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Chemical components and biological properties from acetone extracts of *Conamomum vietnamense*

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Summary

Conamomum vietnamense is an endemic and rare species from Vietnam. The aim of this study is to determine the chemical compositions, antibacterial and antioxidant properties of the acetone extracts obtained from the different organs of this species for the first time. A total of 82 components were identified from the acetone extracts of leaf, flower, and rhizome of *C. vietnamense* using Gas chromatography–mass spectrometry (GC/MS) technique. Furthermore, the agar disk-diffusion method was also used to determine the antibacterial activity of the *C. vietnamense* extracts. Accordingly, the leaf extract was found to be effective against eight out of nine bacterial strains while the flower and rhizome extracts displayed activity against four out of nine tested bacteria. In addition, the three organs of *C. vietnamense* also possessed the high DPPH scavenging properties. The results of this study indicate that *C. vietnamense* extracts have the potential to be developed into pharmaceutical products in the future.

Keywords: Antibacterial activity, Antioxidant activity, Chemical components, *Conamomum vietnamense*, GC/MS, Zingiberaceae.

Introduction

Zingiberaceae is an important and large family including 4 sub-families, 6 tribes, 57 genera, and more than 1900 species distributed in subtropical and tropical areas, but commonly found in southeast Asia (BOONMA et al., 2023). Members of Zingiberaceae are considered as the medicinal plants in ethnobotanical studies (INTA et al., 2023). Besides, they are also used for cosmetic, ornamental plants, spices, food (TRIMANTO et al., 2018), perfume, and dye (SAENSOUK and SAENSOUK, 2021). The chemical components in the Zingiberaceae plants determine the main factor responsible for their biological activities. For instance, Curcumin, a major component in some *Curcuma* species, contributes many bioactivities like anticancer, expectorant, hepatoprotective, antibacterial, antioxidant, antimutagenic, anti-inflammatory, and antidiabetic activities (CHATZINASIOU et al., 2019; VAN et al., 2021b). In addition, bornyl acetate, eucalyptol, geraniol and β -linalool make to the unique aroma of Zingiberaceae species (PANG et al., 2017) as well as possess so many biological properties such as antimicrobial, antioxidant, anti-inflammatory, anti-proliferative, anticancer, antihyperlipidemic, antinociceptive and analgesic (PEREIRA et al., 2018; VAN et al., 2021c).

Conamomum Ridl. is a genus belonging to Zingiberaceae family which possesses 10 species commonly distributed in montane and lowland forests in Borneo, Sumatra, Singapore, Malay Peninsula, and Indochina (DE BOER et al., 2018; LUU et al., 2019). Only four *Conamomum* species have been discovered in Vietnam, including *Conamomum pierreanum* collected from Dak Nong province (LUU

et al., 2021), *C. odorum* from Khanh Hoa province (LUU et al., 2019), *C. rubidum* from Lam Dong and Khanh Hoa provinces (LAMXAY and NEWMAN, 2012), and *C. vietnamense* from Lam Dong and Dak Nong provinces (LY et al., 2022).

Conamomum vietnamense N.S.Lý & T.S. Hoang is an endemic plant from Vietnam which was recently described as a new species by Ly et al. (LY et al., 2022). More recently, studies provided the chemical compositions and the biological activities of the essential oils obtained from *C. vietnamense* (LE et al., 2023; NGUYEN et al., 2023; VAN et al., 2024). For instance, the chemical components and the antimicrobial effects of the *C. vietnamense* essential oils have been investigated by our prior work (VAN et al., 2024). Additionally, the chemical constituents, mosquito larvicidal, and antimicrobial properties of the essential oils obtained from this plant have been reported by recent studies (LE et al., 2023; NGUYEN et al., 2023). So far, the chemical profiles and biological activities of the different solvent extracts isolated from *C. vietnamense* has been unknown. The present study, therefore, first investigated the chemical compositions and antibacterial properties of the acetone extracts obtained from the different organs of *C. vietnamense*.

Materials and methods

Plant materials

The rhizomes, flowers, and leaves of *C. vietnamense* were collected from Dong Nai commune, Bu Dang district, Binh Phuoc province by Van Hop Nguyen, coordinates at 11°47'156" N and 107°22'862" E in July 2022. The vouchered specimen, BÐ-BP06072022, was deposited in the Herbarium of Faculty of Natural Resources and Environment, Vietnam National University of Forestry at Dongnai.

Bacterial strains

Nine bacterial strains were used to identify the antibacterial activity of *C. vietnamense* acetone extracts, including five Gram-negative strains (*Escherichia coli* ATCC 25922, *Salmonella typhimurium* ATCC 13311, *Klebsiella pneumoniae* ATCC 13883, *Shigella flexneri* ATCC 9199, *Enterobacter hormaechei* ATCC 700323) and four Gram-positive strains (*Staphylococcus saprophyticus* BAA750, *Staphylococcus aureus* ATCC 29213, *Staphylococcus aureus* ATCC 25923, *Bacillus cereus* ATCC 13883).

