PLANT SCIENCE TODAY
ISSN 2348-1900 (online)
Vol x(x): xx–xx
https://doi.org/10.14719/pst.1993

HORIZON
e-Publishing Group
HePG

RESEARCH COMMUNICATION

Chemical profiles and antibacterial activity of acetone extract of two *Curcuma* species from Vietnam

Hong Thien Van[1], Minh Dang Tran[1], Thi Kim Ngan Tran[1], Hanh Thi Dieu Nguyen[1], Ngoc An Nguyen[1], Nguyen Tuong An Huynh[2], Van Son Le[3], Quoc Hung Nguyen[4], Thanh Tho Le[4], Nga Nguyen-Phi[5,6] & Tan Viet Pham[1*]

[1]Institute of Biotechnology and Food technology, Industrial University of Ho Chi Minh City, 12 Nguyen Van Bao Street, Go Vap District, Ho Chi Minh City, Vietnam
[2]Institute of International and Postgraduate Education, Industrial University of Ho Chi Minh City, 12 Nguyen Van Bao Street, Go Vap District, Ho Chi Minh City, Vietnam
[3]Binh Chau-Phuoc Buu Nature Reserve, Bung Rieng Ward, Xuyen Moc District, Ba Ria-Vung Tau Province, Vietnam
[4]Center of Analytical Services and Experimentation HCMC, Vietnam, 2 Nguyen Van Thu, Dakao Ward, District 1, Ho Chi Minh City, Vietnam
[5]Faculty of Biology and Biotechnology, University of Science, Vietnam National University HCMC, 227 Nguyen Van Cu Street, District 5, Ho Chi Minh City, Vietnam
[6]Vietnam National University, Ho Chi Minh City, Thu Duc City, Ho Chi Minh City, Vietnam

*Email: phamtanviet@iuh.edu.vn

OPEN ACCESS

ARTICLE HISTORY

Received: 09 July 2022
Accepted: 17 October 2022

Available online
Version 1.0 : 07 March 2023

Check for updates

Additional information

Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

Reprints & permissions information is available at https://horizonepublishing.com/journals/index.php/PST/open_access_policy

Publisher's Note: Horizon e-Publishing Group remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Indexing: Plant Science Today, published by Horizon e-Publishing Group, is covered by Scopus, Web of Science, BIOSIS Previews, Clarivate Analytics, NAAS, UGC Care, etc
See https://horizonepublishing.com/journals/index.php/PST/indexing_abstracting

CITE THIS ARTICLE

Van H T, Tran M D, Tran T K N, Nguyen H T D, Nguyen N A, Huynh N T A, Le V S, Nguyen Q H, Le T T, Phi N N, Pham TV. Chemical profiles and antibacterial activity of acetone extract of two *Curcuma* species from Vietnam. Plant Science Today (Early Access). https://doi.org/10.14719/pst.1993

Abstract

Curcuma thorelii Gagnep. and *Curcuma cotuana* Luu, Škorničk. & H.Đ.Trần are the rare species only found in Southeast Asia. The present study was the first to explore the chemical compositions and antibacterial effects of the whole plant acetone extracts of these 2 species. Altogether 41 and 31 compounds have been identified in *C. thorelii* and *C. cotuana* extracts by gas chromatography/mass spectrometry. Accordingly, the *C. thorelii* extract contained (E)-labda-8(17),12-diene-15,16-dial (33.37%), vitamin E (12.33%), phytol (9.83%) as the major compounds while *C. cotuana* extract contained predominantly (E)-labda-8(17),12-diene-15,16-dial (14.58%), n-exadecanoic acid (10.96%), 3,7,11,15-tetramethylhexadec-2-en-1-yl acetate (8.13%), γ-sitosterol (7.97%). In addition, results from disc diffusion assay have shown that *C. thorelii* acetone extract had inhibitory effects on 5 out of 10 pathogenic bacterial strains such as *Bacillus cereus* (ATCC 11778), *Listeria monocytogenes* (ATCC 19111), *Staphylococcus aureus* (ATCC 25923), *S. aureus* (ATCC 29213) and *S. saprophyticus* (BAA750) while *C. cotuana* acetone extract was found to be effective only against *B. cereus.* The obtained results showed that the acetone extracts of *C. thorelii* and *C. cotuana* possessed several valuable bioactive compounds as well as promising antibacterial activity, which place a good foundation for future pharmaceutical product development.

