

Zenoh et al. Afr. J. Clin. Exper. Microbiol. 2024; 25 (2): 219 - 226

<https://www.afrcem.org>African Journal of Clinical and Experimental Microbiology. ISSN 1595-689X  
AJCEM/2329. <https://www.ajol.info/index.php/ajcem>

Apr 2024; Vol.25 No.2

Copyright AJCEM 2024: <https://dx.doi.org/10.4314/ajcem.v25i2.13>**Original Article****Open Access****Evaluation of antimicrobial properties of five medicinal plants used against bacterial infections in Jalingo, Nigeria**<sup>\*1</sup>Zenoh, D. A., <sup>2</sup>Josephus, B., <sup>3</sup>Halley, N., <sup>1</sup>Endurance Okpan., <sup>1</sup>Henry Chukwuemeka., and <sup>1</sup>Akumbo Gemenen<sup>1</sup>Department of Medical Laboratory Science, Faculty of Health Sciences, Taraba State University, Jalingo, Nigeria<sup>2</sup>Department of Nursing Science, Faculty of Health Sciences, Taraba State University, Jalingo, Nigeria<sup>3</sup>Federal Medical Center Jalingo, Taraba State, Nigeria\*Correspondence to: [zenoh.d@tsuniversity.edu.ng](mailto:zenoh.d@tsuniversity.edu.ng); +234 8053852525**Abstract:****Background:** The prevalent utilization of medicinal plants in communities underscores their promise as antimicrobial agents amid rising antibiotic resistance. This study assesses five medicinal plants; *Bambusa vulgaris*, *Hibiscus sabdariffa*, *Heteropogon contortus*, *Moringa oleifera*, and *Carica papaya* against clinical isolates of *Salmonella* Typhi and *Shigella dysenteriae*.**Methodology:** Five medicinal plants were chosen based on traditional knowledge and ethnobotanical practices. Phytochemical analysis followed standard methods. Plant extracts were prepared using ethanol, ethyl acetate, dichloromethane, and hexane. Various concentrations (R conc., D1 conc., D2 conc, D3 conc, and D4 conc) of the extracts were evaluated using Kirby-Bauer disk diffusion and broth dilution methods to ascertain antimicrobial properties, including minimum inhibitory concentrations (MIC) and minimum bactericidal concentrations (MBC).**Results:** Phytochemical analysis revealed abundant saponins, cardiac glycosides, terpenoids, steroids, flavonoids, phenolics, and tannins, notably higher with ethanol extraction. *Hibiscus sabdariffa* demonstrated potent activity against *S. Typhi* with inhibition zone diameters of 29.00 mm (R conc), 27.00 mm (D1 conc), 14.00 mm (D2 conc), and 4.00 mm (D3 conc). *Heteropogon contortus* exhibited activity against *S. dysenteriae* with inhibition zone diameter of 25.05 mm (R conc), 15.00 mm (D1 conc), 10.00 mm (D2 conc), and 5.00 mm (D3 conc). The inhibition zone diameters of *B. vulgaris* were 18.50 mm (R conc), 17.00 mm (D1 conc), and 10.00 mm (D2 conc) against *S. dysenteriae*. The MIC and MBC were similar for both organisms, with *H. sabdariffa* (MIC: D3-4.27 mg/mL, MBC: D1-68.25 mg/mL) and *H. contortus* (MIC: D3-4.69 mg/mL, MBC: R-75.00 mg/mL), while *M. oleifera*, *C. papaya*, and *B. vulgaris* had negligible antimicrobial activity.**Conclusion:** *Hibiscus sabdariffa* and *H. contortus* exhibited potent antimicrobial effects against *Salmonella*, with MICs of 4.27 mg/mL and 4.69 mg/mL, and MBCs of 68.25 mg/mL and 75.00 mg/mL respectively. Their consistent low MICs against *Shigella* suggest their potentials for antibiotic production.**Keywords:** Antimicrobial agent; Antibiotic resistance; Plant extracts; MIC; MBC

Received Jan 19, 2024; Revised Feb 17, 2024; Accepted Feb 22, 2024

Copyright 2024 AJCEM Open Access. This article is licensed and distributed under the terms of the Creative Commons Attribution 4.0 International License <http://creativecommons.org/licenses/by/4.0/>, which permits unrestricted use, distribution and reproduction in any medium, provided credit is given to the original author(s) and the source. Editor-in-Chief: Prof. S. S. Taiwo**Évaluation des propriétés antimicrobiennes de cinq plantes médicinales utilisées contre les infections bactériennes à Jalingo, Nigeria**<sup>\*1</sup>Zenoh, Z. A., <sup>2</sup>Josephus, B., <sup>3</sup>Halley, N., <sup>1</sup>Endurance Okpan., <sup>1</sup>Henry Chukwuemeka., et <sup>1</sup>Akumbo Gemenen<sup>1</sup>Département des Sciences de Laboratoire Médical, Faculté des Sciences de la Santé, Université d'État de Taraba, Jalingo, Nigeria<sup>2</sup>Département des Sciences Infirmières, Faculté des Sciences de la Santé, Université d'État de Taraba, Jalingo, Nigeria<sup>3</sup>Centre Médical Fédéral Jalingo, État de Taraba, Nigeria\*Correspondance à: [zenoh.d@tsuniversity.edu.ng](mailto:zenoh.d@tsuniversity.edu.ng); +234 8053852525