Extraction procedures

The fresh rhizomes, flowers, and leaves of *C. vietnamense* were washed and dried at temperature of 50 °C, ground into powder using fine grinder. 100 g of sample powder was ground and soaked in 500 mL of acetone solution (Thermo Fisher Scientific, USA). The mixture solution was filtered through Whatman filter paper. The filter residue then was soaked again in 300 mL of acetone solution two more times

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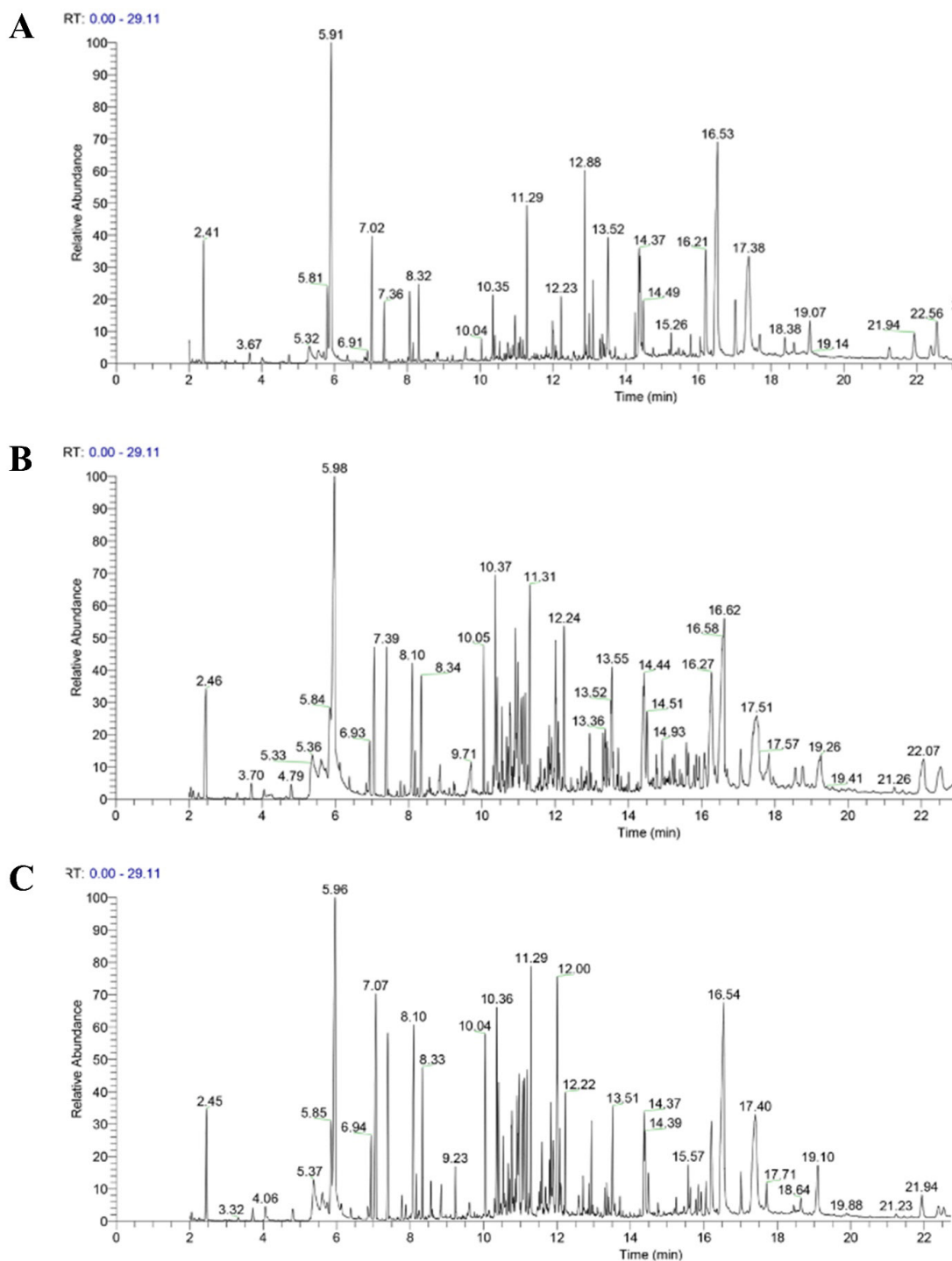


Fig. 1: The GC chromatogram of the acetone extracts from *C. vietnamense*. (A) leaf, (B) flower, and (C) rhizome

to collect the final filtrate. A vacuum evaporator was used to evaporate the filtrate at temperature of 45 °C to obtain the brown extract (VAN et al., 2021a).

