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Antibacterial Activities of some Medicinal Plants Used for Treatment of Infectious Diseases in the Vina and Mayo-Louti Divisions of Cameroon

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Authors' contributions

This work was carried out in collaboration between all authors. Authors BTT and GZ designed the study, performed the statistical analysis, wrote the protocol and first draft of the manuscript. Authors SPCF, MTF and JRK managed the analyses of the study. Author LNT managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Background: In Cameroon, most peoples use traditional medicine treating infectious diseases. To verify the scientific bases of these locally used medicinal plants, an ethnobotanical survey was carried out in some villages of Vina and Mayo-Louti Divisions.

Materials and Methods: Interviews were conducted through structured questionnaires among 31 traditional healers living in these divisions. With the medicinal plants revealed, a literature investigation on their therapeutic effects, as well as *in vitro* antimicrobial activity of these plants

were conducted. The agar diffusion method was used to determine the antibacterial activities of the methanol extracts against the pathogens while the Minimum Inhibitory Concentration (MIC) was determined using the Broth dilution method.

Result: A total of 15 medicinal plants species belonging to 12 families are being used in the treatment of numerous infectious diseases in the Vina and Mayo-Louti Divisions. *Khaya senegalensis* (Meliaceae), *Terminalia glaucescens* (Combretaceae), *Flacourtia flavescens* wild (Salicaceae), *Pterocarpus erinaceus* (Fabaceae) and *Boswellia dalzielii* (Burseraceae) were the mostly used plants for the treatment of infectious diseases in the study areas. Maceration (43.75%) was the common mode of preparation, followed by infusion (31.25%) and decoction (25.00%). Bioassay showed that crude methanol extract of *Pterocarpus erinaceus* and *Flacourtia flavescens* were the most active plant extract with a MIC of 0.8 mg/ml on many tested bacteria. The antibacterial activity of *Boswellia dalzeilii* from Cameroon are reported here for the first time. **Conclusion:** Many herbals remedies are used in these divisions for the treatment infectious diseases. The plants can be used as source of antibacterial drugs to treat infections caused by susceptible bacteria

Keywords: Antibacterial activities; medicinal plants; infectious diseases; Cameroon.

1. INTRODUCTION

Infectious diseases are one of the leading causes of morbidity and mortality worldwide, especially in developing countries [1]. Indeed, human commensal bacteria could become pathogenic either due to a change in their normal behaviour /habitat or a failure in the immune system [2]. Furthermore, enteric fever caused by *Salmonella enterica* serotypes Typhi and Paratyphi A, B, and C is mainly a disease in developing countries, and it is occasionally diagnosed as an imported disease in countries where the disease is not endemic [3].

The discovery of antibiotics has decreased the spread and severity of a wide variety of infectious diseases. However, as a result of their uncontrolled use, the efficiency of many antibiotics is being threatened by the emergence of microbial resistance to existing chemotherapeutic agents [4] such as direct destruction of antibiotics by penicillinase producing bacteria, or resistance to the wide range use of Amphotericin B and azole derivatives by pathogenic fungi [5].

While bioactive natural compounds have been isolated mainly from cultivable microbial strains, unexploited biologically active metabolites of different sources including plants remain to be investigated [6] to alleviate or help respond to current health care situations.

Plant-derived natural products, therefore, represent an attractive source of antimicrobial agents since they are natural, have manageable

side effects and available at affordable prices [7]. Also, plants derived agents may have different mechanisms of action than conventional drugs [8].

The diversity of Cameroon flora has a dominance of plants that have been used so fare for many pharmacological purposes. Although traditional medicine has played and continue to play a critical role worldwide in treating infectious diseases [9]; no scientific work has been carried out to the best of our knowledge, in many parts of the country to index medicinal plants used in these localities for the treatment of infectious diseases. Thus the present proposal, which aims at carrying out an activities of medicinal plants used for the treatment of infectious diseases in some localities of Vina and Mayo-Louti Division (Northern Region of Cameroon).

2. MATERIALS AND METHODS

2.1 Ethnobotanical Survey and Plant Collection

This study took place in Northern Cameroon, and more specifically in the Vina Division (6° 37' 60" latitude North and 13° 24' 0" longitude East) and the Mayo-Louti Division (9° 37' 60" latitude North and 13° 55' 60" longitude East). To identify some plants used for the treatment of infectious diseases such as malaria, typhoid, diarrhoea and dysentery in these regions, an ethnobotanical survey was carried out from April 2013 to November 2013 involving thirty-one traditional healers aged between 20 and 50 years. A questionnaire was used for the survey and it comprised: types of medication (medicinal plants or pharmaceutical products) used when sick; and for each medicinal plant used, its vernacular name, parts of the plant used, methods of preparation and administration, diseases used for, the solvent used.

The interviews were conducted in the local language (foufouldé) to facilitate communication. Fresh samples of plants were harvested, and their identification confirmed at the Cameroon National Herbarium (CNH), where their full scientific names and voucher number were obtained. Further literature investigations were also conducted relative to their therapeutic and pharmacological effects and their phytochemical composition. The plants of interest in this study should be traditionally used for the treatment of infectious diseases in the Department of Vina and the Department of Mayo-louti and should have been cited by at least five traditional healers in both divisions. After the ethnobotanical survey, (Meliaceae), Khava senegalensis glaucescens (Combretaceae), Terminalia wild Flacourtia flavescens (Salicaceae), Pterocarpus erinaceus (Fabaceae) and Boswellia dalzielii (Burseraceae) were selected for antimicrobial analyses.

