

IN VITRO ANTIBACTERIAL EFFECT OF AQUEOUS, ACETONIC, AND ETHANOLIC STEM BARK EXTRACTS OF *COMBRETUM COLLINUM* FRESEN. (COMBRETACEAE) ON MULTIDRUG-RESISTANT *STAPHYLOCOCCUS AUREUS* STRAINS AND PHYTOCHEMICAL SCREENING

Sanogo Yacouba^{*123}, Kone Dramane¹, Bamba Moussa²

¹Laboratory for Environment, Climate, Health, Engineering and Sustainable Development, Peleforo Gon Coulibaly University, BP 1328 Korhogo, Côte d'Ivoire.

²Laboratory of Bacteriology-Virology, Pasteur Institute of Côte d'Ivoire, 01 BP 490 Abidjan 01, Côte d'Ivoire.

³Phytochemistry Laboratory, Swiss Center for Scientific Research, Côte d'Ivoire, 01 BP 1303 Abidjan 01, Côte d'Ivoire.

Article Received: 31 June 2026 | Article Revised: 22 July 2026 | Article Accepted: 12 August 2026

***Corresponding Author: Sanogo Yacouba**

Laboratory of Bacteriology-Virology, Pasteur Institute of Côte d'Ivoire, 01 BP 490 Abidjan 01, Côte d'Ivoire.

DOI: <https://doi.org/10.5281/zenodo.21949081>

How to cite this Article: Sanogo Yacouba, Kone Dramane, Bamba Moussa (2026) IN VITRO ANTIBACTERIAL EFFECT OF AQUEOUS, ACETONIC, AND ETHANOLIC STEM BARK EXTRACTS OF *COMBRETUM COLLINUM* FRESEN. (COMBRETACEAE) ON MULTIDRUG-RESISTANT *STAPHYLOCOCCUS AUREUS* STRAINS AND PHYTOCHEMICAL SCREENING. World Journal of Pharmaceutical Science and Research, 5(8), 774-785.



Copyright © 2026 Sanogo Yacouba | World Journal of Pharmaceutical Science and Research.

This work is licensed under creative Commons Attribution-NonCommercial 4.0 International license (CC BY-NC 4.0).

ABSTRACT

Combretum collinum Fresen. (Combretaceae) is a plant native to Côte d'Ivoire commonly used to traditionally treat various ailments, including envenomations, ear infections, diarrhea, infertility, gonorrhea, epilepsy, and dengue fever. This work contributes to the development of this plant, aiming to promote its sustainable management and conservation among local populations. Like many medicinal plants, it is subject to human pressure that could lead to its extinction. The study aims to evaluate the antibacterial activity of aqueous, acetone, and ethanolic extracts of *Combretum collinum* stem bark on the *in vitro* growth of methicillin-resistant *Staphylococcus aureus* strains. Diffusion in agar and macrodilution in liquid medium were used to evaluate the sensitivity of bacterial strains to the extracts and to determine their antibacterial parameters (MIC and MBC). The MICs obtained ranged from 0.60 to 9.72 mg/ml. The MBCs ranged from 0.60 to 9.72 mg/ml for the ethanolic extract and from 1.21 to 4.86 mg/ml for the aqueous extract, demonstrating the bactericidal activity of these extracts. Furthermore, qualitative analysis by thin-layer chromatography on the extracts detected several groups of compounds. These included tannins, flavonoids, polyphenols, polyterpenes, anthocyanins, and anthraquinone glycosides, most of which have known antibacterial effects. This work justifies the traditional use of *C. collinum* in the treatment of various bacterial infections, particularly those induced by *Staphylococcus aureus*.

KEYWORDS: Antibacterial, *Combretum collinum*, Ivorian flora, Qualitative analysis.

1. INTRODUCTION

Modern medicine and its advances in treating bacterial infections have led populations to turn away, somewhat, from traditional medicine. Indeed, the advent of antibiotics has been a true boon for humanity, as they have contributed to significantly reducing the spread of these diseases. However, bacterial resistance to these molecules is becoming increasingly common.^[1] This is the case with methicillin-resistant *Staphylococcus aureus* (MRSA). In fact, this highly adaptive bacterium has developed various resistance mechanisms to anti-staphylococcal agents. More than 90% of strains produce penicillinase. These strains now exhibit cross-resistance between M-penicillins (methicillin, oxacillin) and other β -lactams through the production of a protein, PLP2a, which binds penicillins (PLP) and has a low affinity for these compounds. The gene (*mecA*) encoding PLP2a is carried by a chromosomal element which also contains other genes for resistance to heavy metals and other antibiotics, hence the multi-resistance profile of *S. aureus* to methicillin.