Gas chromatography/mass spectrometry (GC/MS)

The chemical compounds of the *C. vietnamense* extracts were determined using the TRACE 1310 Gas Chromatograph (Thermo Fisher Scientific Inc., Waltham, MA, USA) coupled with ISQ 7000 single quadrupole mass spectrometer. The injector temperature was held at 250 °C in the flow rate 36 mL/min, split ratio of 30:1. The helium

carrier gas flow rate at 1.2 mL/min using the DB-5MS column (30 m × 0.25 mm × 0.25 μm) as the stationary phase. The mass spectrometer was run at an ionization voltage 70 eV and ion source temperature at 250 °C. The oven temperature was programmed at 80 °C for 5 min, and was increased 20 °C/min until reached 280 °C, the oven was subsequently held at 280 °C for 10 min. The acquisitions scan mass range of MS was 29-650 m/z with the scanning frequency of 2 scans/sec. Chemical compositions of the acetone extracts of the studied samples were determined based on mass spectral comparison through the NIST 2017 library.

Antibacterial activity

The antibacterial effect was determined by agar disk-diffusion method as recommended by The Clinical & Laboratory Standards Institute (CLSI, 2016). Initially, the bacterial strains were grown on the LB broth until turbidity of 0.5 McFarland standards. One hundred microliters of each bacterial culture were spread on a sterile Mueller Hinton plate. Subsequently, the studied extracts were diluted with 15% DMSO solution at concentration ranges of 100, 150, and 200 mg/mL and then, 10 microliters of diluted samples were added to each disc (diameter 6 mm). The plates were incubated under aerobic conditions for 24 h at 37 °C, and then the zone of inhibition of each studied sample was measured. The gentamicin (Nam Khoa BioTek, Vietnam; for antibacterial tests) was used as the positive control whereas 15% DMSO solution was used as negative control. The antibacterial tests were carried out in three separate times. The mean and standard deviation of data from three replicates were presented. Fisher's least significant difference (LSD) approach and one-way analysis of variance (ANOVA) were used as statistical methods (Statgraphics program, Centurion XV).

Antioxidant activity

The antioxidant activity was determined using the DPPH radical scavenging assay, which was based on the procedure described by (LE et al., 2021a) with minor modifications. The extract was completely solved in 99.8% methanol (Thermo Fisher Scientific, USA) to obtain different concentrations. Then, 0.3 mL of this solution was mixed with 3.7 mL of 0.1 mM DPPH solution. The resulting mixture was shaken and then incubated in a dark room at room temperature for 30 min.

The absorbance was subsequently measured at a wavelength of 517 nm using a UV-Vis spectrophotometer (Genesys 20, USA). The antioxidant effects of the extracts were compared with ascorbic acid, as a control and it was estimated by the IC₅₀ value (concentration of the sample required to inhibit 50% of DPPH free radical scavenging). The antioxidant activity was calculated using the following equation:

$$\%DPPH = \frac{A_0 - A_i}{A_0} \times 100$$

Where A_0 and A_i represent the absorbance of the blank solution (DPPH solution) and the sample (DPPH solution and extract), respectively.

Results and discussion

Chemical compositions of acetone extracts from *C. vietnamense* leaf, flower, and rhizome

The chemical compositions from the acetone extracts of *C. vietnamense* leaf, flower and rhizome were presented in Tab. 1 and Fig. 1. A total of 82 chemical compounds were identified from 3 studied samples of which 40 components were in the leaf, 53 constituents were in the flower, and 55 compounds were in the rhizome. The major components of the leaf included eucalyptol (21.90%), dihydrochrysin (7.31%), neophytadiene (5.34%), *cis*-vaccenic acid (4.96%), 1-(+)-ascorbic acid 2,6-dihexadecanoate (4.92%), linalool (4.86%), *D*-limonene (4.62%), and 2-pentanone, 4-hydroxy-4-methyl- (4.39%). The flower was mainly composed of eucalyptol (13.29%), *trans*-nerolidol (3.56%), dihydrochrysin (5.91%), octadecanoic acid (4.45%), *D*-limonene (3.32%), linalool (3.16%), fenchol (3.16%), and α -santalene (3.16%). The rhizome was characterized by the predominance of eucalyptol (9.98%), linalool (5.11%), α -epi-7-epi-5-eudesmol (4.19%), endo-borneol (3.54%), and fenchol (3.14%).

