Investigational
Medicinal Chemistry & Pharmacology

Research Article Open Access

Synergistic antibacterial activity of *Erigeron floribundus* extracts combined with erythromycin against multidrug-resistant bacteria

Vincent Ngouana[1]*, Mado Solange Ngo Yane[1], Patrick Valere Tsouh Fokou[2,5]**, Germaine Matsuete Takongmo[3], Boniface Pone Kamdem[4,5], Jean Polidor Nguetsa Demafo[1], Prisca Laure Nobosse Pewoupa[1], Jean-De-Dieu Tamokou[3]

Abstract

Background: This research assessed the in vitro antibacterial activity of *Erigeron floribundus* extracts against multidrug-resistant bacterial strains and explored their potential to enhance the effectiveness of standard antibiotics through synergistic combinations.

Methods: Leaf, stem, and root crude extracts of *E. floribundus* were obtained using methanol and ethyl acetate. Major phytochemical groups (total phenols, flavonoids, tannins, saponins) were quantified. The antimicrobial activity of the extracts alone and in combination with amoxicillin, levofloxacin, and erythromycin was evaluated against laboratory strains of *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa* using the broth microdilution assay. Minimum inhibitory concentrations (MICs) and minimum bactericidal concentrations (MBCs) were determined. Synergy was quantified using the fractional inhibitory concentration (FIC) index.

Results: The methanol leaf extract contained the highest levels of phenolic and flavonoid compounds and exhibited the strongest intrinsic antibacterial activity. When combined with erythromycin, this extract demonstrated potent synergy against all tested bacterial strains. Combinations with amoxicillin and levofloxacin produced additive or indifferent effects depending on the bacterial species. The observed synergy indicates that the extract potentiates erythromycin's antibacterial mechanism.

Conclusion: The methanol leaf extract of *E. floribundus* possesses considerable antibacterial properties and, more importantly, exhibits universal synergistic interaction with erythromycin against multidrug-resistant bacteria. This finding corroborates the ethnomedical application of the plant and highlights its potential as a source of resistance-modifying compounds. Such an approach could help "rescue" existing antibiotics and provide a new strategy against antibiotic resistance.

Keywords: Antibiotic synergy; combination therapy; *Erigeron floribundus*; multidrug resistance; phytochemicals; resistance-modifying agent.

*Correspondence: *Tel.: +237 67379910; E-mail address: ngouanavincent@yahoo.fr; ORCID: https://orcid.org/0000-0002-1390-6050 (Vincent Ngouana); **Tel.: +237 676620503; E mail address: ptsouh@gmail.com; ORCID: https://orcid.org/0000-0003-3724-3527 (Patrick Valere Tsouh Fokou).*

[1] Centre de Recherche en Santé Humaine et Développement de Médicaments (CRESHDEM), Department of Pharmaceutical Sciences, Faculty of Medicine and Pharmaceutical Sciences, University of Dschang, Dschang, Cameroon; [2] Department of Biochemistry, Faculty of Science, University of Bamenda, Bambili, Cameroon; [3] Department of Biochemistry, Faculty of Science, University of Dschang, Dschang, Cameroon; [4] Department of Biochemistry, Faculty of Science, University of Yaounde 1, Yaounde, Cameroon; [5] Advanced Research & Health Innovation Hub, Yaounde, Cameroon.

Other authors
Email: ngoyanemadosolange@gmail.com (Mado Solange Ngo Yane); Email: germainetakongmo@gmail.com (Germaine Matsuete Takongmo); Email: ponekamdemboniface@gmail.com, ORCID: https://orcid.org/0000-0002-4982-0507 (Boniface Pone Kamdem); Email: polidornguetsa@gmail.com (Jean Polidor Nguetsa Demafo); Email: pnobose@gmail.com (Prisca Laure Nobosse Pewoupa); Email: jtamokou@yahoo.fr, ORCID: https://orcid.org/0000-0002-8088-463X (Jean De Dieu Tamokou).