In Côte d'Ivoire, numerous cases of multidrug-resistant bacteria have been reported.^[2] This bacterial resistance, resulting from the continuous and even uncontrolled use of antibiotics, has contributed to making microbial-related illnesses the leading cause of death worldwide.^[3] Bacteria are responsible for 70% of these deaths.^[4] To address this situation, which is compounded by the high cost of modern medicines, much hope remains in the secrets of medicinal plants, and the emergence of alternative medicine based on these plants is more relevant than ever.

Indeed, the modern pharmaceutical industry itself still relies heavily on the diversity of plant secondary metabolites to find new molecules with novel biological properties.^[5] This source seems inexhaustible since each of the 400,000 known species can contain several thousand different constituents whose various uses aim to alleviate suffering and improve human health.^[6]

In this context, some researchers have conducted antimicrobial tests on many of these plants.^[6,7] However, several of them are threatened with extinction due to poor human management. This is the case for *Combretum collinum* Fresen (Combretaceae), a promising plant widely used in traditional African medicine to treat various ailments, some of which are bacterial in origin. These include, in particular, diarrhea, urinary tract infections, and ear infections caused by *Staphylococcus aureus*.^[8]

To further develop the field of Ivorian traditional medicine, we chose to evaluate the antibacterial activity of *Combretum collinum* stem bark against strains of *Staphylococcus aureus* resistant to conventional antibiotics and to conduct a phytochemical screening. This research could help the population understand the urgency and necessity of conserving and protecting this medicinal plant.

2. MATERIALS AND METHODS

2.1 Bacterial strains

The germs originate from the bacterial strain bank of the Department of Bacteriology and Virology of the Pasteur Institute of Côte d'Ivoire and the University Hospitals (CHU) of Cocody and Yopougon (municipalities of Abidjan). They consist of eight isolates of *Staphylococcus aureus*, including one reference strain (*Staphylococcus aureus* ATCC 25923) allowing quality control, five strains of resistant phenotype and two strains of wild or susceptible phenotype (Table 1).

Table 1: Selected *Staphylococcus aureus* bacterial strains.

<i>Staphylococcus aureus</i> strain code	Organic product	Resistance phenotypes
64y	Pus	MRSA, Kana.R
10y	Pus	MRSA, Kana.R
114c	Pus	Savage
75c	Urine	MRSA, FQR
56c	Tooth decay	MRSA, Kana.R
48c	Urine	MRSA, FQR
66c	Blood	Sauvage
ATCC 25923	Reference strain	

MRSA: Methicillin resistance; Kana R: Kanamycin resistance; FQR: Fluoroquinolone cross-resistance; ATCC: American Type Culture Collection.

Laboratory Equipment

The laboratory equipment used to obtain the crude extracts consisted of a rotary evaporator, a freeze dryer, a Voltex shaker, a magnetic stirrer, a Densimat, an oven, a fume hood, a refrigerator, automatic micropipettes (P200 and P1000), and a balance for determining the collected masses. A UV lamp, a chromatography tank, a digital camera, a hot plate, and reagents were used for phytochemical sorting. As for the glassware used to carry out the antibacterial activities, it consists of beakers (100 mL and 250 mL), test tubes, graduated cylinders, funnels, sterile Petri dishes of 90 mm diameter, jars, Pasteur pipettes, hemolysis tubes, a stand, syringes (5 mL and 10 mL), sterile glass pipettes (10 mL), microfilters (0.22 µm and 0.45 µm), swabs, Whatman® 3 mm filter paper, aluminum foil, spatulas, latex gloves, masking tape, absorbent cotton, a timer, 100 and 1000 µL tips, F254 silica gel plates, micropillar tubes and bulbs. Sterile distilled water, hexane, acetone, and ethanol were the solvents used to prepare the crude extracts.

2.2 Preparation of crude extracts

The samples were collected at the beginning of the dry season in the savannas of northern Côte d'Ivoire. These harvests took place just before sunset, allowing the plants sufficient time to secrete active compounds to combat the day's stresses. The plant samples were dried and then ground into a powder, which underwent several extractions using a method described by,^[9] adapted for this study. To do this, 25 g of plant powder were macerated in 250 ml of hexane for 24 hours under magnetic stirring to degrease the powder. The residue was dried on blotting paper and weighed, then added to 250 ml of acetone. After 24 hours of maceration with stirring, the filtrate was evaporated using a rotary evaporator at 40 °C to remove the alcohol and dried under a fume hood to obtain the acetone extract. Repeating the same process with ethanol yielded the ethanolic extract. The aqueous extract was obtained by macerating 50 g of plant powder in 500 ml of distilled water, placed on a magnetic stirrer for 24 hours. Freeze-drying the filtrate produced the aqueous extract. All three extracts (acetonic, ethanolic, and aqueous) were stored in a freezer.