Keywords

Curcuma thorelii, *Curcuma cotuana*, acetone extract, GC/MS, chemical composition, antibacterial activity

Introduction

Curcuma genus includes about 108 species widely distributed throughout the tropical and subtropical regions, particularly in northern Australia, Malaysia, Thailand, Indochina, India and Vietnam (1, 2). Previous studies have recorded about 29 *Curcuma* species for the flora of Vietnam (2-4). Several *Curcuma* species were commercially used for their good flavoring, preservative properties (1), as well as for their uses in traditional medicine to treat abscesses, diarrhea, infectious wounds, leucorrhea, pneumonia, bronchial complaints and insect bites (5). Moreover, bioactive compounds and biological activities of many *Curcuma* of plants have been experimentally studied

(6-10). For instance, xanthorrhizol, a bioactive component isolated from the rhizome of *Curcuma xanthorrhiza* was found to effectively act against *Candida albicans* biofilms development (6). The essential oil extracted from *C. xanthorrhiza* has been reported to possess cytotoxic activity against YMB-1 breast cancer cell line (7). *Curcuma longa* extracts and their major bioactive compound, curcumin had anti-parasitic and anti-inflammatory potency by parenteral and oral application in animal models (10). Recently, study revealed that essential oil of *Cucurma gracillima* possessed several bioactive compounds such as α-cucumene, caryophylene, *allo*-aromadendrene, caryophylene oxide, 3-carene, α-pinene (9).

Materials and Methods

Plants

The whole plant sample of *Curcuma cotuana* was collected from Tay Giang district, Quang Nam province, Vietnam and *Curcuma thorelii* were collected from Binh Chau-Phuoc Buu Nature Reserve, Ba Ria-Vung Tau province, Vietnam. The vouchered specimens of *C. cotuana* and *C. thorelii* was deposited at the Herbarium of Department of Ecology and Evolutionary Biology, Faculty of Biology & Biotechnology, University of Science, Vietnam National University HCMC (PHH) with the vouchered specimen numbers Van HT 132b and Van HT 132c respectively (Fig. 1).