## Résumé:

**Contexte:** L'utilisation répandue des plantes médicinales dans les communautés souligne leur promesse en tant qu'agents antimicrobiens dans un contexte de résistance croissante aux antibiotiques. Cette étude évalue cinq plantes médicinales; *Bambusa vulgaris*, *Hibiscus sabdariffa*, *Heteropogon contortus*, *Moringa oleifera* et *Carica papaya* contre les isolats cliniques de *Salmonella* Typhi et *Shigella dysenteriae*.

**Méthodologie:** Cinq plantes médicinales ont été choisies sur la base des connaissances traditionnelles et des pratiques ethnobotaniques. L'analyse phytochimique a suivi les méthodes standard. Des extraits de plantes ont été préparés en utilisant de l'éthanol, de l'acétate d'éthyle, du dichlorométhane et de l'hexane. Diverses concentrations (R conc., D1 conc., D2 conc., D3 conc et D4 conc) des extraits ont été évaluées à l'aide des méthodes de diffusion sur disque Kirby-Bauer et de dilution en bouillon pour vérifier les propriétés antimicrobiennes, y compris les concentrations minimales inhibitrices (CMI) et les concentrations minimales concentrations bactéricides (MBC).

**Résultats:** L'analyse phytochimique a révélé une abondance de saponines, de glycosides cardiaques, de terpénoïdes, de stéroïdes, de flavonoïdes, de composés phénoliques et de tanins, notamment plus élevés avec l'extraction à l'éthanol. *Hibiscus sabdariffa* a démontré une activité puissante contre *S. Typhi* avec des diamètres de zone d'inhibition de 29,00 mm (conc R), 27,00 mm (conc D1), 14,00 mm (conc D2) et 4,00 mm (conc D3). *Heteropogon contortus* a présenté une activité contre *S. dysenteriae* avec un diamètre de zone d'inhibition de 25,05 mm (R conc), 15,00 mm (D1 conc), 10,00 mm (D2 conc) et 5,00 mm (D3 onc). Les diamètres des zones d'inhibition de *B. vulgaris* étaient de 18,50 mm (conc R), 17,00 mm (conc D1) et 10,00 mm (conc D2) contre *S. dysenteriae*. La CMI et la MBC étaient similaires pour les deux organismes, avec *H. sabdariffa* (CMI: D3-4,27 mg/mL, MBC: D1-68,25 mg/mL) et *H. contortus* (CMI: D3-4,69 mg/mL, MBC: R -75,00 mg/mL), tandis que *M. oleifera*, *C. papaya* et *B. vulgaris* avaient une activité antimicrobienne négligeable.

**Conclusion:** *Hibiscus sabdariffa* et *H. contortus* ont présenté de puissants effets antimicrobiens contre *Salmonella*, avec des CMI de 4,27 mg/mL et 4,69 mg/mL et des MBC de 68,25 mg/mL et 75,00 mg/mL respectivement. Leurs CMI constamment faibles contre *Shigella* suggèrent leur potentiel de production d'antibiotiques.

**Mots-clés:** Agent antimicrobien; Résistance aux antibiotiques; Extraits de plantes; CMI; MBC

## Introduction:

Throughout history, certain plants have played significant roles in traditional medicine, serving as remedies for various ailments and contributing to preservation methods. Long before discovering microorganisms, the belief in the healing potential of certain plants was widely accepted. Human utilization of plants for medicinal purposes dates back millennia, reflecting the enduring relationship between humanity and therapeutic properties of plants. The extensive array of plant species employed for healing purposes encompasses a wide spectrum, including analgesics, anticancer, antipyretic and antihypertensive agents (1).

Earth is home to approximately 250,000 higher plant species, with over 80,000 reported to possess medicinal value and around 5,000 recognized for specific therapeutic benefits. Notably, more than 80% of the global population relies primarily on plants and plant extracts for healthcare (2,3). There is a growing interest in plant-derived drugs and dietary supplements, particularly in the treatment of infectious diseases. Traditional healers, inspired by the historical use of plants, have contributed to the integration of plant-based remedies into Western medicine. The secondary metabolites found in plants, such as tannins, terpenoids, alkaloids, and flavonoids, are well-known for their in vitro antimicrobial properties. Plant-based antimicrobials may offer alternative mechanisms for inhibiting bacteria, potentially contributing to the treatment of resistant microbial pathogens (4).