2.2 Preparation of Extracts

The bark and roots of four of the above plants were harvested in the Vina Division while roots and bark of Boswellia dalzielii were harvested in Mayo-Louti Division, shade-dried, the and ground. 500 grams of each powder was macerated in 2000 mL of methanol in an Erlenmever flask. The mixture was filtered, and the residue returned to the methanol after every 24 hours. The operation was repeated twice. At the end of the 72 hours, the filtrate was concentrated using a Rotary evaporator.at 45°C. The resulting extract was weight, and the extraction yield was 7.36, 11.28, 8.4, 5.5, 9.82, 3.00 and 4.75 for Terminalia glauscescens stem, glauscescens Terminalia root. Khava senegalensis, Pterocarpus erinaceus, Boswellia dalzielii root, Boswellia dalzielii stem and Flacourtia flavescens respectively. These extracts were then stored in sterile, clean and dry flasks before usage

2.3 Microorganisms

Microorganisms were obtained from the Laboratory of Microbiology and Antimicrobial Substances of the University of Dschang. All the

ten microorganisms investigated, were associated with numerous cases of human infections. The bacteria (clinical isolates) used in this study were collected from Centre Pasteur (Yaoundé-Cameroon), and constitute of two Gram-positive bacteria (Bacillus cereus and Staphylococcus aureus), and eight Gramnegative bacteria (Escherichia coli, Klebsiella pneumonia, Proteus mirabilis, Pseudomonas aeruginosa, Salmonella typhi, Salmonella typhimurium, Salmonella paratyphi A, Salmonella paratyphi B). All the microorganisms were maintained at 4°C on Muller Hinton Agar.

2.4 Phytochemical Screening of Plants Extracts

Phytochemical screening aims to estimate the nature of secondary metabolites responsible for the biological activity of plant extracts. Phytochemical Screening was carried out, according to the methods described by Prashant et al. [10] and Shaheen et al. [11].

2.5 Antibacterial Test

Microbiological screening of the crude extracts was evaluated by the agar diffusion method (agar well diffusion method) as described by Abubakar[12] and Titilope et al. [13]. The Petri dish was incubated overnight at 37°C, and microbial growth was determined by measuring the inhibition zone diameter. The extracts were considered active when the inhibition zone diameter was greater than or equal to 14 mm. For each bacterial strain, controls were done, and pure solvents were used instead of the extract. The experiment was done three times, and the mean values are presented.

2.6 Determination of Minimal Inhibitory Concentration (MIC) and Minimal Bactericidal Concentration (CMB)

The MIC and CMB were determined by the method of micro dilution in a liquid medium [9]. In the 96-well microplates, 100 μ L of double concentrated culture broth (MHB) was added to each well. These wells were then supplemented with the various extracts (100 μ L) and reference antibiotic (ceftriaxone) concentrations ranging from 12000 to 93.75 μ g / mL and from 50 to 0.39 μ g / mL respectively. Finally, ten μ L of bacteria inoculum 1.5 × 10⁶ CFU/mL were introduced into each well. Control wells containing only the broth and those containing the inoculum without extract or antibiotics were made.

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After 24 hours of incubation at 37°C, 20 µL of 2% (Methyl thiazolyl-diphenyl tetrazolium MTT bromide) solution were added in one test wells of each repeated concentration. This compound is instantly metabolized (reduced) by the active living cell mitochondrial succinate dehydrogenase to formazan to form a purple precipitate whose intensity is proportional to the number of living cells and the metabolic activity of each cell. The lowest concentration at which no visible colour change was observed was considered as the MIC. For wells that did not receive a developer, 20 µl of a solution of the wells corresponding to the concentrations that did not display the blue-violet colour were removed and streaked on the surface of the MHA previously poured into Petri dishes. After 24 hours of incubation at 37°C, the concentrations of wells having less than three bacterial colonies were considered bactericidal and the smallest of these, noted as CMB.

2.7 Data Analyses

Descriptive statistic was principally used in this study. Initially, information on the popular uses of the species collected, along with botanical information, was compiled into a database. The species were listed in alphabetical order by scientific name, a popular name in the region (vernacular name), voucher number and frequency of use. The frequency of citation (FC) of the used plant species was evaluated using the following formula:

FC = Number of times a particular species was mentioned total number of times that all species were mentioned

Quantitative data were subjected to one-way analysis of variance, and differences between samples at P \leq 0.05 were determined by Duncan Multiple Range Test using the Statistical Package for the Social Sciences (SPSS) program. The experimental results were expressed (where appropriate) as the mean ± standard deviation of three replicates.

3. RESULTS

3.1 Ethno-Pharmacological Survey and Phytochemical Screening of Extracts

The set of plants recorded during the ethnobotanical survey and their frequency of citation are grouped in Table 1. A total of 15 plants belonging to 12 botanical families were

identified. The leaves were the most used parts followed by the bark. The 05 (five) most common plants (cited five times and more) were: *Boswellia dalzielii, Pterocarpus erinaceus, Khaya senegalensis, Flacourtia flavecens* and *Terminalia glaucescens*.

About three of the plants obtained is used to treat at least three others diseases. This is the case of senegalensis (Rheumatoid Khaya arthritis. Syphilis, Jatropha leprosy); curcas (Hypertension, rheumatism, diabetes); Euphobia hirta (Skin diseases, bronchitis and asthma). Three of these plants (Boswellia dalzielii, Flacourtia flavescens and Terminalia glaucescens) presented higher frequencies of citation.