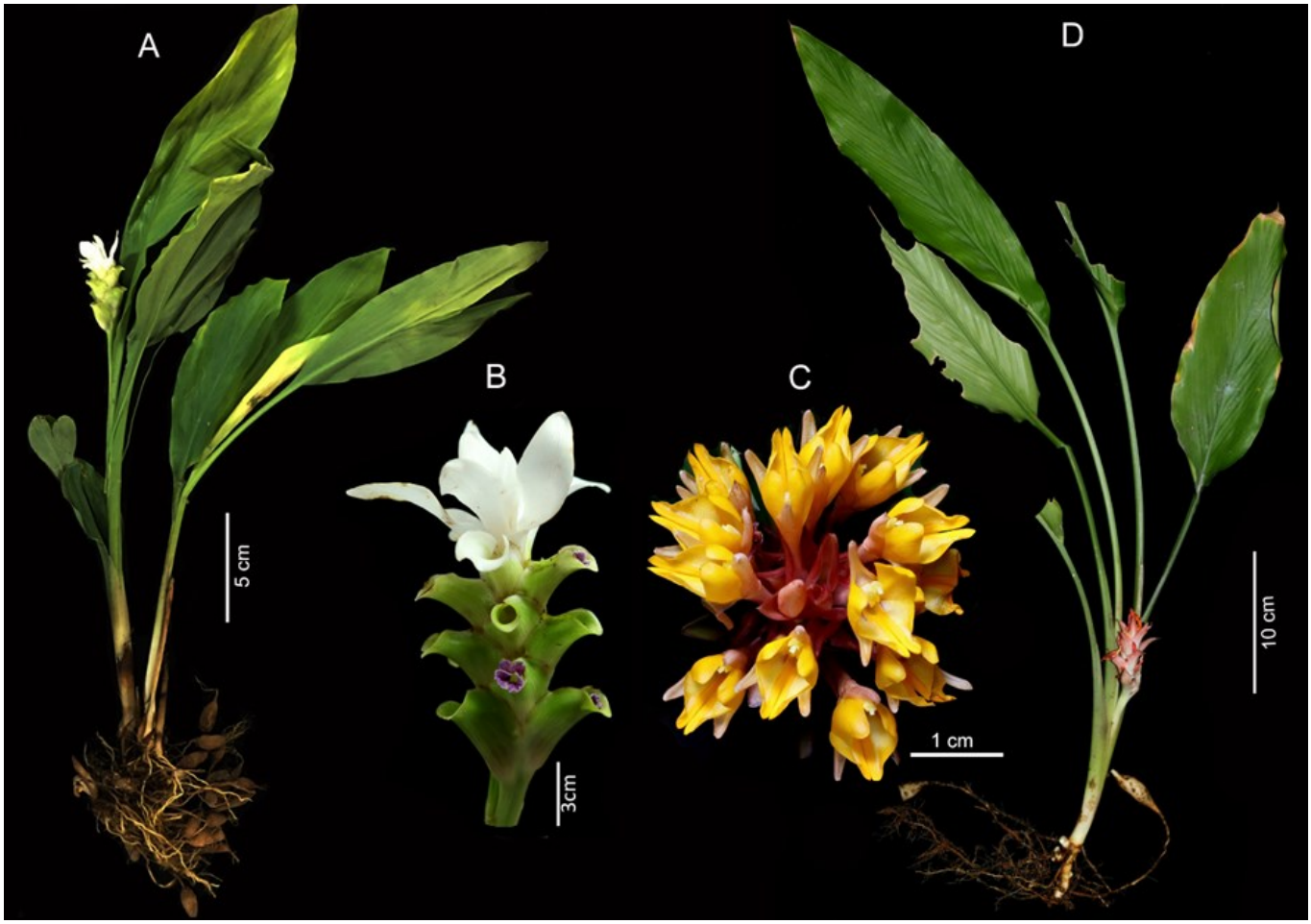

Fig. 1. Two *Curcuma* species in this study. *C. thorelii:* A. whole plant, B. flower. *C. cotuana*: C. flower, D. whole plant.

Curcuma thorelii Gagnep. was first discovered in Laos by Gagnepain in 1907. To date, this plant is only found in Laos, Cambodia, Thailand and Vietnam (11). Meanwhile, the first specimen of *Curcuma cotuana* Luu, Škorničk. & H.Đ.Trần was collected from Quang Nam province, Vietnam in 2017 (12). To date, only one study on chemical compositions and antibacterial effects of *C. thorelii* essential oils has been conducted (13). Therefore, this study was the first to explore the information about chemical component and biological property of the acetone extracts from *C. thorelii* and *C. cotuana*, thereby enhancing their potential especially in medicinal applications.

Bacterial strains

Five Gram positive bacteria *Bacillus cereus* (ATCC 11778), *Listeria monocytogenes* (ATCC 19111), *Staphylococcus aureus* (ATCC 25923), *Staphylococcus aureus* (ATCC 29213), *Staphylococcus saprophyticus* (BAA750) and Gram negative bacteria *Escherichia coli* (ATCC 25922), *Klebsiella pneumoniae* (ATCC 13883), *Salmonella enteritidis* (ATCC 13072), *Salmonella typhimurium* (ATCC 13311), *Shigella flexneri* (ATCC 9199) were used to identify antibacterial activity of *C. thorelii* and *C. cotuana* extracts.

