus	0	0	0	30
Aspergillus nigre	0	0	0	31
Aspergillus flavus	0	0	0	30

Conclusion

The findings of this research revealed that *Euphorbia dendroides* Pax leaves possess significant antimicrobial potential, attributable to the diverse phytochemicals identified in its extracts. Among the three solvent extracts, the ethyl acetate extract exhibited the most pronounced inhibitory activity against a wide range of bacterial species, including *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella typhi*. The presence of bioactive compounds such as flavonoids, alkaloids, saponins, tannins, and steroids suggests that these constituents contribute synergistically to the observed antimicrobial effects. The methanol extract showed moderate activity, while the n-hexane extract displayed minimal inhibition, highlighting the importance of solvent polarity in optimizing the extraction of bioactive molecules. Overall, *E. dendroides* Pax demonstrates considerable promise as a natural source of antimicrobial agents capable of addressing the

growing challenge of multidrug-resistant pathogens.

REFERENCE

[1]. Idris, Y. B., Sirajo, M., Hassan, L. G., Izuagie, T., Tukur, M., & Abubakar, A. U. (2025). Millipede secretions as a potential source of antibiotics: A mini review. *Caliphate Journal of Science and Technology*, *7*(1), 44-54.

[2]. Abdallah, E. M., Alhatlani, B. Y., de Paula Menezes, R., & Martins, C. H. G. (2023). Back to nature: Medicinal plants as promising sources for antibacterial drugs in the post-antibiotic era. *Plants*, *12*(17), 3077.

[3]. Guedes, B. N., Krambeck, K., Durazzo, A., Lucarini, M., Santini, A., Oliveira, M. B. P., ... & Souto, E. B. (2024). Natural antibiotics against antimicrobial resistance: sources and bioinspired delivery systems. *Brazilian Journal of Microbiology*, *55*(3), 2753-2766.

[4]. Benjamaa, R., Moujanni, A., Kaushik, N., Choi, E. H., Essamadi, A. K., & Kaushik, N. K. (2022). Euphorbia species latex: A comprehensive review on phytochemistry and biological activities. *Frontiers in plant science*, *13*, 1008881.

[5]. Hassan, A. R. (2022). Chemical profile and cytotoxic activity of a polyphenolic-rich fraction from Euphorbia dendroides aerial parts. *South African Journal of Botany*, *147*, 332-339.

[6].Shaikh, J. R., & Patil, M. (2020). Qualitative tests for preliminary phytochemical screening: An overview. *International journal of chemical studies*, *8*(2), 603-608.

[7]. Asfa, B., Woldemichael, D. N., Tesfaw, L., Asefa, L., Desta, S., Girma, S., ... & Tufa, T. B. (2025). Evaluating antimicrobial activity of selected medicinal plant extracts against pasteurellosis-causing bacteria in small ruminants. *Frontiers in Veterinary Science*, *12*, 1563208.

[8].Sharma, A., & Kaur, A. (2025). Catharanthus roseus leaf-based green synthesis of silver oxide nanoparticles: Characterization, phytochemicals screening and antimicrobial activity. *The Microbe*, 100469.

[9]. Rakholiya, K. D., Kaneria, M. J., Shaikh, A. M., Wandhekar, S. S., & Chanda, S. V. (2025). Medicinal plants as alternative sources of therapeutics against multidrug resistant pathogenic microorganisms based on their antimicrobial potential and synergistic properties. *Fighting multidrug resistance with herbal extracts, essential oils and their components*, 271-289.

[10]. M. Ali, S. U. Diso, S. A. Waiya, and M. S. Abdallah, 'Phytochemical Screening and Antibacterial Activity of Bitter Leaf (Vernonia amygdalina)', *Annals of Microbiology and Infectious Diseases*, vol. 2(4): 1–7, 2019, doi: 10.22259/2637-5346.0204001.

[11]. Shamsudin, N. F., Ahmed, Q. U., Mahmood, S., Shah, S. A. A., Sarian, M. N., Khattak, M. M. A. K., ... & Latip, J. (2022). Flavonoids as antidiabetic and anti-inflammatory agents: A review on structural activity relationship-based studies and meta-analysis. *International journal of molecular sciences*, *23*(20), 12605.

[12]. Cosme, F., Aires, A., Pinto, T., Oliveira, I., Vilela, A., & Gonçalves, B. (2025). A comprehensive review of bioactive tannins in foods and beverages: functional properties, health benefits, and sensory qualities. *Molecules*, *30*(4), 800.

[13]. Dar, R. A., Shahnawaz, M., Ahanger, M. A., & Majid, I. U. (2023). Exploring the diverse

bioactive compounds from medicinal plants: a review. *J. Phytopharm*, *12*(3), 189-195.

[14]. Smeriglio, A., Denaro, M., Trombetta, D., Ragusa, S., & Circosta, C. (2021). New insights on Euphorbia dendroides L.(Euphorbiaceae): polyphenol profile and biological properties of hydroalcoholic extracts from aerial parts. *Plants*, *10*(8), 1621.

[15].Benjamaa, R., Moujanni, A., Kaushik, N., Choi, E. H., Essamadi, A. K., & Kaushik, N. K. (2022). Euphorbia species latex: A comprehensive review on phytochemistry and biological activities. *Frontiers in plant science*, *13*, 1008881.