3.2 Demography/Personal Information on Respondents

The characteristics of the population are presented in Figs. 1 to 4. From Fig. 1 it is noted that thirty-one (31) traditional healers were interviewed. The informants were between 20 and 50 years old (Fig. 3), and the modal class was 39 to 49 years old. The age distribution of informants showed that most of the traditional healer encountered during the survey are within the age range 29-49. Among these informants, there were 9 women and 22 men (Fig. 2).

3.3 Parts of the Plant Used, Mode of Preparation

Leaves, roots, stems, whole plant, fruits, seeds and barks were used for numerous preparations. Amount these, the most commonly used plant parts (Fig. 3). were the leaves (40.91%), followed by stems (31.81%), roots (18.18%), fruits (4.55%) and whole plant (4.55%). Water was the only solvent used for different preparations that included maceration (43.75%), infusion (25.25%) and decoction (25.00%) (Fig. 4).

After an investigation, five most used plants caught our attention. They were latter collected and used for biological tests.

3.4 Phytochemical Screening

Plants samples were screened for the following classes of compounds: polyphenols, tannins, terpenes steroids, flavonoids, alkaloids, anthraquinone and saponins. This test revealed the presence of different classes of chemical compounds in each of these extracts. (Table 2).

N°	Botanical/ Scientific name	family	Local name (fourfold)	Parts used	Methods of preparation	Frequency	Others use/effects
1	Khaya senegalensis	Meliaceae	Delhi	Barks, roots	Maceration	6/59	Rheumatoid arthritis, Syphilis, leprosy [14,15]
2	Pterocarpus erinaceus	Fabaceae	Banohi	barks	Maceration	7/59	Febrifuge [16]
3	Eucalyptus sp	Myrtaceae		Leaves	Decoction	3/59	Antidiabetic, anticancer [17,18]
4	Boswellia dalzielii	Burseraceae	Andakedje	Barks, roots	Maceration	8/59	Venereal diseases, rheumatism [19]
5	Flacourtia flavescens	Flacourtiaceae		Barks	Decoction	5/59	Jaundice, stomach pain [20]
6	Terminalia glaucescens	Combretaceae	Koulahi	Barks, roots	Maceration	5/59	Hemorrhoids [21]
7	Jatropha curcas	Euphorbiaceae	Kolkoladjé/Magalehi	Leaves	Infusion	2/59	Hypertension, rheumatism, diabetes [22]
8	Euphobia hirta	Euphorbiaceae	Kosam-yel	Whole plant	Maceration	3/59	Skin diseases, bronchitis and asthma [23]
9	Psorospermum febrifugum	Hypericaceae	Cawayki	Leaves, barks	Maceration /Infusion	4/59	Epilepsy disease [24]
10	Harungana madagascarensis	Hypericaceae	Bourgal	Leaves	Infusion	4/59	Anti-haemorrhage, skin diseases[25, 26]
11	Vitellaria paradoxa	Sapotaceae	Karehi	Leaves, barks	Infusion	3/59	Cutaneous infection, stomach ailments [27]
12	Aloes buettneri	Liliaceae		Leaves	Maceration	4/59	Dysmenorrhea, general stomach aches [28]
13	Citrus limonum	RutaceaeMyrtaceae	Lemou	Leaves, Fruit	Infusion	1/59	Arthritis [29]
14	Carica papaya	Caricaceae	Dukudjee	Leaves, roots	Decoction	3/59	Colon cancer, heart attacks [18]
15	Ocimum gratissimum	Lamiaceae	kacuke	Leaves	Decoction	1/59	respiratory tract infections, skin diseases,and conjunctivitis [30]

Table 1. Keck-list of medicinal plants species inventoried during the survey

Samples	Polyphenols	Tannins	Flavonoids	Sterols and tri terpenes	Glycosides	Alkaloids	Anthraquinones	Saponins
Bde	+	+	-	+	+	-	+	+
Bdr	+	+	+	+	+	+	+	-
Pe	+	+	+	+	+	+	+	-
Ff	+	+	+	+	-	-	+	-
Ks	-	-	+	+	-	+	-	-
Tge	+	+	-	+	-	+	-	+
Tgr	+	+	+	-	-	+	+	+

Table 2. Phytochemical profile of methanolic crude extracts

Key: + = Present; - = Absent; Tge: Terminalia glaucescens stem; Tgr: Terminalia glaucescens root; Ks: Khaya senegalensis; Pe : Pterocarpus erinaceus; Bde: Boswellia dalzielii root; Ff: Flacourtia flavescens