In response to the escalating issue of

antibiotic resistance, numerous studies have explored plants as natural antimicrobials (5). The rise in resistance to conventional antimicrobial agents, particularly antibiotics, has led to alarming mortality rates, with an estimated 100,000 deaths annually and a projected global increase to 10 million by 2050, with Africa accounting for 40% of these deaths (6, 7,8).

In response, the World Health Organization (WHO) published a list of antibiotic-resistant bacteria of public health concerns, signaling the urgent need for new measures (9). Investigating the phytochemical constituents and antimicrobial properties of herbal plants holds promise for developing new drugs to combat antibiotic resistance, with the added benefit of lower toxicity compared to chemically synthesized antimicrobials.

This study aims to assess the antimicrobial properties of five medicinal plants; *Bambusa vulgaris* (Bamboo), *Heteropogon contortus* (Spear grass), *Hibiscus sabdariffa*, *Moringa oleifera*, and *Carica papaya*, traditionally used for medicinal purposes in treating infections caused by *Salmonella* Typhi and *Shigella dysenteriae*.

## Materials and method:

### Plant collection and identification:

Plants were gathered from Gateri village of Kurmi Local Government Area (LGA) and Kona area of Jalingo LGA Taraba State, Nigeria, in August 2022. The Department of Botany, Taraba State University, Jalingo, identified the plants. The selection was based on

traditional and ethnobotanical usage indicating potential antimicrobial properties.

#### Plant preparation:

The plant parts to be used were air dried under shade, cut into pieces, and stored at 40°C. 300g of the plant part(s) were extracted with solvents of 80% 900ml each of ethanol, ethyl acetate, dichloromethane, and hexane in a shaking incubator at 28°C for 12 hours. The residues were re-extracted three times. The extracts were pooled and filtered, and the solvent-combined specimen was evaporated to dryness using a vacuum rotary evaporator and weighed to determine the yield of a soluble constituent, labeled and stored in a desiccator, subject to further analysis.

To vary the concentration, extract(s) were dissolved in phosphate-buffered saline (PBS) at a concentration of 100mg/ml for each extract. Five-fold serial dilution were carried out in 10ml sterile test tubes containing PBS. This was used in the preparation of the antibiotic plates. PBS has no antimicrobial properties.

#### Isolation and identification of test clinical bacteria isolates:

*Salmonella* Typhi and *S. dysenteriae* used were isolated from patients attending the Specialist Hospital, Jalingo Taraba State. Stool samples were collected into a sterile container and processed immediately. The stool samples were cultured on *Salmonella-Shigella* (SS) agar and incubated at 37°C (98.6°F) for 24 hours. *Salmonella* colonies on SS agar appeared colorless with a black center due to hydrogen sulfide production, while *Shigella* colonies

appeared colorless because of their inability to produce hydrogen peroxide.

To confirm the identity of the isolates, biochemical tests such as Triple Sugar Iron (TSI) agar test was used to differentiate based on sugar fermentation and gas production, Urea agar test was used to test for urease production, and motility test with *Salmonella* motile, while *Shigella* is non-motile.

#### Determination of antimicrobial concentrations of the extracts by serial dilution:

The concentrations (mg/mL) of extracts from *H. sabdariffa*, *M. oleifera*, *C. papaya*, *B. vulgaris*, and *H. contortus* are presented following various dilution levels as shown in Table 1. For *H. sabdariffa*, the undiluted (raw) concentration (R conc) was 273 mg/mL, decreasing to 0.027 mg/mL at the fifth dilution (D5 conc). The concentration of the positive control (ceftriaxone) was a constant of 0.030 mg/mL, while the concentration of the negative control (Dilution Solvent: ethanol) remained 0.00 mg/mL. Other plant extracts had similar concentration trends at different levels of dilution, giving varying concentrations for each extract.

The initial concentrations before dilution (R conc.) for *H. sabdariffa*, *M. oleifera*, *C. papaya*, *B. vulgaris*, and *H. contortus* were 273 mg/mL, 82 mg/mL, 111 mg/mL, 266 mg/mL, and 75 mg/mL, respectively. After 5-fold serial dilutions (D1 to D5 conc), concentrations varied as shown. Ceftriaxone, the positive control, maintained a constant concentration of 0.030 mg/mL. *Hibiscus sabdariffa* had the highest concentrations, followed by *B. vulgaris*, *C. papaya*, *M. oleifera*, and *H. contortus*.