Citation on this article: Ngouana V, Ngo Yane MS, Tsouh Fokou PV, Takongmo GM, Kamdem BP, Demafo JPN, Pewoupa PLN, Tamokou JDD. Synergistic antibacterial activity of Erigeron floribundus extracts combined with erythromycin against multidrug-resistant bacteria. Investigational Medicinal Chemistry and Pharmacology (2026) 9(1):131; Doi: https://dx.doi.org/10.31183/imcp.2026.00131

Background

The antibiotic-resistant bacteria are continually increasing, becoming a major threat to the public health and medicine field all over the world [1]. In fact, infections due to multidrug-resistant (MDR) pathogens not only cause prolonged illness and higher death rates but also bring about enormous healthcare costs, thus reversing decades of medical progress [2]. The main reasons for this have been the excessive use of antibiotics both in humans and animals and the almost complete stop in the development of new antibacterial drugs [3]. As a result, there an urgent global demand for new ways of therapy against resistant infections and for the preservation of the effectiveness of our current antibiotic arsenals [4]. A promising strategy is combination therapy in which standard antibiotics are combined with bioactive compounds from natural sources such as herbal medicines [5, 6]. These substances extracted from plants can serve as resistance-modifying agents (RMAs). They may counter antibiotic resistance mechanisms by preventing bacterial efflux pumps, damaging cell membranes, or capturing resistance enzymes; thus, bacteria will be susceptible again to the drugs they have learned to evade [7, 8]. This kind of combination not only brings the advantage of lower antibiotic dose when treatment is enhanced which in turn reduces side effects but also it helps in slowing the development of newer resistance [9]. *Erigeron floribundus* (H.B.K) Sch. Bip. Compon (Asteraceae) is a medicinal plant whose use dates back centuries and plays a crucial role in the traditional medicine of several African areas including Cameroon and Côte d'Ivoire [10, 11]. It is frequently employed to treat bacterial infections such as dental abscesses, skin disorders, and abdominal complaints. This is a strong indication that the plant has inherent antimicrobial properties [12]. Studies on the phytochemical profiles of this plant have unveiled that it harbors bioactive secondary metabolites such as phenols, flavonoids, and tannins which are usually associated with antimicrobial activity [13, 14]. To date, ant-inflammatory, pain relief, and antifungal properties of the plant have been described. However, little is known about its exact antibacterial action, especially against today's main clinically relevant MDR bacterial strains, and whether it could work in synergy with conventional antibiotics [15, 16]. Therefore, this investigation focused on the antibacterial and antibiotic-potentiating proficiency of *E. floribundus*. Specifically, the study aimed to assess the *in vitro* antibacterial activity of crude extracts derived from the different organs of *E. floribundus* against a panel of MDR bacteria and to determine the outcome of their combination with standard antibiotics (amoxicillin, levofloxacin, and erythromycin). By thoroughly elucidating this plant–antibiotic synergy, our work seeks to validate the traditional use of *E. floribundus* and provide a scientific, plant-based strategy for combating antimicrobial resistance globally, potentially reviving the efficacy of present-day antibiotics.

Methods

Plant material, authentication, and extraction

Wild *Erigeron floribundus* (H.B.K) Sch. Bip. Compon plants were collected from the West Region of Cameroon. Identification and authentication of the plant were done at the National Herbarium of Yaoundé, where the voucher specimen was deposited under reference number 7750/HNC. The leaves, stems, and roots were harvested, air-dried at room temperature (20-25°C) in the shade for one week, and then pulverized into powder with a mechanical grinder.

Each powdered plant part (100 g) was separately subjected to extraction by cold maceration in 500 mL of solvent (1:5 w/v ratio) for 48 hours with intermittent shaking every 12 hours. Two different solvent systems were applied: methanol (99.8% purity, Sigma-Aldrich) and ethyl acetate (99.5% purity, Sigma-Aldrich). The mixtures were filtered through Whatman No.1 filter paper. The filtrates were then concentrated under reduced pressure at 65°C (methanol) and 78°C (ethyl acetate) using a rotary evaporator (Heidolph). The crude extracts obtained were placed into pre-weighed vials and were oven-dried at 40°C to a constant weight. The extraction yield was calculated using the equation below:

$$\text{Yield (\%)} = \frac{\text{Weight of dry extract (g)}}{\text{Weight of dry plant powder (g)}} \times 100$$

Phytochemical Analysis (Quantitative Determination)

The dried crude extracts were analyzed quantitatively for the major groups of secondary metabolites.

Total Phenolic Content (TPC) was evaluated by using the Folin-Ciocalteu colorimetric method according to the description of Ramde, Tiendrebeogo et al. [17]. In short, 20 µL of the extract solution (2 mg/mL) was combined with 100 µL of diluted Folin-Ciocalteu reagent and 80 µL of sodium carbonate solution (20% w/v). The mixture was then incubated at 45°C for 30 min, and the absorbance was measured at 765 nm. The calibration curve was generated using gallic acid as standard, with the linear regression equation y = 0.0375x (R^2 = 0.9988). The results were expressed as milligrams of Gallic Acid Equivalent per gram of dry extract (mg GAE/g).