Extrait souche	Tge	Tgr	Ks	Pe	Bde	Bdr	Ff	Cef
Ec	17.5 ± 0.5 ^{de}	17.5 ± 0.5 ^{bc}	19.5 ± 0.5 ^e	16.5 ± 0.5 ^d	18.5 ± 1.5 ^d	5.0 ±0.5 ^{ab}	19.5 ± 0.5 ^e	26.8 ± 0.3
Кр	18.5 ± 0.5 ^e	20.0 ± 0.0 ^c	22.0 ± 0.0 ^f	11.5 ± 0.5 ^a	16.5 ± 0.5 ^{cd}	10.5 ± 0.5 ^{bc}	10.5 ± 0.5 ^b	24.3 ± 0.8
PM	16.5 ± 0.5 ^d	18.0 ± 1.0 ^c	20.5 ± 0.5 ^{ef}	10.5 ± 0.5 ^a	13.5 ± 0.5 ^{ab}	11.5 ± 0.5 ^c	0 ± 0.0^{a}	27.5 ± 0.5
Pa	15.0 ± 0.0 ^c	15.5 ± 0.5 ^{ab}	15.5 ± 0.5 ^b	14.5 ± 0.5 ^{bc}	13.5± 0.5 ^{ab}	0.0 ± 0.0^{a}	10.5 ± 0.5 ^b	26.0 ± 0.5
St	14.5 ± 0.5 [°]	18.0 ± 2.0 ^{bc}	20.5 ± 0.5 ^{ef}	15.5 ± 0.5 ^{cd}	16.0 ± 1.0 ^{cd}	10.5 ± 0.5 ^{bc}	20.5 ± 0.5 ^e	28.0 ± 1.0
Stm	14.5 ± 0.5 [°]	18.5 ± 0.5 ^{bc}	12.0 ± 0.0 ^a	18.5 ± 0.5 ^e	17.5 ± 0.5 ^d	9.5 ± 0.5^{bc}	19.5 ± 0.5 ^e	22.5 ± 1.0
Spa	10.5 ± 0.5 ^ª	14.5 ± 0.5 ^a	17.5 ± 0.5 ^{cd}	13.5 ± 0.5 ^b	13.0 ± 1.0 ^{ab}	0.0 ± 00^{a}	19.5 ± 0.5 ^e	29.3± 0.8
SPb	14.5 ± 0.5 ^c	17.5 ± 0.5 ^{bc}	19.0 ± 1.0 ^{de}	15.5 ± 0.5 ^{cd}	18.5 ± 1.5 ^d	10.0 ± 00 ^{bc}	20.5 ± 0.5 ^e	26.0 ± 0.0
Sa	12.5 ± 0.5 ^b	17.5 ± 0.5 ^{bc}	16.5 ± 0.5 ^{bc}	10.5 ± 0.5 ^a	11.5 ± 0.5 ^a	11.5 ± 0.5 [°]	13.5 ± 0.5 ^c	29.8 ± 0.3
BC	$20.5 \pm 0.5^{\circ}$	27.0 ± 1.0 ^d	$22.0 \pm 0.0^{\circ}$	19.0 ± 1.0 ^e	18.5 ± 1.5 ^d	13.5 ± 0.5 ^c	17.5 ± 0.5 ^d	29.5 ± 0.5

Table 3. Diameter of inhibition zones of methanolic crude extracts on bacteria at 25 mg/mL

The values are the mean of the inhibition diameter ± standard error of the mean of 3 repetitions. a, b, c, d, and f: in the same column, the assigned values of the same letters are not significantly different (p <0.05); Ec: Escherichia coli; Kp: Klebsiella pneumoniae; PM: Proteus mirabilis; Pa: Pseudomonas aeruginosa; St: Salmonella tyhpi; Stm: Salmonella typhimurium; Spa: Salmonella paratyhi A; Spb: Salmonella paratyhi B; Sa: Staphylococcus aureus; BC, Bacillus cereus; Tge: Terminalia glaucescens stem; Tgr: Terminalia glaucescens root; Ks: Khaya senegalensis; Pe: Pterocarpus erinaceus; Bde: Boswellia dalzielii stem; Bdr: Boswellia dalzielii root; Ff: Flacourtia flavescens; ceftriaxone

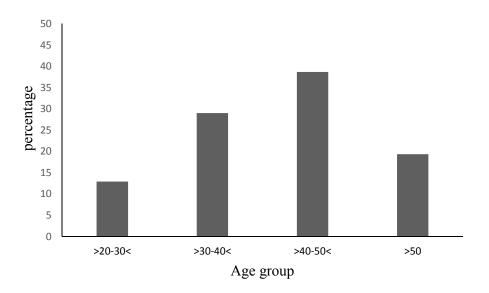
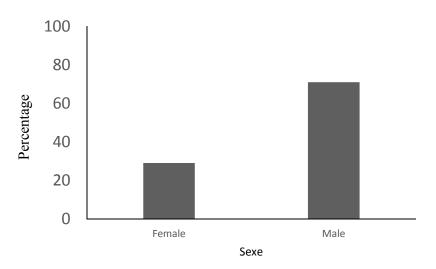


Fig. 1. Distribution of 31 traditional healers surveyed by age group





3.5 Sensitivity of Bacteria to Methanolic Crude Extracts

Table 3 summarizes the behaviour of different germs vis-à-vis the tested extracts. Except for *Flacourtia flavescens* extract, *Bacillus cereus* was the most sensitive bacterium. On the other hand, *Pseudomonas aeruginosa* is resistant to 4 extracts (Tgr, Ks, Bde and Bdr) followed by *Salmonella* para typhi A which is resistant to The, Tgr, Bde and Bdr.

3.6 MIC and CMB of the Crude Methanolic Extracts

It can be seen from Table 4 that *Terminalia* glaucescens bark extracts as well as root extract of *Pterocarpus erinaceus, and Flacourtia* flavecens showed a good activity about Grampositive bacteria with MICs varying between 0.75 mg/mL and 1.5 mg/mL. In contrast, *Boswellia* dalzielii bark extract had the smallest activity with a MIC of 12 mg/mL and CMB> 12 mg/mL

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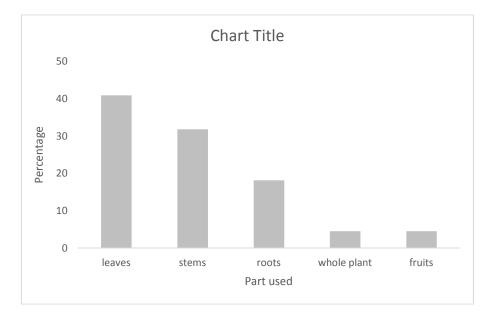


Fig. 3. Repartition of plants according to the parts used

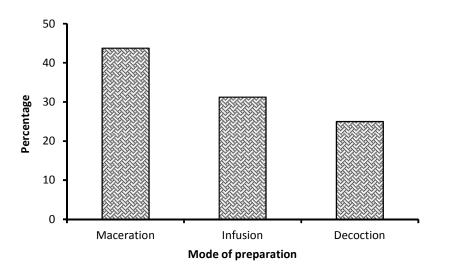


Fig. 4. Repartition of plants according to the mode of preparation

4. DISCUSSION

According to the survey in the Vina and mayo-Louti divisions, a total 15 medicinal plants belonging to 12 botanical families were used in treating different infectious diseases.