Table 1: Concentrations of the medicinal plant extracts from serial dilution and the controls

Plants	Concentrations in milligrams per milliliter (mg/mL)							Cef (PC)	DS (NC)
	R conc	D1 conc	D2 conc	D3 conc	D4 conc	D5 conc			
<i>Hibiscus sabdariffa</i>	273	68.25	17.06	4.27	1.066	0.027	0.030	0.00	
<i>Moringa olifera</i>	82	20.5	5.13	1.28	0.32	0.08	0.030	0.00	
<i>Carica papaya</i>	111	27.75	6.93	1.73	0.44	0.11	0.030	0.00	
<i>Bambusa vulgaris</i>	266	66.5	16.63	4.156	1.04	0.26	0.030	0.00	
<i>Heteropogon contortus</i>	75	18.75	4.69	1.17	0.30	0.075	0.030	0.00	

R con= Undiluted (Raw) concentration, D1 conc = First dilution concentration, D2 conc = Second dilution concentration, D3 conc = Third dilution concentration, D4 conc = Fourth dilution concentration, D5 conc = Fifth dilution concentration, Cef = Ceftriaxone, PC = Positive Control, DS = Dilution Solvent (ethanol), NC = Negative Control

**Analysis of phytochemical constituents of the medicinal plants:**

The secondary metabolites were identified using previously described standard methods. Cardiac glycosides were identified by the Kellar-Kiliani test, which involves dissolving 50mg of methanolic extract in 2ml chloroform, and adding H<sub>2</sub>SO<sub>4</sub> to produce brown ring at the interface. Flavonoid was detected by the Shinoda test which involves adding to 2-3ml of ethanolic extract, Mg ribbon and HCl to give a pink-red or red solution or by the NaOH test in which extract is treated with dilute NaOH, and then dilute HCl to give a yellow solution that turns colorless. Phenol was identified by the phenol test in which to a spot extract on filter paper is added phosphomolybdic acid reagent, which is then exposed to ammonia vapors to give blue spot coloration.

Phlobatannin was identified by boiling 2ml of the extract with 2ml of 1% HCl leading to the formation of red precipitates. Saponin was identified by the Frothing or Foam test in which 0.5ml filtrate was added to 5ml distilled water and shaken to give persistent frothing. Steroid was detected by the Liebermann-Burchardt test which involves mixing 1ml of the extract with chloroform, acetic anhydride, and sulfuric acid to give dark green coloration, which can be confirmed by mixing 1ml extract with acetic anhydride and sulfuric acid to produce blue or green color change.

Tannin was detected by the Braemer's test which involved mixing ferric chloride with extract to produce dark blue or greenish-grey solution. Terpenoid was also detected by the Liebermann-Burchardt test that involved mixing extract with chloroform, acetic anhydride, and sulfuric acid to produce pink or red coloration or by the Salkowski test in which extract is combined with chloroform and sulfuric acid to produce reddish-brown color interface.

**Test of antimicrobial properties by the disk diffusion method:**

The Kirby-Bauer disk diffusion method was used to assess the antimicrobial properties of the test extracts. First, the test organisms were inoculated on nutrient agar plates and impregnated sterile filter paper disks with plant extract solution at different concentrations (D1, D2, D3, D4, D4, and R conc.) were placed on inoculated agar using sterile forceps, evenly distributed across plates. The plates were incubated at 37°C for 24 hours, followed by measurement of diameters of zones of inhibition using a ruler and determination of antimicrobial activity. The control disks were prepared with PBS (negative control) and commercially prepared ceftriaxone

(Hardy diagnostics, Santa Maria, USA) as positive control. Inhibition zone diameter of 24–30 mm indicated sensitivity, 13–23 mm intermediate and ≤12 mm resistance (10).

**Determination of minimum inhibitory concentration (MIC) and minimum bacterial concentration (MBC) of the extracts:**

The MIC of the extracts against the test bacterial isolates (*S. Typhi* and *S. dysenteriae*) was determined by diluting an overnight culture growth of a pure culture of the test organisms in nutrient broth to a concentration of 1×10<sup>5</sup> CFU/ml. A 5-fold serial dilution of the antimicrobial test extract was performed in a series of test tubes to establish varying concentrations using the ethanolic extract. One milliliter of each test extract dilution was inoculated with an equal volume of the standardized microorganism. Positive control (ceftriaxone) and negative control (PBS) tubes were included for each test microorganism. The tubes were incubated at 37°C for 24 hrs. The MIC was determined as the lowest concentration of the extract in the tube where no growth was visually observed.

For the MBC determination, dilutions representing the MIC and concentrations above were plated on nutrient agar, incubated overnight, and colonies were enumerated to determine CFU/ml. The MBC is the lowest concentration showing no viable bacterial growth.