Total Flavonoid Content (TFC) was determined by means of the aluminum chloride colorimetric method [18]. 100 µL of the extract (2 mg/mL) was mixed with 100 µL of a solution containing 50 µL of $AlCl_3$ (1.2% w/v) and 50 µL of potassium acetate (120 mM). After the mixture was left to incubate 30 min at room temperature, the absorbance was read at 415 nm. The calibration curve was generated using quercetin as standard, with the linear regression equation y = 0.0298x + 0.1024 (R^2 = 0.9952). The content was reported in terms of milligrams of Quercetin Equivalent per gram of extract (mg QE/g).

Total Tannin Content (TTC) was measured by the method of Singleton and Rossi [19]. 100 µL of the extract (2 mg/mL) was added to a mixture of 500 µL of Folin-Ciocalteu reagent and 1 mL of sodium carbonate (35%). The solution was diluted, kept for 30 min, and the absorbance was read at 700 nm. The calibration curve was generated using tannic acid as standard, with the linear regression equation y = 0.0005x + 0.0081 (R^2 = 0.9904). Tannic acid was the standard, and the results were expressed as mg Tannic Acid Equivalent per gram of extract (mg TAE/g).

Total Saponin Content (TSC) was determined using the foam index (afrosimetric) method of Koziol [20]. Briefly, 0.5 g of an extract was mixed with 5 mL of distilled water in a graduated test tube and shaken vigorously for 30 seconds. After 10 seconds, the height of the stable foam layer was measured. The saponin content was calculated using the following formula and expressed in mg per 100 g of sample (mg/100g).

$$\text{Saponins (mg/100g sample)} = \frac{(0.432 \times \text{foam height in cm}) + 0.008}{\text{sample mass (g)}}$$

Bacterial strains and culture conditions

The work was done with four bacterial strains, namely *Staphylococcus aureus* ATCC 29213, *Escherichia coli* ATCC 25922, *Klebsiella pneumoniae* 1 (a clinical isolate), and *Pseudomonas aeruginosa* 2 (a clinical isolate). The strains were streaked on Mueller-Hinton Agar (MHA) slants and stored at 4°C. Before each test, they were sub-cultured on fresh MHA plates and incubated for 24 hours. The bacterial inocula were prepared by picking colonies from the plate, suspending them in sterile saline solution (0.85% NaCl), and then adjusting the turbidity to a 0.5 McFarland standard (about 1.5 × 10^8 CFU/mL). The suspension was finally diluted 1:100 in Mueller-Hinton Broth (MHB) to obtain a working inoculum of approximately 1.5 × 10^6 CFU/mL [21].

Antibiotics and chemicals

Reference antibiotics were purchased as pure powders: amoxicillin (β-lactam), levofloxacin (fluoroquinolone), and erythromycin (macrolide). Stock solutions (8192 µg/mL) were created in sterile distilled water or dimethyl sulfoxide (DMSO, ≤ 1% final concentration) according to the requirement. Folin-Ciocalteu reagent, gallic acid, quercetin, tannic acid, $AlCl_3$, and all solvents were of analytical grade. Culture media (MHA and MHB) were obtained from HiMedia Laboratories.

Determination of minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC)

The antibacterial activity of the plant extracts and antibiotics was tested by using the broth microdilution method in 96-well microplates according to the guidelines of the Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS) with slight changes [22]. Briefly, two-fold serial dilutions of the test agents were prepared in MHB to obtain a concentration range of 0.25 to 2048 µg/mL. Each well was filled with 100 µL of the diluted agent, 100 µL of MHB, and 100 µL of the standardized inoculum (approximately 1.5 × 10^6 CFU/mL). Wells with only MHB and inoculum acted as growth controls, whereas wells with MHB, DMSO (at the highest concentration used, ≤1%), and inoculum acted as solvent controls. The plates were incubated at 37°C for 24 hours. The MIC was defined as the minimum concentration showing no visible growth. For MBC determination, 10 µL from each well not showing visible growth in the MIC test was sub-cultured on new MHA plates which were further incubated for 24 hours. The MBC was considered the minimum concentration that killed ≥99.9% of the initial inoculum (no colony growth on subculture). Each assay was done in triplicate (n = 3).