2.3 Performing antibacterial tests

2.3.1 Preparing the inoculum for solid-state tests

Young 24-hour strains were emulsified in 2 ml of 85% NaCl suspension, and the optical density was adjusted to 0.5 MaC Farland using a densimat. A volume of 1 ml was diluted in 10 ml of physiological saline (0.9% NaCl), thus constituting the bacterial inoculum estimated at 10⁶ bacteria/ml.

2.3.2 Germ susceptibility testing of extracts

Before evaluating any activity, the extracts underwent a sterility test to verify whether or not they were contaminated by a germ. Antibigrams by diffusion in agar wells and the macrodilution method in liquid medium were used to perform the tests.^[10] Petri dishes containing Muller-Hinton (MHA) Agar were inoculated by swabbing with the bacterial inoculum. Fifty μ l of a solution of extracts prepared at 50 mg/ml was added to wells in the agar (Figure 1).

The plates were incubated at 37 °C for 24 h. After this time, the inhibition zone diameter around each well was measured using calipers. The effectiveness of the extracts was assessed according to the criteria of.^[11] A substance is considered ineffective if the inhibition diameter (ID) is less than 8 mm, while it is considered effective if the ID is between 9 and 14 mm. It is judged to be very effective when the ID is between 15 and 19 mm, and extremely effective if it is greater than 20 mm.



Figure 1: Sensitivity test of *S. aureus* 48c to the ethanolic extract at 50 mg/mL.

2.3.3 Preparation of the inoculum for liquid-based tests

Young colonies (24 hours old) were collected and emulsified in a test tube containing 10 ml of sterile Muller-Hinton broth. The mixture was incubated at 37°C for 3 hours. After this incubation, a 0.3 ml suspension of this preculture was diluted in 10 ml of sterile Muller-Hinton broth and then homogenized.

2.3.4 Preparation of the concentration range

The concentration range was obtained by the double dilution method. To do this, a 50 mg/ml solution of the extracts was prepared in sterile distilled water. A series of 2:1 dilutions were performed on this solution to obtain a concentration range from 50 to 0.19 mg/ml.

2.3.5 Determination of Antibacterial Parameters

This activity was carried out using the most active extracts in the sensitivity tests, namely the aqueous and ethanolic extracts. The determination of antibacterial parameters was performed by dilution in liquid medium according to the method used by.^[5] Thus, in 10 experimental hemolysis tubes, 0.2 ml of each extract concentration was mixed with 1.8 ml of bacterial inoculum. The growth control tube received 0.2 ml of sterile distilled water in addition to the inoculum, while the sterility control received only 2 ml of sterile Muller-Hinton broth. This distribution yielded a new range of concentrations from 5 to 0.019 mg/ml, representing a 1:10 dilution. The tubes were incubated for 24 hours at 37°C.

After this incubation period, the lowest concentration at which no bacterial growth was observed with the naked eye corresponds to the Minimum Inhibitory Concentration (MIC). The Minimum Bactericidal Concentration (MBC) yields 0.01% viable bacteria after 24 hours of incubation at 37°C. Its determination began with enumeration. This consisted of diluting the initial inoculum from 10^{-1} to 10^{-4} and inoculating these different dilutions using a 2 µl calibrated loop in 5 cm streaks onto a MHA (Muller Hinton Agar) and then incubating for 24 hours. These Petri dishes were labeled A.

After reading the MICs, the contents of the tubes in which no visible growth was observed were used to inoculate the MHA in 5 cm streaks. This series of Petri dishes is named B. The MBC was determined by comparing the bacterial growth of dishes A and B. Thus, the smallest concentration in the tube that has less than 0.01% viable bacteria relative to the initial inoculum is the MBC.

The MBC/MIC ratio helped to clarify the mode of action of the extracts.^[13] According to^[14], an extract is bactericidal when its MBC is equal to its MIC or if the MBC/MIC ratio is less than or equal to 4. It is considered bacteriostatic when the MBC/MIC ratio is greater than 4. When this ratio is equal to 32, the strain is considered tolerant.