In this study, the chemical compositions of the acetone extracts from *C. vietnamense* were reported for the first time. However, the chemical components of the essential oils obtained from this species were

Tab. 1: The chemical compounds of acetone extracts of *C. vietnamense* leaf, flower, and rhizome

№	RT	Compounds	Relative percentage		
			Leaf	Flower	Rhizome
1	2.41	2-Pentanone, 4-hydroxy-4-methyl-	4.39	3.10	2.13
2	3.67	α -Pinene	0.53	0.38	-
3	3.71	2-Thujene	-	-	0.28
4	4.04	Camphene	-	0.25	0.48
5	4.65	β -Pinene	-	0.10	-
6	4.75	β -Myrcene	0.50	0.39	0.33
7	5.32	α -Phellandrene	1.87	-	2.16
8	5.61	α -Terpinene	-	-	1.35
9	5.81	<i>D</i> -Limonene	4.62	3.32	-
10	5.85	m-Mentha-6,8-diene, (R)-(+)-	-	-	2.61
11	5.91	Eucalyptol	21.90	13.29	9.98
12	6.35	γ -Terpinene	0.33	-	-
13	6.82	p-Mentha-2,4(8)-diene	1.71	0.20	0.24
14	6.91	Fenchone	0.50	0.97	1.38
15	7.02	Linalool	4.86	3.16	5.11
16	7.36	Fenchol	2.12	3.16	3.14
17	7.68	cis-p-Menth-2-en-1-ol	-	0.10	-
18	7.78	(+)-2-Bornanone	-	0.22	0.30
19	7.88	Camphene hydrate	-	0.20	0.18
20	8.02	Terpineol	0.21	-	-
21	8.10	endo-Borneol	2.29	2.66	3.54
22	8.15	p-Menth-1-en-4-ol, (R)-(-)-	0.59	-	0.46
23	8.17	Terpinen-4-ol	-	0.51	-
24	8.25	2-Cyclohexen-1-one, 4-(1-methylethyl)-	-	0.10	-
25	8.32	3-Cyclohexene-1-methanol, $\alpha,\alpha,4$ -trimethyl-, (R)-	2.43	1.80	1.93
26	8.57	Fenchyl acetate	-	0.18	0.39
27	8.85	Geraniol	-	0.79	0.60
28	9.10	(Z)-Undec-6-en-2-one	0.23	-	-
29	9.23	Bornyl acetate	-	0.14	0.53
30	9.26	2-Undecanone	-	0.11	-
31	9.60	Hydrocinnamic acid	0.83	-	0.35
32	9.71	Benzenepropanoic acid, silver (1+) salt	-	1.18	-
33	10.04	α -Cubebene	0.63	-	-
34	10.05	Copaene	-	1.88	1.93
35	10.31	Di-epi- α -cedrene	-	0.33	-
36	10.35	α -Santalene	1.59	3.16	2.34
37	10.41	β -Caryophyllene	0.70	1.65	1.73
38	10.47	Lemnalol	-	0.16	-
39	10.53	cis- β -Farnesene	0.45	0.90	0.78
40	10.57	Epi- β -Santalene	-	0.21	-
41	10.68	1,4,7,-Cycloundecatriene, 1,5,9,9-tetramethyl-, Z,Z,Z-	-	0.50	0.55
42	10.72	4-epi- α -Acoradiene	-	0.64	-
43	10.76	Guaia-6,9-diene	0.77	-	1.88
44	10.77	Cadina-1(10),4-diene	-	0.95	-
45	10.78	γ -Muulolene	-	0.53	-
46	10.83	2-Tridecanone	0.46	-	-
47	10.87	Aristolochene	-	0.36	0.50
48	10.91	1 β ,4 β H,10 β H-Guaia-5,11-diene	0.94	-	2.38
49	10.92	Longifolene-(V4)	-	2.06	-
50	10.96	α -Himachalene	1.73	1.75	-
51	10.97	4-epi- α -Acoradiene	-	-	2.52
52	11.06	Cadina-1(10),4-diene	0.75	2.03	2.63

53	11.10	o-Menth-2-ene, 4-isopropylidene-1-vinyl	0.92	-	-
54	11.11	Epizonarene	-	-	2.34
55	11.12	Zonarene	-	1.27	-
56	11.17	Cubenene	0.70	1.31	1.91
57	11.29	trans-Nerolido	1.44	3.56	3.15
58	11.51	Caryophyllenyl alcohol	-	-	0.42
59	11.56	Caryophyllene oxide	-	-	0.41
60	11.59	Guaiol	-	-	0.80
61	11.69	Ledol	-	-	0.40
62	11.80	Cubenol	-	0.52	0.75
63	11.84	epi- γ -Eudesmol	-	0.64	1.11
64	11.90	tau-Muurolol	-	0.66	1.13
65	11.99	Eudesm-4(14)-en-11-ol	1.92	-	-
66	12.01	α -epi-7-epi-5-Eudesmol	-	2.32	4.19
67	12.08	Allohimachalol	-	0.77	-
68	12.11	7-epi-cis-sesquisabinene hydrate	-	0.45	0.61
69	12.23	Farnesol	1.47	2.51	1.24
70	12.71	2-Naphthalenemethanol, decahydro-8-hydroxy- $\alpha,\alpha,4a,8$ -tetramethyl-	-	-	0.35
71	12.88	Neophytadiene	5.34	-	0.30
72	13.30	(2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl) benzene	0.71	-	-
73	13.52	l-(+)-Ascorbic acid 2,6-dihexadecanoate	4.92	2.51	1.86
74	14.26	Phytol	1.24	-	-
75	14.37	9(E),11(E)-Conjugated linoleic acid	4.60	-	1.69
76	14.40	cis-Vaccenic acid	4.96	-	-
77	14.17	17-Octadecynoic acid	-	-	0.97
78	14.44	cis-13-Octadecenoic acid	-	4.45	-
79	14.49	Octadecanoic acid	2.79	-	0.54
80	15.26	Falcarinol	0.73	-	-
81	15.57	7-(Iodomethyl)-3a-isopropenyl- 6,7-dimethyloctahydro-1H-indene	-	-	0.53
82	16.21	Dihydrochrysin	7.31	5.91	2.44
83	16.62	Unknown compound	-	10.78	-
84	16.54	Unknown compound	-	-	9.33
85	17.40	Unknown compound	-	-	7.42
86	17.51	Unknown compound	-	7.10	-
Total			96.98	98.18	98.60