Evaluation of extract-antibiotic combination effects

The combined effect of the drugs was analyzed using the checkerboard microdilution method [23]. The most potent plant extract (methanol leaf extract) was used in combination with each antibiotic. Serial dilutions of the extract and the antibiotic were mixed in a two-dimensional checkerboard arrangement in the microplate. The final concentrations ranged from 1/8× to 2× the predetermined MIC of each agent. The plates were inoculated and incubated as previously described. All tests were repeated three times. The degree of interaction was measured by determining the Fractional Inhibitory Concentration (FIC) index:

$$FIC_{extract} = \frac{MIC_{extract\ in\ combination}}{MIC_{extract\ alone}}$$

$$FIC_{antibiotic} = \frac{MIC_{antibiotic\ in\ combination}}{MIC_{antibiotic\ alone}}$$

$$\sum FIC = FIC_{extract} + FIC_{antibiotic}$$

The interaction was interpreted as follows: synergy (∑FIC ≤ 0.5), additivity (0.5 < ∑FIC ≤ 1), indifference (1 < ∑FIC ≤ 4), or antagonism (∑FIC > 4) [5].

Statistical analysis

Quantitative phytochemical data and MIC values are presented as mean ± standard deviation (SD). Data were analyzed by one-way analysis of variance (ANOVA) using SPSS software (Version 22.0). The differences between means were compared by Waller-Duncan's post-hoc test at a significance level of $p < 0.05$.

Results

Extraction yields and phytochemical profile

The extraction yields of the six crude extracts were in the range of 1.36% to 2.59% (w/w), with the methanol root extract giving the highest yield (Table 1). The quantitative phytochemical analysis showed that the extracts contained widely different amounts of various compounds (Table 2). The methanol leaf extract had the greatest total phenolic content (TPC: 282.21 ± 0.17 mg GAE/g) and total flavonoid content (TFC: 145.47 ± 0.33 mg QE/g). The ethyl acetate stem extract recorded the highest tannin content (14.12 ± 0.76 mg TAE/g).

Antibacterial activity of extracts and antibiotics alone

The extracts possessed some degree of intrinsic antibacterial activity as indicated by the MICs ranging from 256 to >2048 µg/mL and MBCs from 256 to >2048 µg/mL (Table 3). The methanol leaf extract showed MIC values of 128 µg/mL, 256 µg/mL, 256 µg/mL and 1024 µg/mL against *S. aureus*, *E. coli*, *P. aeruginosa*, and *K. pneumoniae*, respectively. Based on its MBC/MIC ratios, the extract exhibited a bactericidal effect (MBC/MIC ≤ 4) against *S. aureus* and *P. aeruginosa* and a bacteriostatic effect (MBC/MIC > 4) against *E. coli* and *K. pneumoniae*. Among the reference antibiotics, levofloxacin showed the lowest MICs (range: 0.25–128 µg/mL) and was bactericidal against all susceptible strains. Erythromycin had a moderate level of activity (MIC range: 2-128 µg/mL), whereas amoxicillin was ineffective against *K. pneumoniae* (MIC >256 µg/mL).

Potentiation of antibiotic activity by methanol leaf extract

The MIC for each antibiotic was assessed with sub-inhibitory concentrations (1/2, 1/4 and 1/8 MIC) of the methanol leaf extract. Table 4 presents the MIC values of each antibiotic when combined with the extract. The greatest degree of potentiation was with erythromycin, the MIC being lowered 4-fold (to 0.5 µg/mL) for *S. aureus*, 16-fold (to 1 µg/mL) for *E. coli*, and 32-fold (to 0.5 µg/mL) for *P. aeruginosa* when 1/2 MIC of the extract was used. Additionally, the combination of the two agents made amoxicillin effective against an otherwise resistant *K. pneumoniae*, as the MIC was reduced from >256 µg/mL to 64 µg/mL.

Synergy Assessment via Fractional Inhibitory Concentration (FIC) Index

The checkerboard method and FIC index were used to measure the interaction between the methanol leaf extract and antibiotics. With erythromycin, the combination was synergistic (∑FIC ≤ 0.5) against *S. aureus*, *E. coli*, and *P. aeruginosa*, and additive (∑FIC = 0.75) against *K. pneumoniae*. With amoxicillin, the combination was synergistic against *K. pneumoniae* (∑FIC = 0.5) and *P. aeruginosa* (∑FIC = 0.375), additive against *E. coli* (∑FIC = 0.5625), and indifferent against *S. aureus* (∑FIC = 1.125). With levofloxacin, the combination was synergistic against *S. aureus* (∑FIC = 0.3125) and *E. coli* (∑FIC = 0.1875), additive against *K. pneumoniae* (∑FIC = 0.75), and indifferent against *P. aeruginosa* (∑FIC = 1.125).