2.4 Phytochemical Screening

The characterization of the different chemical compounds in the extracts was performed by thin-layer chromatography (TLC) according to the method used by.^[12] This method allows the detection of several groups of secondary metabolites by specific colorations, either in the visible spectrum or at a given wavelength.^[15] A 10 mg mass of extracts was dissolved in 1 ml of absolute methanol to obtain a 10 mg/ml solution. Ten microliters (10 µl), or 100 µg, of this solution were spot-deposited onto a Silicagel 60F254 plate (stationary phase) using a microcapillary tube. The chromatograms were developed in tanks previously saturated with CHCl₃-MeOH-H₂O eluent or mobile phase (65:35:5 v/v/v), and then dried. These plates were observed before and after development either in the visible spectrum or under a UV lamp at 254 and 366 nm.

2.4.1 Detection of Terpenoids and Saponins

These compounds are detected using Godin's reagent. After spraying the plate with Godin's reagent and heating it at 100 °C for 10 minutes, various colors appear. In the visible spectrum, violet and red spots indicate the presence of monoterpenes, and blue spots indicate saponins.^[16]

2.4.2 Detection of Alkaloids

After spraying with Dragendorff's reagent and heating the chromatogram at 100 °C for 10 min, the alkaloids appear as orange spots in the visible spectrum.

2.4.3 Detection of Polyphenols

After spraying the chromatogram with 10% Folin-Ciocalteu reagent, then heating at 100 °C for 10 min, the blue spots observed in the visible range confirm the presence of polyphenols.^[17]

2.4.4 Detection of Coumarins

5% (w/v) basic lead acetate (CH₃COO)₂ Pb was used for sputtering the chromatogram. Green and blue fluorescence spots under UV light at 366 nm indicate the presence of coumarins.

2.4.5 Detection of Tannins

The appearance of spots of various colors (blue, green, black), observable in the visible spectrum, after sputtering the chromatogram with a 10% FeCl₃ solution, indicates the presence of tannins.

2.4.6 Detection of Anthraquinones and Anthrones

A 5% ethanolic solution of KOH was sprayed onto the chromatogram. The red spots observed in the visible range and at 366 nm confirm the presence of anthraquinones. In contrast, anthrones are visible at 366 nm as yellow spots.^[18]

2.5 Statistical Analyses

Analysis of variance (one-way ANOVA) followed by Tukey's test was used to compare the variations in MICs and MBCs, and to determine whether the activity of the *Combretum collinum* stem bark extracts was statistically influenced by the bacterial phenotypes. The results are expressed as means $\pm$ standard deviations. Probability values ($P < 0.05$) were considered statistically significant. The R software^[19] was used to perform these statistical tests.

3. RESULTS

3.1 Antibacterial Activity

Extracts from the stem bark of *Combretum collinum* were active to varying degrees against all the bacterial strains studied. The ethanolic extract yielded the largest mean inhibition zone diameter (CID) of 22.0 ± 0.1 mm. This was followed by the aqueous and acetone extracts, with CIDs of 21.7 ± 0.6 mm and 17.2 ± 4.3 mm, respectively. These mean CIDs were observed against *Staphylococcus aureus* 48c. This strain, exhibiting a resistant phenotype (MRSA, cross-resistance to fluoroquinolones), was isolated from the urine of a patient with a urinary tract infection (Figure 2).

The ethanolic and aqueous extracts exhibited minimum inhibitory concentrations (MICs) ranging from 0.60 to 9.72 mg/ml against all bacteria. The lowest value (0.60 mg/ml) was obtained with the ethanolic extract against *S. aureus* strains 10y and 48c, and with the aqueous extract against *S. aureus* strain 48c. Minimum bactericidal concentrations (MBCs) ranged from 0.60 to 9.72 mg/ml for the ethanolic extract and from 1.21 to 4.86 mg/ml for the aqueous extract (Table 2). The latter was bactericidal against all bacterial strains tested, unlike the ethanolic extract, which only showed bactericidal activity against 50% of the bacteria.

Statistical analysis compared the mean $\pm$ standard deviation values obtained for wild-type bacterial strains (*S. aureus* 114c, 66c) and resistant bacterial strains (*S. aureus* 64y, 10y, 75c, 56c, 48c). Analysis of variance showed that the observed differences in MICs were not significant, and the same was true for MBCs (Tables 3 and 4 ; $P > 0.05$).