From Table 1, *Pterocarpus erinaceus* and *Boswellia dalzielii* were the most common plants reported to be used in the management of infectious diseases in the northern region with frequencies of 8 (13.6%) and 7 (11.86%)

respectively. *Boswellia dalzeilii* is a species of the Genus Boswellia that grows in the northern part of Cameroon[31].

Three plants species (*Boswellia dalzielii*, *Flacourtia flavescens* and *Terminalia glaucescens*) presented higher frequencies of citation. This may be linked to their efficacy since these plants are reported, in Cameroon as well as in other parts of the world, to be used for the treatment of infectious diseases [31-33].

Extract		Ec	Кр	РМ	Ра	St	Stm	Sa	Вс	SpA	SpB
Tge	CMI	3.0	3.0	3.0	3.0	1.5	1.5	1.5	1.5	3.0	1.5
(mg/mL)	CMB	>12	12	12	>12	>12	>12	12	12	>12	>12
	CMB/CMI	>4	4	4	>4	>4	>4	>4	>4	>4	>4
Tgr	CMI	1.5	3.0	1.5	3.0	3.0	3.0	1.5	1.5	3.0	3.0
(mg/mL)	CMB	>12	>12	12	>12	>12	>12	>12	6	12	>12
	CMB/CMI	>4	>4	>4	>4	>4	>4	>4	4	>4	>4
Pe	CMI	0.8	6.0	3.0	3.0	0.8	0.8	0.8	0.8	1.5	1.5
(mg/mL)	CMB	6	12	12	12	6	3.0	6	6	12	>12
	CMB/CMI	>4	2	4	4	>4	4	>4	>4	>4	>4
Bde	CMI	3.0	12.0	12.0	12.0	12.0	12.0	12.0	12.0	6.0	3.0
(mg/mL)	CMB	>12	>12	>12	>12	>12	>12	>12	>12	>12	12
	CMB/CMI	>4	>4	>4	>4	>4	>4	>4	>4	>4	4
Bdr	CMI	3.0	3.0	0.8	3.0	3.0	3.0	1.5	3.0	1.5	3.0
(mg/mL)	CMB	6	>12	6	6	>12	>12	>12	6	12	>12
	CMB/CMI	2	>4	>4	2	>4	>4	>4	2	>4	>4
Ks	CMI	1.5	3.0	1.5	6.0	1.5	3.0	3.0	1.5	6.0	1.5
(mg/mL)	CMB	6	6	6	>12	>12	12	>12	>12	6	12
	CMB/CMI	4	2	4	>4	>4	4	>4	>4	1	>4
Ff	CMI	3.0	0.8	6.0	6.0	3.0	3.0	0.8	0.8	1.5	0.8
(mg/mL)	CMB	>12	6	12	>12	>12	>12	6	3.0	6	12
	CMB/CMI	>4	>4	2	>4	>4	>4	>4	4	>4	>4
Cef	CMI	6.25	6.3	12.5	12.5	3.1	6.25	3.1	6.3	12.5	6.3
(µg/mL)	CMB	25	25	50	50	25	25	25	50	50	25
	CMB/CMI	4	4	4	4	>4	4	>4	>4	4	4

Table 4. Minimal Inhibitory Concentrations (MIC) in mg/ml of Methanolic crude extract against
bacterial strains

Ec : Escherichia coli ; Kp : Klebsiella pneumoniae ; Pm : Proteus mirabilis ; Pa : Pseudomonas aeruginosa ; St : Salmonella tyhpi ; Stm : Salmonella typhimurium ; Spa : Salmonella paratyhi A ; Spb : Salmonella paratyhi B ; Sa : Staphylococcus aureus ; BC, Bacillus cereus Tge: Terminalia glaucescens stem; Tgr: Terminalia glaucescens root; Ks: Khaya senegalensis; Pe: Pterocarpus erinaceus; Bde: Boswellia dalzielii stem; Bdr: Boswellia dalzielii root; Ff: Flacourtia flavescens; cef : ceftriaxone

Indeed. ethnobotanical surveys in many Cameroonian localities reported that Boswellia dalzielii, Flacourtia flavescens and Terminalia glaucescens are used for the treatment of malaria, shingles, ringworm [34,35]. These traditional knowledge on the therapeutic effects of the above medicinal plants are confirmed by various pharmacological study data, which demonstrated immunomodulatory activities of B. dalzielii aqueous extract and methanol fraction [31], the antibacterial activities of T. glaucescens of ethanol extract of bark and root with MIC between 0.625 to 1.25 mg/ml [21], the antioxidant properties of methanolic extracts of Flacourtia flavescens [36].

From Fig. 1 it is noted that thirty-one (31) traditional healers were interviewed. The informants were between 20 and over 50 years old, and the modal class was 40 to 50 years old. Among these informants, there were nine women and 22 men (Fig. 2). These small number could be because traditional healers are scarce

throughout our country [37] and thus represent a very little portion of the general population and also because some of them refused to be interviewed. This might also be because, young people to whom traditional knowledge on medicinal plants effects could have been transmitted are not eager to learn and exile to city for jobs and better living condition purposes [37]. This situation is the same worldwide since cultural changes as a result of westernization and modernization have contributed enormously in making the younger generation undermine African traditional values [38]. The low representativeness of women in this study is due, on the one hand, to the fact that traditional medicine has long been exercised by men and also to the fact that in these Regions women are not allowed to interact with foreigners.