reported by recent reports. For instance, in our prior work, the chemical compounds of the essential oils isolated from the rhizome and the aerial part of *C. vietnamense* collected from Binh Phuoc province, Vietnam have been investigated. Accordingly, the aerial part essential oil was dominated by 1,8-cineole (41.3%), limonene (15.1%), linalool (10.2%) while 1,8-cineole (30.7%), linalool (19.4%), endo-borneol (10.8%) were the major constituents of the rhizome essential oil (VAN et al., 2024). Similarly, the leaf and rhizome essential oils of this species grown in Bao Loc province, Vietnam have been reported, in which eucalyptol (49.49 and 40.47%) and limonene (26.20 and 18.74%) were the main compounds of both organs (NGUYEN et al., 2023). More recently, the leaf and rhizome essential oils from *C. vietnamense* were also provided in which its oils contained 1,8-cineole (47.9-62.0%) and limonene (10.3-16.2%) as the major components (LE et al., 2023). Furthermore, the chemical compositions from another *Conamomum* plant have been also reported by prior studies. For examples, the rhizome essential oil of *Amomum rubidum*, a synonym of *Conamomum rubidum* (DE BOER et al., 2018) collected from Vietnam, was reported to contain β -phellandrene (16.1%), limonene (14.4%), and δ -3-carene (13.9%) as the major compounds (LE et al., 2020). Besides, the chemical constituents of the essential oils ob-

tained from *Conamomum rubidum* (syn.: *Amomum rubidum*) leaf and stem were also reported, thereby the stem essential oil was found to be rich in d-3-carene (21.9%), limonene (17.8%), and β -phellandrene (14.6%) whereas 1,8-cineole (37.7%), d-3-carene (19.5%), and limonene (16.3%) were the major compounds in the leaf essential oils (LE et al., 2021b). In addition, four new chemical compounds isolated from *Conamomum rubidum* grown in Vietnam, including conarubins A–D have been recently provided (HOANG et al., 2023).

In addition, the chemical constituents of the acetone extracts obtained from other species belonging to Zingiberaceae using GC/MS techniques have been also provided. For instance, the acetone extract of the *Curcuma caesia* rhizome from India was reported to contain β -elemenone (22.25%), methyl stearolate (14.29%), and α -selinene (5.47%) (CHATURVEDI et al., 2021). Similarly, the acetone extract isolated from *C. caesia* rhizome grown in India was characterized by the predominance of 2-pentanone, 4-hydroxy-4-methyl acid (7.68%), 4,5,6,6a-tetrahydro-2(1H)-pentalenone (7.92%), and trans-2-Decen-1-ol (4.04%) (CHITRA et al., 2020). In addition, the chemical compounds of the acetone extracts from the six organs of *Alpinia zerumbet* collected from Japan were also reported. Accordingly, sitosterol was the richest compound in stem, rhizome, and leaf extracts while the flower and pericarp were found to be rich stigmasterol, and cholest-4-ene-3,6-dione was the most abundant compound in the seed extract (CHOMPOO et al., 2012). The acetone extract of the *Zingiber zerumbet* rhizome grown in India was mainly composed of zerumbone (64.58%), diacetone alcohol (10.64%), and α -humulene (8.93%) (DASH et al., 2020). Moreover, the acetone extract of the *A. subulatum* contained 1,8-cineole (19.70%), eugenol (12.50%), α -terpineol (8.10%) as the major compounds (KAPOOR et al., 2008).