Discussion

This study reveals that *Erigeron floribundus* leaves exhibit the strongest antibacterial activity, which correlates with the high phenolic and flavonoid content of the methanol leaf extract (Table 2). It is noteworthy that for the same plant part, methanol extracts generally had higher phenolic and flavonoid contents than their ethyl acetate counterparts, reflecting the greater extraction efficiency of polar solvents for these phytochemicals. Phenolic and flavonoid compounds are well-recognized antimicrobial agents that exert their effects through multiple mechanisms, including disruption of cell membranes, inhibition of bacterial enzymes, and interference with microbial toxin production [24, 25]. The bactericidal activity (MBC/MIC ≤ 4) observed against *S. aureus* and *P. aeruginosa* suggests a mechanism beyond simple growth inhibition, supporting the therapeutic potential of this extract. These findings provide a phytochemical rationale for the traditional use of *E. floribundus* leaves as remedies for bacterial infections [12].

The intrinsic MIC values of the extract (128–1024 µg/mL) are modest compared to pure antibiotics but remain within the range considered significantly active for crude plant extracts [26]. More importantly, the primary value of this extract lies not in direct antimicrobial action but in its ability to potentiate existing antibiotics against resistant bacteria.

The results demonstrate that sub-inhibitory concentrations of the methanol leaf extract substantially enhance the activity of conventional antibiotics (Table 4). Notably, the extract reduced the MIC of erythromycin against *P. aeruginosa* by 32-fold and restored susceptibility of *K. pneumoniae* to amoxicillin, effectively reversing resistance that was present prior to combination treatment.

A key finding of this study is the consistent synergistic interaction between the methanol leaf extract and erythromycin across all tested Gram-positive and Gram-negative MDR strains, with ∑FIC values ≤0.375 for three of four strains (Table 5). Erythromycin, a macrolide antibiotic, frequently fails due to resistance mechanisms such as efflux pumps (e.g., Mef, Msr) and ribosomal modification [27]. We hypothesize that the high concentration of flavonoids and phenolics in the extract (145.47 mg QE/g and 282.21 mg GAE/g, respectively) acts as resistance-modifying agents (RMAs). Specifically, these phytochemicals may competitively inhibit macrolide-specific efflux pumps or increase bacterial membrane permeability, thereby allowing erythromycin to accumulate intracellularly and reach its ribosomal target at lower doses [7, 8]. This hypothesis is supported by the bactericidal activity observed against some strains, suggesting that the extract targets the cell envelope, which could facilitate antibiotic entry.

In contrast to the universal synergy with erythromycin, the combination outcomes with amoxicillin and levofloxacin were variable, ranging from synergistic to indifferent depending on the bacterial species. This variability is consistent with the distinct resistance mechanisms associated with each antibiotic class. β-Lactam resistance (e.g., amoxicillin) is primarily mediated by β-lactamase enzymes, while fluoroquinolone resistance (e.g., levofloxacin) often involves mutations in DNA gyrase or topoisomerase IV [28]. The phytochemical profile of the extract appears to be particularly effective against macrolide-specific resistance (e.g., efflux pumps) but may not directly inhibit β-lactamases or alter fluoroquinolone target sites. However, the synergistic effect observed with amoxicillin against *K. pneumoniae* and *P. aeruginosa* suggests an additional, non-specific mechanism, such as generalized membrane disruption that enhances antibiotic penetration. Further research is required to elucidate this mechanism.

This study has several limitations that should be acknowledged. First, the research was conducted exclusively *in vitro*; the observed synergistic effects require validation *in vivo* to assess pharmacokinetic compatibility, safety, and efficacy. Second, the active compounds responsible for the synergistic effect have not been identified. Bioassay-guided fractionation is needed to isolate and characterize the specific flavonoids or phenolics that mediate erythromycin potentiation. Third, no cytotoxicity evaluation was performed on mammalian cells, which is essential to determine the therapeutic window of the extract-antibiotic combination. Fourth, the plant material was collected from the wild, which may introduce batch-to-batch variability in phytochemical composition; chemical standardization will be necessary for future development. Finally, the clinical isolate of *K. pneumoniae* was not genotyped; whole-genome sequencing would help correlate synergy with specific resistance genes. Despite these limitations, the robust and consistent synergy observed with erythromycin provides a strong foundation for further investigation.