Data from Fig. 3 showed that, the most commonly used plant parts were the leaves (40.91%). In fact, leaves are known to accumulate plants secondary metabolites such

as alkaloids, tannins and saponin, which are active components responsible for many medicinal properties [39]. Moreover, utilization of leaves and stems is advantageous since their harvest does not induce irreversible destruction of plants like that of roots or whole plant [40].

Medicinal plants were prepared in different forms including maceration (43.75%), infusion (25.25%) and decoction (25.00%) (Fig. 4). These preparations are made only with water and orally administered. These modes of preparation and administration are the most used in traditional medicine. Similar results were obtained in previous ethnobotanical surveys carried out in Cameroon and another part of the world [38,41, 42]. The high frequency of maceration is related to the fact that this method does not alter the active principle of infusion and decoction do.

4.1 Antimicrobial Activity of Selected Plants

According to Popova et al., [43] scale, which state that natural products with minimum inhibitory concentrations (MIC) range between $100-1000 \ \mu g \ mL^{-1}$ *in vitro* on at least one Grampositive and one Gram-negative bacteria can be classified as antimicrobials, crude extract of *Pterocarpus erinaceus* and *Flacourtia flavescens* shown good antibacterial activity (MIC = 750 $\ \mu g/ml$).

These observations corroborate those authors, who confirmed the use of these plants in the treatment of infectious diseases [34,44,45]. This may be ascribed to the different classes of compounds found in these extracts. In fact, the phytochemical screening of these extracts revealed the presence of phenols, tannins, terpenoids. flavonoids. steroids. alkaloids. anthraquinones, anthocyanins, saponins and coumarins. Individual antibacterial activities of these secondary metabolites have been demonstrated [46,47]. Nevertheless, Boswellia dalzielii root extract was active only against P. mirabilis. But, Terminalia glaucescens stem extract, Terminalia glaucescens root extract and Khaya senegalensis extract shown moderate activity (MIC 1500 to 3000 µg/mL) depending on the bacterium.

The antibacterial activity of *Boswellia dalzeilii* from Cameroon are reported here for the first time. The values of the diameters of inhibition, MICs and MBC showed that the degree of activity varied with the bacteria and the extracts. This variation of the activity could be due to the difference of solubility of the active ingredient in each extract on the one hand and to the constitutional or structural variability of the tested germs on the other hand. Also, it could be due to the capacity of the organisms to modify the structure of the active principle [2]. Moreover, the differences in susceptibility may be explained by the differences in cell wall composition and genetic content of plasmids that can be easily transferred among strains.

5. CONCLUSION

This ethnobotanical survey study has revealed that there are high knowledge and use of medicinal plants in northern Cameroon. Methanol crude extract of different plants showed different degrees of antibacterial activities against bacterial strains enteropathogenic used. These biological results allowed us to conclude that the methanol extracts of the *Boswellia dalzielii* roots as well *Flacourtia flavescens* and *Pterocarpus erinaceus* barks are the most active. The information from this study can serve as guide for the discovery of new antibacterial drugs from medicinal plant

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

 Voukeng IK, Kuete V, Dzoyem JP, Fankam AG, Noumedem JAK, Kuiate JR, Pages J. M. Antibacterial and antibiotic-potentiation activities of the methanol extract of some cameroonian spices against Gramnegative multi-drug resistant phenotypes. BMC Research Notes. 2012;5(299):1–10. Available:<u>https://doi.org/10.1186/1756-</u>0500-5-299

- Gatsing D, Nkeugouapi FNC, Nji-Nkah F. B. Antibacterial activity, bioavailability and acute toxicity of the leaf extract of *Alchornea cordifolia* (Euphorbiaceae). International Journal of Pharmacology; 2010.
- Robert-Jan H, Goessens WHF, Wilfrid van P, Dik JM, Bruno HS, Molhoek N, Perry JJ, van G. Salmonella Subtypes With Increased MICs for Azithromycin in Travelers returned to the Netherlands. Emerging Infectious Diseases. 2014;20(4): 705–708.
- Diseases, N. I. of allergy and infectious. (2014). The Antimicrobial Resistance Research Program of the National Institute of Allergy and Infectious Diseases: Current Status and Future Directions.
- 5. Alexander B, Perfect J. Antifungal resistance trends toward the year 2000, implication for therapy and new approaches, Drugs. 1997;54:657-678.
- Quiroga E, Sampietro A, Vattuone A. Screening antifungal activities of selected medicinal plants. Journal of Ethnopharmacology. 2001;74:89-96.
- Ghosh A., Das B, Roy A, Mandal B, Chandra G. Antibacterial activity of some medicinal plant extracts. Journal of Natural Medicines. 2008;62:259–262.
- Eloff J. Which extractant should be used for the screening and isolation of antimicrobial components from plants? Journal of Ethnopharmacology. 1998;60:1–8.
- Fodouop DG, Ngo TG, Cheseto X, Talom TB, Nji-kah B, Torto B. A chemical constituent, the antibacterial and antioxidant activity of essential oil from of L. abyssinica. International Journal of Phytomedicine. 2014;6(2).
- 10. Prashant Tiwari B, Kumar MK, Gurpreet Kaur HK. Phytochemical screening and extraction - A review. Internationale Pharmaceutica Sciencia. 2011;1(1):98– 106.