The acetone extract of the *Globba macrocarpa* aerial part collected from Vietnam was found to be rich in linolenic acid (19.89%), palmitic acid (13.05%), and phytol (7.52%) whereas germacrene D (15.25%), 1H-indole, 4-(3-methyl-2-butenyl) (14.33%), and (*E*)- β -farnesene (11.28%) were the major compounds in the rhizome extract (VAN et al., 2022a). The acetone extract obtained from the whole plants of *C. xanthella* grown in Vietnam were characterized by the predominance of 1,8-cineole (11.3%), n-hexadecanoic acid (11.13%), and stigmasterol (8.2%) (VAN et al., 2022b). The chemical components of the acetone extracts from the two *Curcuma* species collected from Vietnam were performed by recent reports. Accordingly, the *C. cotuana* extract possessed (*E*)-labda-8(17),12-diene-15,16-dial (14.58%), n-exadecanoic acid (10.96%), and 3,7,11,15-tetramethylhexadec-2-en-1-yl acetate (8.13%) as the main components while *C. thorellii* extract was found to be rich in (*E*)-labda-8(17),12-diene-15,16-dial (33.37%), vitamin E (12.33%), and phytol (9.83%) (VAN et al., 2023). More recently, the chemical compositions of the acetone extracts of four Zingiberaceae species collected from Vietnam. Consequently, the *Amomum biflorum* extract was found to contain (-)-alcanfor (14.12%), and reynosin (8.74%), neophytadiene (8.31%) as the major compounds while *Globba marantina* extract was found the majority of n-hexadecanoic acid (13.42%), neophytadiene (7.35%), and (*E*)-labda-8(17),12-diene-15,16-dial (6.65%). The major compounds of *Curcuma pierreana* extract were n-hexadecanoic acid (12.07%), (*E*)-labda-8(17),12-diene-15,16-dial (11.97%), and cis-13-octadecenoic acid (6.86%) whereas n-hexadecanoic acid (14.03%), β -caryophyllene (11.76%), and (*E*)-labda-8(17),12-diene-15,16-dial (9.88%) were the main compounds in the *Curcuma gracillima* extract (HUYNH et al., 2024).

Antibacterial activity of acetone extracts from *C. vietnamense* leaf, flower, and rhizome

The antibacterial activities of the acetone extracts obtained from the *C. vietnamense* leaf, flower and rhizome were presented in Tab. 2, 3, and 4. Overall, there was no significant difference in the zone of inhibition among different dilutions of the studied samples. The leaf extract had an inhibitory effect on eight out of nine bacterial strains, in-

cluding *E. coli*, *S. typhimurium*, *S. saprophyticus*, *B. cereus*, *K. pneumoniae*, *S. flexneri*, *S. aureus* ATCC 29213, and *S. aureus* ATCC 25923 (Tab. 2) while the flower and rhizome extracts were found to be effective against four out of nine bacteria such as *S. saprophyticus*, *B. cereus*, *S. aureus* ATCC 29213, and *S. aureus* ATCC 25923 (Tab. 3 and 4).

Prior studies reported that the essential oils from *C. vietnamense* also possessed antimicrobial activities. For instances, the rhizome essential oil of this species showed antimicrobial effect against bacterial and fungal strains, including *Salmonella enterica*, *Pseudomonas aeruginosa*, *E. coli*, *B. cereus*, *S. aureus*, *Enterococcus faecalis*, and *Candida albicans* (NGUYEN et al., 2023). Moreover, The rhizome and

Tab. 2: Antibacterial property of acetone extract of *C. vietnamense* leaf on nine tested bacterial strains

Bacterial strains	Zone of inhibition (mm)			
	100 mg/mL	150 mg/mL	200 mg/mL	Gentamycin
<i>E. coli</i> ATCC 25922	9.33±1.44 ^a	8.00±0.87 ^a	8.33±0.58 ^a	20.33±1.53 ^b
<i>S. typhimurium</i> ATCC 13311	7.33±0.58 ^a	8.33±1.53 ^a	7.33±0.58 ^a	24.17±0.29 ^a
<i>S. saprophyticus</i> BAA750	8.67±0.29 ^a	9.83±1.04 ^{ab}	10.67±0.29 ^b	26.17±1.26 ^c
<i>B. cereus</i> ATCC 13883	11.17±1.26 ^a	10.67±0.29 ^a	11.33±0.58 ^a	20.50±0.50 ^b
<i>K. pneumoniae</i> ATCC 13883	7.33±0.29 ^a	8.00±1.00 ^a	7.50±0.50 ^a	16.67±0.58 ^b
<i>S. flexneri</i> ATCC 9199	7.67±0.58 ^a	8.50±1.00 ^a	7.83±1.04 ^a	20.00±1.00 ^b
<i>S. aureus</i> ATCC 29213	8.33±0.58 ^a	11.17±0.76 ^b	10.33±1.04 ^b	22.67±1.53 ^c
<i>S. aureus</i> ATCC 25923	9.83±1.26 ^a	11.17±0.76 ^a	11.33±2.25 ^a	24.50±1.50 ^b
<i>E. hormaechei</i> ATCC 700323	-	-	-	18.11±0.15