Table 1. Extraction yields of *Erigeron floribundus* crude extracts.

Plant Part	Solvent	Yield (%)
Leaves	Methanol	1.51
Leaves	Ethyl Acetate	1.36
Stems	Methanol	1.81
Stems	Ethyl Acetate	1.68
Roots	Methanol	2.59
Roots	Ethyl Acetate	1.78

Table 2. Quantitative phytochemical composition of *Erigeron floribundus* extracts (mean ± SD).

Extract	TPC (mg GAE/g)	TFC (mg QE/g)	Tannins (mg TAE/g)	Saponins*
Methanol Leaves	282.21 ± 0.17	145.47 ± 0.33	6.48 ± 0.41	1.11 ± 0.03
Ethyl Acetate Leaves	187.21 ± 0.76	123.06 ± 0.79	5.94 ± 0.55	1.02 ± 0.55
Methanol Stems	109.30 ± 0.11	89.19 ± 0.17	8.33 ± 0.22	0.822 ± 0.06
Ethyl Acetate Stems	173.70 ± 0.33	151.70 ± 0.22	14.12 ± 0.76	0.562 ± 0.02
Methanol Roots	109.90 ± 0.22	87.72 ± 0.68	7.63 ± 0.17	0.476 ± 0.14
Ethyl Acetate Roots	136.44 ± 0.17	88.62 ± 0.52	5.09 ± 0.11	0.303 ± 0.05

*Saponin content expressed as foam height index (cm/g). See Methods. TPC: Total Phenolic Content; TFC: Total Flavonoid Content; GAE: Gallic Acid Equivalent; QE: Quercetin Equivalent; TAE: Tannic Acid Equivalent.

Table 3. Minimum inhibitory concentrations (MIC) and minimum bactericidal concentrations (MBC, µg/mL) of *Erigeron floribundus* extracts and reference antibiotics against multidrug-resistant strains.

Agent	*S. aureus* ATCC 29213		*E. coli* ATCC 25922		*K. pneumoniae* 1		*P. aeruginosa* 2	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
Methanol Leaves	128	256	256	1024	1024	2048	256	512
Ethyl Acetate Leaves	512	1024	512	1024	2048	ND	1024	ND
Methanol Stems	1024	ND	1024	1024	1024	ND	512	1024
Ethyl Acetate Stems	1024	ND	1024	2048	1024	ND	512	2048
Methanol Roots	1024	1024	512	512	1024	ND	512	1024
Ethyl Acetate Roots	1024	ND	>2048	ND	>2048	ND	2048	ND
Amoxicillin	0.25	2	1	4	>256	ND	128	ND
Levofloxacin	8	32	4	8	128	ND	0.25	2
Erythromycin	2	8	16	32	128	256	16	32

ND: Not Determined (MBC > 2048 µg/mL or no growth inhibition at the highest tested concentration).

Table 4. Minimum inhibitory concentrations (MIC, µg/mL) of antibiotics in the presence of sub-inhibitory concentrations of *Erigeron floribundus* Methanol Leaf Extract (FM).

Antibiotic	Strain	MIC Alone	MIC with 1/2 FM	MIC with 1/4 FM	MIC with 1/8 FM
Erythromycin	*S. aureus* ATCC 29213	2	0.5	0.5	1
	E. coli ATCC 25922	16	1	0.5	2
	K. pneumoniae 1	128	32	64	128
	P. aeruginosa 2	16	0.5	1	1
Levofloxacin	*S. aureus* ATCC 29213	8	2	2	4
	E. coli ATCC 25922	4	0.5	0.5	1
	K. pneumoniae 1	128	64	64	128
	P. aeruginosa 2	0.25	0.25	1	1
Amoxicillin	*S. aureus* ATCC 29213	0.25	0.25	1	1
	E. coli ATCC 25922	1	0.5	2	1
	K. pneumoniae 1	>256	64	128	128

Table 5. Fractional inhibitory concentration (FIC) indices for the combination of *Erigeron floribundus* methanol leaf extract with antibiotics.