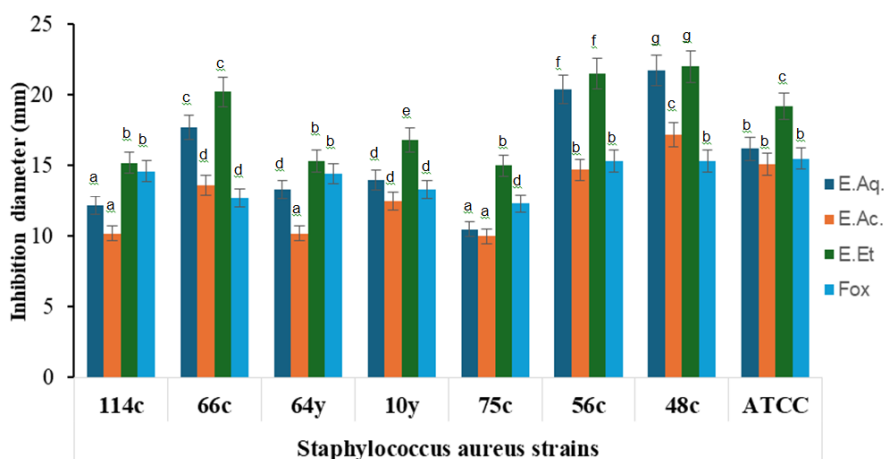


Figure 2: Activity (mean $\pm$ standard deviation) of *Combretum collinum* Fresen trunk bark extracts at 50 mg/ml against *Staphylococcus aureus* strains.

E.Aq.: Aqueous extract; E.Ac.: Acetone extract; E.Et.: Ethanol extract; Fox: cefoxitin; ATCC: American Type Culture Collection.

The same lowercase letters indicate similar activities at the $\alpha = 5\%$ threshold.

Table 2: Mean values (mean $\pm$ SD of three trials) of the antibacterial parameters of the stem bark extracts of *Combretum collinum* on the strains of *S. aureus*.

Bacteria	Ethanol extract (mg/ml)				Aqueous extract (mg/ml)			
	MIC	MBC	MBC/MIC	Power	MIC	MBC	MBC/MIC	Power
<i>S. aureus</i> 64y	1,21	2,43	2	Bcide	1,21	1,21	1	Bcide
<i>S. aureus</i> 10y	0,60	3,60	6	Btique	1,21	2,43	2	Bcide
<i>S. aureus</i> 114c	1,21	6,05	5	Btique	1,21	2,43	2	Bcide
<i>S. aureus</i> 75c	1,21	2,43	2	Bcide	1,21	2,43	2	Bcide
<i>S. aureus</i> 56c	1,21	6,05	5	Btique	1,21	2,43	2	Bcide
<i>S. aureus</i> 48c	0,60	0,60	1	Bcide	0,60	1,21	2	Bcide
<i>S. aureus</i> 66c	1,21	9,72	8	Btique	2,43	4,86	2	Bcide
<i>S. aureus</i> ATCC 25923	2,43	2,43	1	Bcide	2,43	2,43	1	Bcide

MIC: minimum inhibitory concentration ; MBC: minimum bactericidal concentration ; Bcide: bactericidal ; Btique: bacteriostatic.

Table 3: Comparison of mean values (mean $\pm$ SD of three trials) of MIC and MBC of the ethanolic extract according to the phenotypes of the bacterial strains.

Antibacterial parameters	Wild bacterial strains	Resistant bacterial strains	P-value
MIC (mg/ml)	0,648 $\pm$ 4,1	0,900 $\pm$ 3,2	P > 0,05
MBC (mg/ml)	1,9146 $\pm$ 2,0	1,3186 $\pm$ 2,0	P > 0,05

MIC: minimum inhibitory concentration; MBC: minimum bactericidal concentration.

Table 4: Comparison of mean values (mean $\pm$ SD of three trials) of MIC and MBC of the aqueous extract according to the phenotypes of the bacterial strains.

Antibacterial parameters	Wild bacterial strains	Resistant bacterial strains	P-value
MIC (mg/ml)	1,297 $\pm$ 1,3	1,4692 $\pm$ 3,1	P > 0,05
MBC (mg/ml)	1,915 $\pm$ 1,6	1,3548 $\pm$ 2,2	P > 0,05

MIC: minimum inhibitory concentration ; MBC: minimum bactericidal concentration.

3.2 Phytochemical Screening

The results of the phytochemical screening revealed the presence of eight groups of phytochemical compounds in the three extracts. These are saponins, anthraquinone glycosides, polyterpenes, polyphenols, catechin and gallic tannins, anthocyanins, flavonoids, and anthocyanins (Table 5).

Table 5: Chemical compounds identified in the different plant extracts.

Plant extracts	Chemical compounds detected
Aqueous extract	Catechetical and gallic tannins, polyphenols, flavonoids.
Acetone extract	Saponins, gallic tannins, polyphenols, polyterpenes, anthocyanins, flavonoids.
Ethanol extract	Catecholic and gallic tannins, anthraquinone glycosides, polyphenols, Polyterpenes, anthocyanins, flavonoids.