**Results:****Phytochemical constituents of extracts:**

Table 2 indicates the results for the different components (saponins, cardiac glycosides, terpenoids, steroids, flavonoids, phenolics, and tannins) with the four solvents used for the plant extracts (ethanol, ethyl acetate, dichloromethane, hexane). Saponins were extracted by ethanol, ethyl acetate, and hexane. Cardiac glycosides by ethanol, ethyl acetate and hexane. Terpenoids by ethyl acetate and hexane with highest concentration with hexane. Steroids by ethyl acetate, dichloromethane, and hexane with highest concentration in hexane. Flavonoids by ethanol, ethyl acetate, and hexane. Phenolics by ethyl acetate, dichloromethane, and hexane with highest concentration in dichloromethane. Tannins by ethanol, ethyl acetate, and hexane, with the highest concentration in ethanol.

Ethanol had higher yield of the phytochemical components compared to the other solvents. Additionally, ethanol and ethyl acetate also showed the presence of more phytochemical components than other solvents.

Table 2: Phytochemical components of the plant extracts by the four extraction solvents

Phytochemical components	Ethanol					Ethyl Acetate					Dichloromethane					Hexane				
	Hs	Mo	Cp	Bv	Hc	Hs	Mo	Cp	Bv	Hc	Hs	Mo	Cp	Bv	Hc	Hs	Mo	Cp	Bv	Hc
Saponins	+	+	-	+	+	+	-	+	-	-	+	-	+	+	+	-	+	+	+	+
Cardiac glycosides	+	+	-	+	+	+	+	+	+	-	+	-	-	-	+	+	+	-	-	+
Terpenoids	-	+	+	-	+	-	+	-	-	+	+	-	-	-	+	+	+	+	-	-
Steroids	-	+	+	-	+	+	-	-	+	+	+	+	-	-	+	+	+	+	+	+
Flavonoids	+	+	+	+	+	-	+	+	-	+	+	+	+	+	+	-	+	+	-	+
Phenolics	-	-	+	-	+	+	+	+	-	+	-	+	+	+	-	+	-	-	-	+
Tannins	+	+	+	+	+	-	+	+	-	+	+	+	+	-	-	+	-	-	+	-

+ = Positive, - = Negative, Hs = *Hibiscus sabdariffa*, Mo = *Moringa oleifera*, Cp = *Carica papaya*, Bv = *Bambusa vulgaris*, Hc = *Heteropogon contortus*

Table 3: Diameter of zone of inhibition of bacterial isolates at various concentrations of the extracts and the controls

Extracts/concentrations	R conc	D1 conc	D2 conc	D3 conc	D4 conc	D5 conc	Cef (PC)	PBS (NC)
Diameter of zone of inhibition (mm) for <i>Salmonella</i> Typhi								
<i>Hibiscus sabdariffa</i>	29.00	27.00	14.00	4.00	0.00	0.00	14.05	0.00
<i>Moringa oleifera</i>	1.00	0.50	0.00	0.00	0.00	0.00	13.80	0.00
<i>Carica papaya</i>	1.00	0.00	0.00	0.00	0.00	0.00	14.50	0.00
<i>Bambusa vulgaris</i>	5.00	5.00	3.00	0.00	0.00	0.00	15.00	0.00
<i>Heteropogon contortus</i>	10.00	4.00	2.00	1.00	0.50	0.00	14.00	0.00
Diameter of zone of inhibition (mm) for <i>Shigella dysenteriae</i>								
<i>Hibiscus sabdariffa</i>	22.05	16.00	5.50	0.50	0.00	0.00	15.00	0.00
<i>Moringa oleifera</i>	1.00	0.00	0.00	0.00	0.00	0.00	14.00	0.00
<i>Carica papaya</i>	1.00	0.7	0.00	0.00	0.00	0.00	14.00	0.00
<i>Bambusa vulgaris</i>	18.50	17.00	10.00	5.00	0.00	0.00	13.50	0.00
<i>Heteropogon contortus</i>	25.05	15.00	10.00	5.00	0.00	0.00	14.00	0.00

R conc=Raw concentration, D1 conc = First dilution concentration, D2 conc = Second dilution concentration, D3 conc = Third dilution concentration, D4 conc = Fourth dilution concentration, D5 conc = Fifth dilution concentration, ZI = Zone of inhibition, Cef = Ceftriaxone, PC = Positive Control, DS = Dilution Solvent (ethanol), NC = Negative Control

### Antimicrobial properties of the extracts

Table 3 displays zone of inhibition (ZI) values for *S. Typhi* and *S. dysenteriae* in response to varying concentrations of the plant extracts. Undiluted (273 mg/mL) and D1 concentration (68.2 mg/mL) of *H. sabdariffa* extracts had antibacterial effects on *S. Typhi* and *S. dysenteriae* above the diameter of zone of inhibition breakpoint of  $\leq 12$  mm, comparable to 0.030 mg/mL of ceftriaxone control. Also, undiluted (266 mg/mL) and D1 concentration (66.5 mg/mL) of *B. vulgaris* extracts had antibacterial effects on *S. dysenteriae* above the diameter of zone of inhibition breakpoint of  $\leq 12$  mm, comparable to 0.030 mg/mL ceftriaxone control. Similarly, undiluted (75 mg/mL) and D1 concentration (18.75 mg/mL) of *H. contortus* extracts had antibacterial effects on *S. dysenteriae* above the diameter of zone of

inhibition breakpoint of  $\leq 12$  mm, comparable to 0.030 mg/mL ceftriaxone control.