Available: http://www.ipharmsciencia.com

 Shaheen N, Azhar I, Mahmood ZA. Comparative phytochemical investigations for standardization of some spices available in Pakistan. World Journal of Pharmaceutical Sciences. 2015;3(8): 1496–1506.

- 12. Abubakar EM. Antibacterial activity of crude extracts of *Euphorbia hirta* against some bacteria associated with enteric infections. Journal of Medicinal Plants Research. 2009;*3*(7):498–505.
- Titilope KK, Rashidat EA., Christiana OC, Kehinde ER, Omobolaji JN, Jubril A. Invitro antimicrobial activities of *Euphorbia hirta* against some clinical isolates. Agriculture and Biology Journal of North America. 2012;3(4):169–174. Available:https://doi.org/10.5251/abjna.201 2.3.4.169.174
- Abiodun F, Obasuyi O. Phytochemical screening and evaluation of stem bark extract of *Khaya senegalensis* (Meeliaceae) on methicilin. *Canadian* Journal of Pure and Applied Sciences, 2009;3(3):925–928.
- Idu M, Erhabor J, Oshomoh E, Ovuakporie-Uvo PO. Phytochemical composition and antimicrobial properties of the seeds of *Khaya senegalensis* (Desc.) A. Juss. Plant Biology. 2014;1(4):1–4.
- 16. Gabriel AF, Onigbanjo HO. Phytochemical and antimicrobial screening of the stem bark extracts of *Pterocarpus erinaceus* (Pair). 2010;18:1–5.
- Al-Snafi PDAE. The pharmacological and therapeutic importance of Eucalyptus species grown in Iraq. IOSR Journal of Pharmacy. 2017;7(3):72–91. Available:https://doi.org/10.9790/3013-0703017291
- Aravind G, Bhowmik D, Duraivel S, Harish G. Traditional and medicinal uses of carica papaya. Journal of Medicinal Plants Studies. 2013;1(1):7–15.
- Nwinyi FC, Binda L, Ajoku GA, Aniagu S O, Enwerem NM, Orisadipe A, Gamaniel KS. Evaluation of the aqueous extract of *Boswellia dalzielii* stem bark for antimicrobial activities and gastrointestinal effects. African Journal of Biotechnology. 2004;3(5):284–288.
- 20. Kaou AM, Mahiou-Leddet V, Canlet C, Debrauwer L, Hutter S, Laget M, Ollivier E. Antimalarial compounds from the aerial parts of Flacourtia indica (Flacourtiaceae). Journal of Ethnopharmacology, (2010).130(2), 272–274. https://doi.org/10.1016/j.jep.2010.04.045
- 21. Bamidele FA, Ogundipe FO, Adebayo-Oyetoro AO, Aromolaran, Odeyemi OA, Monigbeyin E. *In vitro* Antibacterial activity and preliminary phytochemical screening of four plants on selected clinical

pathogens. International Journal of Scientific and Research Publications. 2013;3(11):1–4.

- Ahoton L, Quenum F, Mergeai G. Evaluation agromorphologique et sélection des meilleures accessions de Pourghère (*Jatropha curcas* L.) introduces au Bénin. International Journal of Biological and Chemical Sciences. 2012;5(4):1619–1627. Available:<u>https://doi.org/10.4314/ijbcs.v5i4.</u> 25
- 23. Mamun-Or-Rashid AN, Mahmud S, Towfique NM, Sen MK. A compendium ethnopharmaceutical review on *Euphorbia hirta* L. 2013;2(2):14–21.
- 24. Ngo Bum E, Soudi NS, Rakotonirina SV, Rakotonirina A. Psorospermum febrifugum Spach (Hypericaceae) decoction antagonized chemically-induced convulsions in mice. International Journal of Pharmacology. 2005;1(2):118–121.
- Iwalewa EO, Omisore NO, Daniyan OM, Adewunmi CO, Taiwo BJ, Fatokun OA, Oluborode IO. Elemental compositions and anti-anemic property of Harungana madagascariensis stem bark. Bangladesh Journal of Pharmacology. 2009;4(2):115– 121. Available:https://doi.org/10.3329/bjp.v4i2.1

Available:<u>https://doi.org/10.3329/bjp.v4i2.1</u> 641

- Afieroho OE, Susan, Izontimi S, Okoroafor Dorcas O, Blessing C. Antibacterial and phytochemical evaluation of *Harungana madagasriensis* L (Hypericaceae) seeds. International Research Journal of Pharmacy. 2012;3(11):75–77.
- Foyet HS, Asongalem AE, Oben EK, Cioanca O, Hancianu M, Hritcu L. Effects of the Methanolic extract of *Vitellaria paradoxa* stem bark against scopolamineinduced cognitive dysfunction and oxidative stress in the rat hippocampus. Cellular and Molecular Neurobiology. 2016;36(7):1139–1149. Available:<u>https://doi.org/10.1007/s10571-015-0310-7</u>
- Metowogo K, Eklu-Gadegbeku K, Agbonon A, Aklikokou KA, Gbeassor M. Gastroprotective Effect of hydroalcoholic extract of aloe Buettneri kossi. Iranian Journal of Pharmaceutical Research. 2011;10(1):69–74.
- Okeke MI, Okoli AS, Eze EN, Ekwume GC, Okosa EU, Iroegbu CU. Antibacterial activity of *Citrus limonum* fruit juice extract. Pakistan Journal of Pharmaceutical Sciences. 2015;28(5):1567–1571.