4. DISCUSSION

The antibacterial activity of *Combretum collinum* stem bark extracts was evaluated on the *in vitro* growth of eight *Staphylococcus aureus* bacterial strains, including one reference strain, five multidrug-resistant strains, and two susceptible strains. The tests revealed varying activities of the extracts against all bacterial strains. This was reflected in the differences observed in the mean inhibition zone diameters. The highest values (22.0 ± 0.1 mm, 21.7 ± 0.6 mm, and 17.2 ± 4.3 mm) were obtained with the ethanolic, aqueous, and acetone extracts, respectively, on the *S. aureus* 48c strain at a concentration of 50 mg/ml. This bacterium, exhibiting a resistant phenotype, was isolated from the urine of a patient with a urinary tract infection. This could therefore justify the traditional use of *Combretum collinum* in the treatment of ear and urinary tract infections.^[20] This antibacterial activity against all germs could provide a scientific basis for the widespread traditional use of this plant in many African countries, such as Kenya and Uganda, in the treatment of several pathologies, some of which are of bacterial origin.^[21]

These results corroborate those of,^[22] who, in their work in Uganda, indicated the antibacterial potential of this plant against Gram-positive bacteria, including methicillin-resistant *Staphylococcus aureus* (MRSA), responsible for diarrhea. Similar large inhibition zone diameters were obtained against bacterial strains collected from patients suffering from diarrhea, sinusitis, otitis, fever, and meningitis in infants. This plant could be recommended for the treatment of these conditions that may be caused by *S. aureus*. In general, the results obtained with the ethanolic and aqueous extracts are almost identical. This indicates a lack of selectivity towards bacterial strains and the solubility of the active ingredients in both types of solvent.

The antibacterial parameters, namely the MIC, MBC, and MBC/MIC ratio, were determined using *C. collinum* extracts. The lowest values at which the extracts exerted inhibitory activity were 0.60 mg/ml for both types of extract. These values were obtained for *S. aureus* 10y and 48c with the ethanolic extract and for *S. aureus* 48c with the aqueous extract. Therefore, it can be concluded that the extracts were more effective against these strains. These results are similar to those of.^[7] Indeed, these authors demonstrated that *C. collinum* extracts have bactericidal activity against a wide range of bacteria, including *S. aureus*, which is responsible for opportunistic infections.

The statistical analysis indicated that the observed differences in MIC and MBC values between the two strain categories were not significant for either type of extract. This suggests that the activity of the extracts was not statistically influenced by bacterial phenotypes. In other words, while the values obtained with the two types of extracts differ, they produce similar effects against the bacteria. This further justifies the non-selectivity of the extracts' action

against the bacteria. Moreover, the aqueous extract was bactericidal against all bacterial strains tested in this study, as its MBC/MIC ratios were all less than 4.^[14]

A plant identified as effective against a microbial infection is often prescribed for the treatment of several other common ailments. It therefore has multiple uses, explained by the presence of numerous groups of biologically active chemical compounds that chemical screening reveals in its organs.^[6] Indeed, phytochemical investigations have identified eight major groups of chemical compounds, including saponins, anthraquinone glycosides, polyterpenes, polyphenols, catechins and gallic tannins, flavonoids, and anthocyanins. These phytoconstituents are certainly responsible for the pharmacological effects observed with the use of this plant and, consequently, determine its therapeutic value. Among these compounds, many are recognized for their antibacterial effects.

The flavonoids present in this plant species are secondary metabolites with significant antibacterial activity. Indeed, due to their high phenolic content, these flavonoid compounds can bind to certain proteins and enzymes, thus altering enzymatic balances.^[23] These constituents are certainly involved in the observed antibacterial activity. Their presence in *C. collinum* could also explain some traditional therapeutic uses of this plant, particularly in the treatment of epilepsy, as flavonoids play a crucial role in maintaining neuronal function.^[24] Similarly, these phytoconstituents could be involved in relieving fever, managing generalized malaise, and treating urogenital disorders traditionally addressed with *C. collinum*.

Saponins are also known for their anti-inflammatory, anti-edematous, analgesic, and antibacterial properties.^[25] The presence of these compounds in *C. collinum* could explain its use in treating stomach aches, fever, general malaise, gonorrhea, and erectile dysfunction. Indeed, these various ailments can be the result of an inflammatory process caused by bacteria.^[26]

Tannins are known for their antiseptic, bactericidal, and astringent properties.^[27] Their presence could also be responsible for this bacterial activity.^[28] Furthermore, these compounds, due to their healing and coagulating properties, could justify the use of *C. collinum* in postpartum treatment.^[29]

Furthermore, *C. collinum*, like many plant species, is being overexploited by local populations for its numerous therapeutic properties. Yet, it plays a crucial role in the functioning of ecosystems in tropical and subtropical regions. For this reason, and many others, it must be included among the cultivated and protected plants in Côte d'Ivoire, or risk disappearing in the near future.