Extraction procedures of total acetone extract

The whole plant samples of each species were dried at 50^0C until constant weights were obtained and then ground into powder. Five hundred mL of acetone solution

99% (Thermo Fisher Scientific, USA) was used to soaked 100 g powder for 72 hrs at room temperature. The extracts were filtered using Whatman paper and this process was repeated thrice. The rotary evaporator was used to concentrate the filtrates under the reduced pressure at 40^{0}C, resulting in the brown extracts (14). Each of the extracts was divided into 2 parts, one for GC/MS analysis and the other for antibacterial assay.

Gas chromatography–mass spectrometry assays

The chemical compositions of the acetone extracts obtained from *C. cotuana* and *C. thorelii* were identified by TRACETM 1310 Gas Chromatograph (Thermo Fisher Scientific Inc., Waltham, MA, USA) coupled with ISQ 7000 single quadrupole mass spectrometer. The DB-5MS column (30 m x 0.25 mm X 0.25 µm) was used as the stationary phase and Helium at a flow rate of 1.2 mL/min was used as the carrier gas. Samples were injected into the GC system by splitting method with split ratio of 30:1, splitless time 1 min, at 250^{0}C in the flow rate of 36 mL/min. The transfer line temperature was set at 250^{0}C. The oven temperature was set at 80^{0}C for 5 min, and was increased 20^{0}C/min until it reached and held at 280 ^{0}C for 10 min. The electron impact ionization was set as 70 eV and the filament source temperature was set at 250^{0}C. The acquisitions scan mass range of MS was 29-650 m/z with the scanning frequency of 2 scans/sec. The chemical components of the acetone extracts were identified based on the comparison between their mass spectra and the NIST 2017 library (15).

Determination of antibacterial activity of the extracts

The antibacterial properties of the acetone extracts obtained from *C. cotuana* and *C. thorelii* were determined by disc diffusion assay (16). The bacterial strains were grown in Luria-Bertani broth until the turbidity reached 0.5 McFarland standards then 100 µL of the cultures was spread on Mueller Hinton agar plates. The sterile paper discs impregnated with 10 µL of acetone extracts were placed on the agar surface. Sterilized distilled water and gentamycin discs (10 µg) (supplied by Nam Khoa BioTek, Vietnam) were used as the negative and positive controls, respectively. The plates were incubated at 37^{0}C for 18-24 hrs. before measuring the zone of inhibition.

Data analysis

Three independent experiments were performed, and the results were expressed as mean ± standard deviation (SD). The Statgraphics Centurion XV software was used for statistical analysis of the results using one-way analysis of variance (ANOVA) and Fisher's least significant difference (LSD) procedure.

Results and Discussion

Chemical compositions of acetone extract from C. thorelii and C. cotuana

The GC/MS data showed that there were 41 and 31 constituents found in the acetone extracts of *C. thorelii* and *C. cotuana*, respectively (Table 1 and Fig. 2). The *C. thorelii*