In *S. Typhi*, the zone of inhibition (ZI) of *H. sabdariffa* decreases from 29.00 mm at undiluted concentration to 0.00 mm at the highest dilution (D5). Positive control (ceftriaxone) had a ZI of 14.05 mm, while negative control (NC) was 0.00 mm. Similar concentration-dependent decreases were observed for other plants, indicating reduced inhibitory effects with dilution. For *Shigella dysenteriae*, a similar concentration-dependent decrease in ZI values was noted. For instance, in *H. sabdariffa*, the ZI decreased from 22.05 mm at undiluted concentration to 0.00 mm at D5. The positive control maintained a constant 15.00 mm ZI, while the negative control remained at 0.00 mm.

Table 4: Minimum inhibitory and minimum bactericidal concentration of extracts against the test bacterial isolates

Plant extracts	MIC (mg/mL)	MBC (mg/mL)
<i>Hibiscus sabdariffa</i>	D3 (4.27)	D1 (68.25)
<i>Moringa oleifera</i>	R (82.00)	-
<i>Carica papaya</i>	D2 (6.93)	-
<i>Bambusa vulgaris</i>	D2 (16.63)	-
<i>Heteropogon contortus</i>	D3 (4.69)	R (75.00)
	<i>Shigella dysenteriae</i>	
<i>Hibiscus sabdariffa</i>	D3 (4.27)	D1 (68.25)
<i>Moringa oleifera</i>	R (82.00)	-
<i>Carica papaya</i>	D2 (6.93)	-
<i>Bambusa vulgaris</i>	D2 (16.63)	D1 (66.5)
<i>Heteropogon contortus</i>	D2 (4.69)	D1 (18.75)

MIC= Minimum inhibitory Concentration, MBC= Minimum Bactericidal Concentration, R conc= Raw concentration, D1 conc = First dilution concentration, D2 conc = Second dilution concentration, D3 conc = Third dilution concentration

As the test extracts were diluted, the zone of inhibition against both bacteria decreased, suggesting diminishing antimicrobial effects. This dose-dependent response indicates antimicrobial activity with variations among plants and dilutions. The positive control consistently showed approximately 14 mm ZI for both bacteria, highlighting antibacterial efficacy of ceftriaxone, while the negative control consistently exhibited no ZI, indicating the solvent's lack of antibacterial properties.

#### Minimum inhibitory and bactericidal concentrations of the extracts:

Table 4 represents the MIC and MBC values of the plant extracts for *S. Typhi* and *S. dysenteriae*. For *S. Typhi*, the MIC of *H. sabdariffa* was at D3 conc (4.27 mg/mL) and MBC at D1 conc (68.25 mg/mL). The MIC of *M. oleifera* was at R conc (82.00 mg/mL), with no MBC reached. The MIC of *C. papaya* was at D2 conc (6.93 mg/mL), and *B. vulgaris* at D2 conc (16.63 mg/mL), both without MBC. The MIC of *H. contortus* was at D3 conc (4.69 mg/mL) and MBC at R conc (75 mg/mL).

For *S. dysenteriae*, the MIC of *H. sabdariffa* was at D3 conc (4.27 mg/mL) and MBC at D1 conc (68.25 mg/mL). The MIC of *M. oleifera* was at R conc (82.00 mg/mL), with no MBC reached. The MIC of *C. papaya* was at D2 conc (6.93 mg/mL), and *B. vulgaris* at D2 conc (16.63 mg/mL) and MBC at D1 conc (66.5 mg/mL). The MIC of *H. contortus* was at D2 conc (4.69 mg/mL) and MBC at D1 conc (18.75 mg/mL).

#### Discussion:

This study was a thorough investigation into the phytochemical composition, con-

centrations, and antimicrobial efficacy of extracts from *H. sabdariffa*, *M. oleifera*, *C. papaya*, *B. vulgaris*, and *H. contortus*. Employing various solvents, including ethanol, ethyl acetate, dichloromethane, and hexane, the research explored the diverse phytochemical components using 5-fold serial dilutions, gauging antimicrobial effects against *S. Typhi* and *S. dysenteriae*. Phytochemical analysis revealed the presence of saponins, cardiac glycosides, terpenoids, steroids, flavonoids, phenolics, and tannins, with ethanol consistently exhibiting higher concentrations. The solvent ethanol generally has higher concentrations of these phytochemicals than other solvents. These findings align with prior research (11).