5. CONCLUSION

The work carried out confirmed the antibacterial activity of aqueous, acetone, and ethanolic extracts of *C. collinum* stem bark against *Staphylococcus aureus* strains of different phenotypes. The determination of the MICs and MBCs revealed a bactericidal effect of the aqueous extract on all bacterial strains. Furthermore, the major groups of chemical compounds, likely responsible for this activity, were identified and are more concentrated in the ethanolic and aqueous solvents. It is therefore conceivable to further ethnopharmacological studies, confirm the results obtained *in vitro* with *in vivo* tests, and conduct toxicity tests on *C. collinum*, which could offer hope for alleviating microbial infections, a true public health threat. The range of protections for endangered species must be broadened to guarantee the preservation and safeguarding of biodiversity, a true treasure of Ivorian flora.

ACKNOWLEDGMENTS

We express our deep gratitude to the Departments of Bacteriology and Virology of the Pasteur Institute of Côte d'Ivoire (IPCI) and the Swiss Center for Scientific Research of Côte d'Ivoire (CSRS) for their material and technical support.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest regarding the publication of this article.

CONTRIBUTES OF THE AUTHORS

This work was carried out in collaboration with all the authors mentioned. They participated in all stages of the manuscript's development, up to and including its final version. They therefore read and approved the final manuscript.

REFERENCES

1. Ben FR, Bougerra CH, Sahnoun O, Loussaief CH, Kacem B, Mastouri M, Tabka-Stambouli R, Chakroun M, Bouzouaia N. Multidrug-resistant bacteria isolated from patients hospitalized in an infectious diseases ward. *Tunisian Journal of Infectious Diseases*, 2007; 1(4): 12-15.
2. Guessenn NK, Gbonon VC, Tiékoura KB, Kakou-N'douba A, Ouattara DN, Boni-Cissé C, Dosso M, le GER-BMR, Evolution of bacterial resistance to imipenem in Côte d'Ivoire from 2005 to 2009, Scientific Symposium of the Pasteur Institute of Côte d'Ivoire: emerging pathogens and integrative biology, 2009; 17.
3. INSTITUT PASTEUR, The challenge of infectious diseases. Last accessed March 8, 2014. <http://www.pasteur.fr/ip/easysite/pasteur/fr/presse/dossiers-de-presse/sante-en-voyage/le-defi-des-maladies-infectieuses>.
4. Gangoue PJ. Characterization of Beta-lactamases and their inhibition by medicinal plant extracts, Doctoral thesis in Biochemistry. University of Liège, Belgium, 2007; 104.
5. Waridel P. Phytochemical investigation of aquatic plants *Potamogeton pectinatus* L., *P. lucens* L., *P. perfoliatus* L. and *P. crispus* L. (Potamogetonaceae). University of Lausanne, Switzerland, 2003; 219.
6. Tra Bi FH. Evaluation of the antifungal activity of fifteen (15) plants from the flora of Côte d'Ivoire. Doctoral thesis, University of Abobo-Adjamé, Abidjan, Côte d'Ivoire, 2008; 122.
7. Bamba M, Christel N, Simon B, Soro D, Jules KN, Sanogo Y, Jennifer S, Alexis Z, Tra Bi FH, Sevser S. Phytochemical screening of methanolic extracts from *Combretum collinum* leaves and *Anogeisus leiocarpus* roots and in vitro antibacterial effect on multidrug-resistant *Staphylococcus aureus* strains. *Int. J. Biol. Chem. Sci*, 2020; 14(6): 2362-2372.
8. WHO. Coverage of selected prevention and care services in developing countries in 2001. WHO, Geneva (Switzerland), 2003; 48.
9. Zirihi G, Kra A. Evaluation of the antifungal activity of *Microglossa pirifolia* (LARMARCK) O. KUNTZE (Asteraceae) "PYMI" on the in vitro growth of *Candida albicans*. *Review of African Medicines and Pharmacopoeias*, 2003; 17, 11-19.
10. Koné WM, Kamanzi AK, Terreux C, Hostettmann K, Traore D, Dosso M. Traditional medicine in North Côte d'Ivoire: screening of 50 medicinal plants for antibacterial activity. *Journal of Ethnopharmacology*, 2004; 93: 43-49.
11. Ponce AG, Fritz R, Del Vallec, Rouras I. Antimicrobial activity of essential oils on the native microflora of organic Swiss chard. *Society of Food Science and technology (Elsevier)*, 2003 ; 36(7): 679-684.