Different superscript lower-case letters in the same row denote significant difference ($p < 0.05$). (-) Not active

Tab. 3: Antibacterial property of acetone extract of *C. vietnamense* flower on nine tested bacterial strains

Bacterial strains	Zone of inhibition (mm)			
	100 mg/mL	150 mg/mL	200 mg/mL	Gentamycin
<i>E. coli</i> ATCC 25922	-	-	-	20.83±0.76
<i>S. typhimurium</i> ATCC 13311	-	-	-	26.83±0.76
<i>S. saprophyticus</i> BAA750	11.00±0.87 ^a	11.17±0.29 ^a	10.67±0.58 ^a	29.83±0.76 ^b
<i>B. cereus</i> ATCC 13883	10.83±0.29 ^a	10.50±0.50 ^a	10.50±0.50 ^a	20.83±0.76 ^a
<i>K. pneumoniae</i> ATCC 13883	-	-	-	17.83±0.76
<i>S. flexneri</i> ATCC 9199	-	-	-	21.33±1.15
<i>S. aureus</i> ATCC 29213	12.67±1.54 ^a	13.17±0.29 ^a	13.67±1.53 ^a	24.83±1.04 ^b
<i>S. aureus</i> ATCC 25923	12.33±0.58 ^a	13.57±0.58 ^b	13.50±0.50 ^b	21.50±0.50 ^a
<i>E. hormaechei</i> ATCC 700323	-	-	-	19.33±1.15

Different superscript lower-case letters in the same row denote significant difference ($p < 0.05$). (-) Not active

Tab. 4: Antibacterial property of acetone extract of *C. vietnamense* rhizome on nine tested bacterial strains

Bacterial strains	Zone of inhibition (mm)			
	100 mg/mL	150 mg/mL	200 mg/mL	Gentamycin
<i>E. coli</i> ATCC 25922	-	-	-	19.17±1.26
<i>S. typhimurium</i> ATCC 13311	-	-	-	24.33±0.58
<i>S. saprophyticus</i> BAA750	12.67±1.15 ^a	12.00±1.00 ^a	13.33±1.53 ^a	29.00±1.00 ^b
<i>B. cereus</i> ATCC 13883	11.33±1.15 ^a	10.67±0.58 ^a	11.67±0.58 ^a	22.50±0.50 ^b
<i>K. pneumoniae</i> ATCC 13883	-	-	-	20.17±1.61
<i>S. flexneri</i> ATCC 9199	-	-	-	22.33±1.15
<i>S. aureus</i> ATCC 29213	12.83±1.04 ^a	12.67±1.53 ^a	12.00±1.73 ^a	24.33±0.76 ^b
<i>S. aureus</i> ATCC 25923	10.17±0.29 ^a	10.33±0.29 ^a	10.67±0.29 ^a	22.83±1.04 ^b
<i>E. hormaechei</i> ATCC 700323	-	-	-	18.17±0.29

Different superscript lower-case letters in the same row denote significant difference ($p < 0.05$). (-) Not active

Tab. 5: DPPH scavenging capacity of *C. vietnamense* acetone extracts

	Leaf	Flower	Rhizome	Ascorbic acid
IC ₅₀ (µg/mL)	92.25±8.60	121.64±6.18	48.15±9.32	2.47±0.07

leaf essential oils of *C. vietnamense* were also found to be effective against *S. aureus* and *Bacillus subtilis* (LE et al., 2023). The rhizome and aerial part essential oils obtained from *C. vietnamense* had an inhibitory effect on many bacterial and fungal strains, including *B. cereus*, *E. faecalis*, *Listeria monocytogenes*, *S. aureus*, *S. saprophyticus*, *Enterobacter cloacae*, *E. hormachei*, *K. pneumoniae*, *E. coli*, *S. enteritidis*, *Salmonella flexneri*, *Shigella sonnei*, *C. albicans*, *C. glabrata*, *C. krusei*, and *C. parasilosis* (VAN et al., 2024). In addition, the antimicrobial property of the essential oil obtained from the rhizome, leaf and stem of *Conamomum rubidum* collected from Vietnam was also reported. Accordingly, this essential oil was found to be effective against two fungal strains such as *Aspergillus niger* and *Fusarium oxysporum* (LE et al., 2020) while the leaf and stem essential oils displayed activity against *P. aeruginosa*, *F. oxysporum*, and *C. albicans* (LE et al., 2021b).