Antimicrobial Activities were measured by the zone of inhibition (ZI) against *S. Typhi* and *S. dysenteriae*, the activity decreases with increasing dilutions, indicating a dose-dependent response and antimicrobial activity. Ceftriaxone served as a positive control, consistently demonstrating effectiveness. Dose-dependent responses and antimicrobial activities from plant extracts were also corroborated by existing studies (12-15).

Specifically, Perera et al., (11) highlighted the antibacterial potential of *H. contortus* extracts against *Escherichia coli* and *Staphylococcus aureus*. Eremwanarue and Shittu (16) demonstrated antibacterial activity of *M. oleifera* against multidrug-resistant bacteria. Abdallah (17) reported the efficacy of *H. sabdariffa* calyces against various bacteria, surpassing penicillin. Emad (18) reported significant antibacterial properties of *H. sabdariffa* against *A. baumannii* strains. Bokaeian et al., (19) explored inhibitory effects of *M. oleifera* against *E. coli* and *S. aureus*, exhibiting dose-

dependent responses. Timothy et al., (20) reported antibacterial activity of *H. sabdariffa* against *S. aureus* and *E. coli*. Guteirrez et al., (21) studied the effective inhibition of *H. sabdariffa* calyx extracts against multidrug-resistant *Salmonella* strains.

The MIC and MBC values determined revealed similar MIC values for both bacterial isolates with varying MBC values. *Salmonella Typhi* exhibited greater resistance than *S. dysenteriae*. Individual plants exhibited unique MIC and MBC profiles, highlighting plant-specific antibacterial properties. For instance, *H. sabdariffa* at D3 conc demonstrated an MIC of 4.27 mg/mL against both bacterial strains, while *B. vulgaris* at D2 conc exhibited an MBC of 16.63 mg/mL against *S. Typhi* and 66.5 mg/mL against *S. dysenteriae*.

### Conclusion:

This study provides valuable insights into the phytochemical composition and antimicrobial properties of plant extracts, emphasizing the importance of plant specificity in antibacterial effects. The results have implications for potential therapeutic applications and warrant further investigation into the active compounds responsible for the observed effects. The variations in phytochemical content and antimicrobial efficacy among different solvents and plant species underscore the importance of careful selection in extraction processes for medicinal purposes.

Our findings support the idea that plant extracts could serve as potential sources of antimicrobial agents. However, further research, including toxicity studies and clinical trials, is essential to validate their safety and efficacy for practical applications in medicine. Further studies are also required to determine the bioactive component and antimicrobial mechanisms of actions of the extracts for proper documentation. The study contributes valuable insights into the potential use of these plants in developing natural antimicrobial agents for combating bacterial infections.

### Contributions of authors:

DZ designed the study, coordinated the research, monitored the benchwork and wrote the manuscript. JB performed the phytochemical analysis, while HN, EO, AG and HC carried out the bench work. All authors read and approved the manuscript for submission.

### Source of funding:

This research was sponsored by the Tertiary Education Trust Fund (TETFUND) through the Institutional-Based Research (IBR) fund allocated to Taraba State University Jalingo, Nigeria.

### Conflicts of interest:

No conflict of interest is declared.