12. Kouadio NJ, Guessennd NK, Koné MW, Moussa B, Koffi YM, Guédé KB, Yao K, Bakayoko A, Tra-Bi FH, Dosso M. Evaluation of the activity of *Mallotus oppositifolius* (Geisel.) Müll. Arg. (Euphorbiaceae) leaves against multidrug-resistant bacteria and phytochemical screening. International Journal of Biological and Chemical Sciences, 2015; 9(3): 1252-1262.
13. Fauchere IL, Avril JL. General and Medical Bacteriology. Ellipses Publishers. Paris 1, 2002; 122.
14. Kamanzi AK, Medicinal plants of Côte d'Ivoire: Phytochemical investigations guided by biological tests. Doctoral thesis, University of Cocody, Abidjan, 2002; 176.
15. N'gaman KCC, Békro YA, Mamyrbékova-Békro JA, Béné A, Gooré BS. Secondary metabolite composition and antioxidant activity of crude extracts of *Gmelina arborea* Roxb. (Verbanaceae) from Ivory Coast, West Africa: Analysis by Thin Layer Chromatography. Eur. J. Sc. Res., 2009; 36(2): 161-171.
16. Lhuillier A. Contribution to the phytochemical study of four Malagasy plants: *Agauria salicifolia* Hook. f ex Oliver, *Agauria polyphylla* Baker (Ericaceae), *Tambourissa trichophylla* Baker (Monimiaceae) and *Embelia concinna* Baker (Myrsinaceae), National Polytechnic Institute of Toulouse, France, 2007; 214.
17. Mallikharjuna PB, Rajanna LN, Seetharam YN, Sharanabasappa GK. Phytochemical studies of *Strychnos potatorum* L. F. a medical plant. Journal of Chemistry, 2007; 4(4): 510-223.
18. Dohou N, Yamni K, Tahrouch S, Idrissi Hassani LM, Badoc A, Gmira N. Phytochemical screening of an Ibero-Moroccan endemic, *Thymelaea lithroides*. Bulletin of the Bordeaux Pharmaceutical Society. 2003; 142(1-4): 61-78.
19. R Core Team, R: A Language and environment for statistical computing. R fondation for statistical computing, Vienna, Austria, 2013. <http://www.r-project.org>, Accessed May 8, 2013.
20. Njoroge GN & Bussmann RW. Traditional management of ear, nose and throat (ENT) diseases in central Kenya. J. of Ethnobiol. and Ethnomed, 2006; 2: 54-56.
21. Bethwell OO, Kisangau DP. Kenyan medicinal plants used as antivenin: a comparison of plant usage. J. Ethnobiol. & Ethnomed, 2006; 2: 7-15.
22. Tabuti JRS, Lye KA & Dhillon SS. Traditional herbal drugs of Bulamogi, Uganda: plants, use and administration. J. of Ethnopharm., 2003; 88: 19-44.
23. Tsague MV and Nyemb N. Antibacterial properties and phytochemical composition of *Ficus iteophylla* Linn leaves, International Journal of Chemical Research and Development, 2022; 4(2): 37-38.
24. Muazu J, Kaita AH. A review of traditional plants used in the treatment of epilepsy amongst the Hausa/Fulani tribes of northern Nigeria. Afr. J. Trad. CAM, 2008; 5(4): 387 - 390.
25. Brunet J. Pharmacognosie, phytochimie, plantes médicinales, 3ème édition Tec & doc, Paris (France), 1999; 1560.
26. Silbernag LS & Lang F., 2000. Atlas of Pathophysiology. Flammarion Medicine-Sciences, Paris, France, 2000; 406.
27. Hoffman D. Medical Herbalism: The science and practice of herbal medicine. Inner Traditions bear and Company, Rochester, Vermont (USA), 2003; 83.
28. Denise PSL, Polizello ACMP, Ysabelle YI, Augusto CCS. Antibacterial screening of anthocyanic and proanthocyanic fractions from cranberry juice. J. Med. Food., 2005; 8(1): 36-40.

29. Johanna FP. Traditional medicinal uses and biological activities of some plant extracts of African *Combretum* Loeft., *Terminalia* L. and *Pteleopsis* Engl. Species (Combretaceae). Atlas of Pathophysiology. Flammarion Medicine-Sciences, Paris, France, 2007; 184.