Antibiotic / Strain	∑FIC Index	Interpretation
With Amoxicillin		
S. aureus ATCC 29213	1.125	Indifference
E. coli ATCC 25922	0.5625	Additive
K. pneumoniae 1	0.5	Synergy
P. aeruginosa 2	0.375	Synergy
With Levofloxacin		
S. aureus ATCC 29213	0.3125	Synergy
E. coli ATCC 25922	0.1875	Synergy
K. pneumoniae 1	0.75	Additive
P. aeruginosa 2	1.125	Indifference
With Erythromycin		
S. aureus ATCC 29213	0.375	Synergy
E. coli ATCC 25922	0.1875	Synergy
K. pneumoniae 1	0.75	Additive
P. aeruginosa 2	0.09375	Synergy

Conclusion

In summary, the methanol leaf extract of *Erigeron floribundus* exhibited antibacterial activity against multidrug-resistant bacteria. Furthermore, the extract demonstrated potent resistance-modifying properties by synergistically potentiating erythromycin against all tested bacterial strains. This finding scientifically validates the traditional usage of the plant and highlights its potential as a source of natural antibiotic adjuvants. Future work should focus on

identifying the synergistic compounds and determining their mechanism of action through efflux pump inhibition and membrane integrity assays. The safety and efficacy of the combination should be assessed through *in vivo* studies to develop practical approaches for resistance therapy.

Abbreviations

ATCC: American Type Culture Collection
ATB: Antibiotic
CFU: Colony Forming Unit
CLSI: Clinical & Laboratory Standards Institute
DMSO: Dimethyl Sulfoxide
FIC: Fractional Inhibitory Concentration
GAE: Gallic Acid Equivalent
MBC: Minimum Bactericidal Concentration
MDR: Multidrug-Resistant
MHB: Mueller-Hinton Broth
MIC: Minimum Inhibitory Concentration
QE: Quercetin Equivalent
RMA: Resistance-Modifying Agent
TAE: Tannic Acid Equivalent
TPC: Total Phenolic Content
TFC: Total Flavonoid Content
TTC: Total Tannin Content
TSC: Total Saponin Content

Authors' Contribution

VN: Conceptualization, Supervision, Methodology, Validation, Writing – review & editing; MSNY: Methodology, Investigation, Formal analysis, Writing – review & editing; PVTF: Conceptualization, Methodology, Data curation, Writing – original draft, Writing – review & editing; GMT: Investigation, Methodology, Formal analysis, Writing – review & editing; BPK: Methodology, Formal analysis, Writing – review & editing; JPND: Methodology, Formal analysis, Writing – review & editing; PLNP: Methodology, Formal analysis, Writing – review & editing; JDDT: Supervision, Resources, Validation, Writing – review & editing; All authors read and approved the final manuscript.

Acknowledgments

The authors sincerely thank the technical staff of the Research Laboratory of Microbiology and Antimicrobial Substances at the University of Dschang for their invaluable support and assistance during this work. We also gratefully acknowledge the National Herbarium of Yaoundé for the botanical identification of the plant material.

Conflict of interest

The authors declare no conflict of interest

Article history:

Received: 28 February 2026
Received in revised form: 28 April 2026
Accepted: 01 May 2026
Available online: 01 May 2026