### References:

- Rios, J. L., and Recio, M. C. Medicinal plants and antimicrobial activity. *J Ethnopharmacol.* 2005; 100: 80–84. <https://doi.org/10.33545/27072827.2020.v1.i2a.14>
- Vyshnavi, N. Commentary: Importance of Medicinal Plants in Medicine. *J Med Org Chem.* 2021; 1 (1):
- Okoye, T. C., Uzor, P. F., Onyeto, C. A., and Okereke, E. K. Safe African Medicinal Plants for Clinical Studies. *Toxicological Survey of African Medicinal Plants*, 2014; 535–555. [doi:10.1016/B978-0-12-800018-2.00018-2](https://doi.org/10.1016/B978-0-12-800018-2.00018-2)
- Cowan, M. M. Plant products as antimicrobial agents. *Clin Microbiol Rev.* 1999; 12 (4): 564–582. <https://doi.org/10.1128/cmr.12.4.564>
- Zenoh, D. A., Asuquo, A.B., Eyo, A. O., and Yakubu, N. Antibiogram of Bacteria Associated with Urinary Tract Infection in People Living with HIV/AIDS in Jalingo, Nigeria. *Calabar J Health Sci.* 2018; 2 (2):
- Wenner, M. M. Dangerous New Antibiotic-Resistant Bacteria Reach U.S. America Scientific research article. 2016 <https://www.scientificamerican.com/article/dangerous-new-antibiotic-resistant-bacteria-reach-u-s/>
- Oluwadara, A. What Nigeria can do about antimicrobial Resistance. Article on Antimicrobial Resistance. <http://www.businessdayonline.com/nigeria-can-antimicrobial-resistance>
- Medical World Nigeria: Nigeria Center for Disease Control Antimicrobial Resistance (AMR) Symposium: Call for Abstracts 2017. [www.medicalworldnigeria.com](http://www.medicalworldnigeria.com).
- World Health Organization: Who publishes list of bacteria for which new antibiotics are urgently needed. *Saudi Med J* 2017; 38: 444–445. <https://www.who.int/news/item/27-02-2017-who-publishes-list-of-bacteria-for-which-new-antibiotics-are-urgently-needed>.
- Hudzicki, J. Kirby-Bauer disk diffusion susceptibility test protocol. <https://asm.org/getattachment/2594ce26-bd44-47f6-8287-0657aa9185ad/Kirby-Bauer-Disk-Diffusion-Susceptibility-Test-Protocol-pdf.pdf>
- Perera, H. E. H., Maddumage, R. S., Fernando, R. S. P., Ratnasooriya, W. D., Pathirana, R. N., and Silva, A. R. N. In vitro antibacterial activity of aqueous extracts of the whole plant, flowers, roots, leaves, and stem of grass *Heteropogon contortus* grown in Sri Lanka. *Int J Pharmacogn Life Sci.* 2020; 1 (2): 24–28.
- Yakobi, S. H., Mkhize, S., and Poee, O. J. Screening of antimicrobial properties and bioactive compounds of *Pleurotus ostreatus* extracts against *Staphylococcus aureus*, *Escherichia coli*, and *Neisseria gonorrhoeae*. *Biochem Res Int.* 2023; 2023: 1777039 <https://doi.org/10.1155/2023/1777039>
- Jahan, S., Shahjahan, M., Rasna, S. S., et al. Antibacterial Effect of Moringa (*Moringa oleifera*) Leaf Ethanolic Extract Against *Staphylococcus aureus* and *Escherichia coli*. *Mymensingh Med J.* 2022; 31 (4): 976–982
- Manandhar, S., Luitel, S., and Dahal, R. K. In vitro antimicrobial activity of some medicinal plants against human pathogenic bacteria. *J Trop Med.* 2019; 2019: 1895340 <https://doi.org/10.1155/2019/1895340>
- Fullerton, M., Khatiwada, J., Johnson, J. U., Davis, S., and Williams, L. L. Determination of the antimicrobial activity of sorrel (*Hibiscus sabdariffa*) on *Escherichia coli* O157:H7 isolated from food, veterinary, and clinical samples. *J Med Food.* 2011; 14 (9): 950–956. <https://doi.org/10.1089/imf.2010.0200>

16. Eremwanarue, O. A., and Shittu, H. O. Antimicrobial activity of *Moringa oleifera* leaf extracts on multiple drug-resistant bacterial isolates from urine samples in Benin City. *Nig J Biotechnol.* 2019; 35 (2): 16.  
<https://doi.org/10.4314/njb.v35i2.3>
17. Abdallah, E. Antibacterial efficiency of the Sudanese Roselle (*Hibiscus sabdariffa* L.), a famous beverage from Sudanese folk medicine. *J Inter-cultural Ethnopharmacol.* 2016;5(2):186.  
<https://doi.org/10.5455/iice.20160320022623>
18. Emad, M. A. Antibacterial activity of *Hibiscus sabdariffa* L. calyces against hospital isolates of multidrug-resistant *Acinetobacter baumannii*. *J Acute Dis.* 2016; 5 (6): 512-516.
19. Bokaeian, M., Sheikh, M., Shahi, Z., and Saeidi, S. Antimicrobial activity of *Hibiscus sabdariffa* extract against human pathogen. *Int J Adv Biol Biomed Res.* 2014; 2 (2): 433-439
20. Timothy, S., Anaegbu, J., Yakubu, N., et al. Phytochemical and antimicrobial activity of aqueous extract of *Hibiscus sabdariffa*. *J Pharm Bio-resources.* 2008; 5 (1): 24-30.  
<https://doi.org/10.4314/jpb.v5i1.52989>
21. Gutiérrez-Alcántara, E. J., Gómez-Aldapa, C. A., Román-Gutiérrez, A. D., Rangel-Vargas, E., González-Olivares, L. G., and Castro-Rosas, J. Antimicrobial activity of Roselle *Hibiscus sabdariffa* calyx extracts on culture media and carrots against multidrug resistant *Salmonella* strains isolated from raw carrots. *J Food Safety.* 2016; 36 (4): 450-458.  
<https://doi.org/10.1111/jfs.12259>