The antibacterial properties of the acetone extracts obtained from *C. vietnamense* in this study may be attributed to their chemical components. For example, eucalyptol, another name of 1,8-cineole, was found to be effective against many microorganisms such as *E. coli*, *Cylindrocarpum mali*, *Mycobacterium smegmatis*, *P. aeruginosa*, *S. aureus*, *Streptococcus faecalis*, *Sclerotinia sclerotiorum*, *Stereum purpureum*, *Sotlyfis cinerea*, *A. niger*, and *C. albicans* (HENDRY et al., 2009; PRUDENT et al., 1993). Another major compound in *C. vietnamense* acetone extract, linalool, was also reported to display activity against a large number of bacterial and fungal strains, including *P. aeruginosa*, *S. aureus*, *E. coli*, *C. albicans* (HERMAN et al., 2016), *Porphyromonas gingivalis*, *Prevotella nigrescens*, *Prevotella intermedia*, *Fusobacterium nucleatum* subsp. *polymorphum*, *F. nucleatum* subsp. *fusiforme*, *F. nucleatum* subsp. *nucleatum*, *F. nucleatum* subsp. *animalis*, *F. nucleatum* subsp. *vincentii*, *Streptococcus sobrinus*, *S. mutans*, *Aggregatibacter actinomycetemcomitans* (PARK et al., 2012), *L. monocytogenes* (WU et al., 2014), and *Pasteurella multocida* (GAO et al., 2019). *D*-Limonene was also found to be effective against a wide range of bacterial and fungal strains such as *Saccharomyces cerevisiae*, *B. subtilis*, *S. aureus*, and *E. coli* (ZAHY et al., 2015). β -caryophyllene has also been shown to be resistant to many strains of microorganisms, including *Trichoderma reesei*, *Rhizopus oryzae*, *Penicillium citrinum*, *A. niger*, *P. aeruginosa*, *K. pneumoniae*, *E. coli*, *S. aureus*, *B. subtilis*, and *B. cereus* (DAHAM et al., 2015). In addition, so many chemical compounds such as fenchone, fenchol, borneol, and terpinen-4-ol had an inhibitory effect on a large number of bacterial and fungal strains (KOTAN et al., 2007). Nerolidol was also found to be effective against *Streptococcus mutans*, *E. faecalis*, *Staphylococcus epidermidis*, *S. aureus*, *P. aeruginosa*, *E. coli*, and *K. pneumoniae* (DE MOURA et al., 2021).

Antioxidant activity of acetone extracts from *C. vietnamense* leaf, flower, and rhizome

The antioxidant properties of the acetone extracts isolated from the *C. vietnamense* leaf, flower and rhizome were shown in Tab. 5. Accordingly, the rhizome extract possessed the highest antioxidant effect with the IC₅₀ value of 48.15±9.32 µg/mL, followed by the leaf extract (92.25±8.60 µg/mL) and the flower extract (121.64±6.18 µg/mL).

The chemical constituents of the acetone extracts obtained from *C. vietnamense* could be the main factor contributing to their antioxidant activity. For instance, *D*-limonene have strong antioxidant effects as evaluated using the superoxide radical scavenging assay, hydroxyl radical assay, iron chelating, FRAP, ABTS, and DPPH with IC₅₀ values of 462.89, 442.75, -18475.5, -589.85, 603.23, 384.73 µM, respectively (SHAH and MEHTA, 2018). Moreover, *D*-limonene possessed the potential anti-inflammatory and antioxidant activities because it could prevent the MMP-2 and -9 mRNA expression levels through the control of the iNOS, COX-2, PGE2, TGF- β and ERK1/2 signal-

ing pathways (YU et al., 2017). In addition, linalool could be used as a source of potential antioxidant property because of its hydrogen peroxide (H₂O₂) and DPPH scavenging capacity (JABIR et al., 2018). Besides, in patients with carpal tunnel syndrome, linalool reduces pulse rate and blood pressure as well as increases antioxidant capacity (SEOL et al., 2016). Furthermore, the supplementation of about 20-30mg 1,8-cineole/kg to diets of broilers led to improvements in antioxidant capacity, immunity as well as intestinal morphology (DI et al., 2022). Another chemical compound in the acetone extracts from *C. vietnamense*, nerolidol has been reported to have the antioxidant properties on the DPPH and nitric oxide scavenging activities (UDAVANT et al., 2023).

Conclusion

The present study firstly reported on the chemical components and antibacterial activities of the acetone extracts from *C. vietnamense* leaf, flower and rhizome, a species recently described in Vietnam. Eighty-two chemical compounds were found in three organs of the studied species of which eucalyptol was the most abundant compound in the leaf, flower, and rhizome extracts with the relative percentage of 21.90%, 13.29%, and 9.98%, respectively. Also, the leaf extract had an inhibitory effect on eight out of nine microorganisms while flower and rhizome extracts displayed activity against four out of nine bacterial strains. The *C. vietnamense* rhizome had the high DPPH scavenging capacity with IC₅₀ value of 48.15±9.32 µg/mL, followed by the leaf extract (92.25±8.60 µg/mL), and the flower extract (121.64±6.18 µg/mL).

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Conflict of interest

No potential conflict of interest was reported by the authors.









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