References

1. World Health Organization. 2024. Antimicrobial resistance. Fact sheet. Geneva: WHO. Available from: https://www.who.int/news-room/fact-sheets/detail/antimicrobial-resistance
2. O'Neill J. 2016. Tackling drug-resistant infections globally: final report and recommendations. London: Review on Antimicrobial Resistance (Wellcome Trust and HM Government). Available from: https://amr-review.org/sites/default/files/160525_Final%20paper_with%20cover.pdf
3. Ventola CL. 2015. The antibiotic resistance crisis: part 1: causes and threats. *Pharm Ther*. 40(4):277-83.
4. Brown ED, Wright GD. 2016. Antibacterial drug discovery in the resistance era. *Nature*. 529(7586):336-43.
5. Hemaiswarya S, Kruthiventi AK, Doble M. 2008. Synergism between natural products and antibiotics against infectious diseases. *Phytomedicine*. 15(8):639-52.
6. Cheesman MJ, Ilanko A, Blonk B, Cock IE. 2017. Developing new antimicrobial therapies: are synergistic combinations of plant extracts/compounds with conventional antibiotics the solution? *Pharmacogn Rev*. 11(22):57-72.
7. Stavri M, Piddock LJ, Gibbons S. 2007. Bacterial efflux pump inhibitors from natural sources. *J Antimicrob Chemother*. 59(6):1247-60.
8. Khan IA, Mirza ZM. 2021. Synergistic antibacterial evaluation of plant extracts with antibiotics against multidrug-resistant bacteria. *J Infect Public Health*. 14(1):1-7.
9. Wagner H, Ulrich-Merzenich G. 2009. Synergy research: approaching a new generation of phytopharmaceuticals. *Phytomedicine*. 16(2-3):97-110.
10. Dibong SD, Mpondo Mpondo E, Ngoye A, Priso RJ. 2011. Ethnobotany and phytomedicine of the medicinal plants of Douala (Cameroon). *J Ethnobiol Ethnomed*. 7:33.
11. Tra Bi FH, Koné MW, Kouamé NF. 2008. Antifungal activity of *Erigeron floribundus* (Asteraceae) from Côte d'Ivoire, West Africa. *Trop J Pharm Res*. 7(2):975-9.
12. Asongalem EA, Foyet HS, Ngogang J, Folefoc GN, Dimo T, Kamtchouing P. 2004. Analgesic and antiinflammatory activities of *Erigeron floribundus*. *J Ethnopharmacol*. 91(2-3):301-8.
13. Berto C, Maggi F, Nya PCB, Pettena A, Boschiero I, Dall'Acqua S. 2014. Phenolic constituents of *Erigeron floribundus* (Asteraceae), a Cameroonian medicinal plant. *Nat Prod Commun*. 9(12):1691-4.
14. Petrelli R, Orsomando G, Sorci L, Maggi F, Ranjbarian F, Biapa Nya PC, et al. 2016. Biological activities of the essential oil from *Erigeron floribundus*. *Molecules*. 21(8):1065.
15. Kuiate JR, Tsona AA, Foko J, Bessiere JM, Menut C, Zollo PHA. 2005. Chemical composition and *in vitro* antifungal properties of essential oils from leaves and flowers of *Erigeron floribundus* (H.B. et K.) Sch. Bip. from Cameroon. *J Essent Oil Res*. 17(3):261-4.
16. Yapo F, Yapi F, Ahiboh H, Hauhouot-Attounbre ML, Guédé N, Djaman J, et al. 2011. Immunomodulatory effect of the aqueous extract of *Erigeron floribundus* (Kunth) Sch Beep (Asteraceae) leaf in rabbits. *Trop J Pharm Res*. 10(2):185-91.
17. Ramde-Tiendrebeogo A, Tibiri A, Hilou A, Lompo M, Millogo-Kone H, Nacoulma OG, et al. 2012. Antioxidative and antibacterial activities of phenolic compounds from *Ficus sue* Forssk. *Int J Biol Chem Sci*. 6(1):328-36.
18. Chang C, Yang M, Wen H, Chern J. 2002. Estimation of total flavonoid content in propolis by two complementary colorimetric methods. *J Food Drug Anal*. 10(3):178-82.
19. Singleton VL, Rossi JA. 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *Am J Enol Vitic*. 16(3):144-58.
20. Koziol MJ. 1991. Afrosimetric estimation of threshold saponin concentration for bitterness in quinoa (*Chenopodium quinoa* Willd). *J Food Compos Anal*. 4(3):211-9.
21. Clinical and Laboratory Standards Institute. 2018. *Performance standards for antimicrobial susceptibility testing*. 28th ed. CLSI supplement M100. Wayne (PA): CLSI.
22. National Committee for Clinical Laboratory Standards. 2002. *Reference method for broth dilution antifungal susceptibility testing of yeasts; approved standard*. 2nd ed. NCCLS document M27-A2. Wayne (PA): NCCLS.
23. Odds FC. 2003. Synergy, antagonism, and what the chequerboard puts between them. *J Antimicrob Chemother*. 52(1):1.
24. Cushnie TPT, Lamb AJ. 2011. Recent advances in understanding the antibacterial properties of flavonoids. *Int J Antimicrob Agents*. 38(2):99-107.
25. Kuete V. 2010. Potential of Cameroonian plants and derived products against microbial infections: a review. *Planta Med*. 76(14):1479-91.
26. Rios JL, Recio MC. 2005. Medicinal plants and antimicrobial activity. *J Ethnopharmacol*. 100(1-2):80-4.
27. Fyfe C, Grossman TH, Kerstein K, Sutcliffe J. 2016. Resistance to macrolide antibiotics in public health pathogens. *Cold Spring Harb Perspect Med*. 6(10):a025395.
28. Blair JMA, Webber MA, Baylay AJ, Ogbolu DO, Piddock LJV. 2015. Molecular mechanisms of antibiotic resistance. *Nat Rev Microbiol*. 13(1):42-51.