- Yaouba A, Tchikoua R, Tatsadjieu NL. Antibacterial effect of plant extracts against some pathogenic bacteria. International Journal of Natural Products Research. 2012;1(4):83–87.
- Oumar M, Tume C, Kamtchueng MO, 31. Kamanyi A. In vitro effect of aqueous extract, hexane and methanol fractions of Boswellia dalzeilii, hutch (family: burseraceae) in immunomodulatory human monocvtes activities of 1 macrophages. International Journal of Biological & Pharmaceutical Research. 2014;5(2):201-209.
- Ohemu TL, Agunu A, Olutu PN, Ajimz U, Dafam DG, Awila JJ. Ethnobotanical survey of medicinal plants used in the traditional treatment of viral infections in Jos, Plateau state, Nigeria. Int. J. Med. Arom. Plants. 2014;4(2):74-81.
- 33. Lagnika L, Attioua B, Vonthron-Senecheau C, Kaiser M, Lobstein A, Sanni A, Weniger preliminary B. In vitro studv of antiprotozoal effect of four medicinal plants from Benin In vitro preliminary study of antiprotozoal effect of four medicinal plants from Benin. Journal of Medicinal plantS Research. 2013;7(10):556-560. Available:https://doi.org/10.5897/JMPR012 .253
- Saotoing P, Toua V, Tchobsala, Fohouo FNT, Nloga AMN, Messi J. Medicinal plants used in traditional treatment of malaria in Cameroon. Journal of Ecology and the Natural Environment. 2011;3(3): 104–117.
- Kémeuzé VA, Mapongmetsem PM, Tientcheu MA, Nkongmeneck BA, Jiofack RB. Boswellia dalzielii Hutch: État du peuplement et utilisation traditionnelle dans la région de Mbé (Adamaoua-Cameroun). Secheresse. 2012;23:278– 283.

Available:<u>https://doi.org/10.1684/sec.2012.</u> 0365

36. Ndhlala AR, Chitindingu K, Mupure C, Murenje T, Ndhlala F, Benhura MA, Muchuweti M. Antioxidant properties of methanolic extracts from *Diospyros mespiliformis* (jackal berry), *Flacourtia indica* (Batoka plum), *Uapaca kirkiana* (wild loquat) and *Ziziphus mauritiana* (yellow berry) fruits. International Journal of Food Science and Technology. 2008;43(2):284–288. Available:<u>https://doi.org/10.1111/j.1365-</u>

<u>2621.2006.01431.x</u>

- Yemele DM, Telefo BP, Goka CS, Nguelefack BT, Fodouop CPS, Tagne SR, Lienou LL, Nguemo F, Moundipa FP. *In vitro* cytotoxicity studies plants used for pregnant women's health conditions in Menoua Division-West Cameroon. International Journal of Phytomedicine. 2015;7(2):235-239.
- Giday M, Asfaw Z, Elmqvist T, Woldu Z. An ethnobotanical study of medicinal plant used by Zay people in Ethiopia. Journal of ethnopharmacology. 2003;85:43-52.
- 39. Focho DA, Nkeng EAP, Lucha CF, Ndam WT, Afegenu IA. Ethnobotanical survey of plants used to treat diseases of the reproductive system and preliminary phytochemical screening of some species of malvaceae in Ndop central sub-division, Cameroon. Journal of Medicinal Plants Research. 2009b;3(4):301–314.
- 40. Telefo PB, Lemfack MC, Bayala B, Lienou LL, Goka CS, Yemele MD, Mouokeu C, Tagne SR, Moundipa FP. Enquête ethnopharmacologique des plantes utilisées dans le traitement de l'infertilité féminine dans les localités de Fossong-Wentcheng et Foto, Cameroun. Phytotherapie. 2012;10(1):25–34.
- Focho DA, Ndam WT, Fonge BA. Medicinal plants of Aguambu –Bamumbu in the Lebialem highlands, southwest province of Cameroon. African Journal of Pharmacy and Pharmacology. 2009a;3(1): 1–13.
- Embeya VO, Simbi JBL, Stévigny C, vandenput S, Shongo CP, Duez P. Traditional plant-based remedies to control

gastrointestinal disorders in livestock in the regions of Kamina and Kaniama. Journal of Ethnopharmacology. 2014;153(3):686–693(14).

 Popova M, Dimitrova R, Al-lawati HT, Tsvetkova I, Najdenski H, Bankova V. Omani propolis: chemical profiling, antibacterial activity and new propolis plant sources. Chemistry Central Journal. 2013; 7(1):1–8.

Available:<u>https://doi.org/10.1186/1752-153X-7-158</u>

- 44. Anago E, Lagnika L, Gbenou J, Loko F, Moudachirou M, Sanni A. Antibacterial activity and phytochemical study of six medicinal plants used in Benin. Pakistan Journal of Biological Sciences. 2011;1–7.
- 45. Kota GC, Karthikeyan M, Kannan M. Flacourtia indica (Burm. f.) Merr. -A phytopharmacological review. International Journal. 2012;3(1):78–81.
- 46. Krief S. Métabolites secondaires des plantes et comportment animal : Surveillance sanitaire et observations de l'alimentation de chimpanzés (*Pan Troglodytes schweinfurthii*) en Ouganda ; Activités biologiques et étude chimique de Plantes consommées. Muséum National D'histoire Naturelle; 2003.
- 47. Paiva P, Gomes F, Napoleão T. Antimicrobial activity of secondary metabolites and lectins from plants. Current Research, Technology and Education Topics in Applied Microbiology and Microbial Biotechnology. 2010;396– 406.

Available: https://doi.org/10.3390/v6031037

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