Table 1. Components of acetone extracts from *C. thorelii* and *C. cotuana*

RT (min)	Compounds	*C. thorelii* (%)	*C. cotuana* (%)
3.34	2-Heptanol	0.11	-
3.74	α-Pinene	0.04	-
4.09	Camphene	0.18	-
4.62	Propanoic acid, 3-ethoxy-, ethyl ester	0.15	-
4.69	β-Pinene	0.49	-
5.27	Ethylidene acetate	0.07	-
5.95	Eucalyptol	0.08	-
6.59	Thymine	-	0.52
7.81	(+)-2-Bornanone	-	3.7
8.02	Isoborneol	-	3.02
8.12	endo-Borneol	-	1.14
8.37	2-Pinen-10-ol	0.19	-
8.52	Benzofuran, 2,3-dihydro-	0.08	-
8.63	5-Hydroxymethylfurfural	-	4.89
9.27	(-)-Bornyl acetate	-	0.74
9.3	Isobornyl acetate	-	0.76
9.83	Benzaldehyde, 4-hydroxy-	0.08	-
9.94	β-Ionone	0.14	-
10.09	Sobrerol 8-acetate	-	0.56
10.44	Caryophyllene	1.62	0.50
10.53	trans-Geranylacetone	0.09	-
10.57	α-Farnesene	-	6.29
10.71	Humulene	0.09	-
11.32	(6E)-Nerolidol	0.27	-
11.52	Longipinocarvone	0.19	-
11.59	Caryophyllene oxide	2.39	-
11.92	Caryophylladienol II	0.73	-
12.1	Isoaromadendrene epoxide	0.27	-
12.2	Longifolenaldehyde	0.43	-
12.66	Loliolide	0.24	-
12.92	Ambrial	2.12	5.36
12.95	Hexahydrofarnesyl acetone	0.78	-
13.33	5 Farnesyl acetone	0.34	-
13.55	n-Hexadecanoic acid	1.63	10.96
13.7	2-Butenal	0.30	-
13.83	3-Buten-2-one	0.52	-
14.16	Pimara-7,15-dien-3-one	0.64	-
14.31	Phytol	9.83	1.39
14.40	cis-9,cis-12-Octadecadienoic acid	-	4.36
14.44	Linolenic acid	-	7.30
14.47	5-(7a-Isopropenyl-4,5-dimethyl-octahydroinden-4-yl)-3-methyl-pent-2-enal	1.26	-
14.52	Octadecanoic acid	-	1.62

14.88	β-Sitosterol	1.04	-
14.99	7α-Hydroxydehydroepiandrosterone	-	1.57
15.02	Benzyl α-d-glucoside	-	0.99
15.09	Labd-14-ene-8,13-diol, (13R)-	1.08	-
15.65	(E)-Labda-8(17),12-diene-15,16-dial	33.37	14.58
16.25	Palmitin, 2-mono	-	1.72
16.78	3,7,11,15-Tetramethylhexadec-2-en-1-yl acetate	-	8.13
17.45	β-Monolinolein	-	3.02
17.5	2-Monolinolenin	-	1.53
17.64	5β-Stigmastane-3,6-dione	-	0.69
18.47	Squalene	9.62	0.74
20.82	Lupeol	3.13	-
21.38	γ-Tocopherol	2.55	0.17
22.71	Vitamin E	12.33	0.04
24.66	Campesterol	1.42	0.97
25.21	Stigmasterol	3.17	2.31
26.2	γ-Sitosterol	5.63	7.97
28.19	β-Tocopheryl acetate	1.18	0.69
	Total:	99.87	98.23

extract was characterized by the predominance of (E)-labda-8(17),12-diene-15,16-dial (33.37%), vitamin E (12.33%), phytol (9.83%), squalene (9.62%), γ-sitosterol (5.63%), stigmasterol (3.17%), and lupeol (3.13%). On the other hand, the *C. cotuana* extract was found to be rich in (E)-labda-8(17), 12-diene-15,16-dial (14.58%), n-exadeca noic acid (10.96%), 3,7,11,15-etramethylhexadec-2-en-1-yl acetate (8.13%), γ-sitosterol (7.97%), linolenic acid (7.30%), α-farnesene (6.29%) and ambrial (5.36%).

The most abundant component in the acetone extracts of *C. thorelii* and *C. cotuana*, (E)-labda-8(17), 12-diene-15,16-dial, has also been found in some other Zingiberaceae members such as *Renealmia alpinia* (17), *Alpinia speciose* (18), *Aframomum daniellii, A. alboviolaceum* and *A. stipulatu* (19). This compound has been reported to possess cytotoxic activity against four cancer cell lines, including MCF-7, Ca Ski, PC-3 and HT-29 (20). Vitamin E was considered as a strong peroxyl radical scavenger and chain-breaking antioxidant which prevented the multiplication of free radical damage in biological membranes (21) and showed strong cytotoxic effect against human breast cancer cells (22, 23). In addition, stigmasterol has been demonstrated to have strong cytotoxic activity against human hepatoma HepG2 cell (24). Phytol has been found to display a wide range of biological properties, including anxiolytic, cytotoxic, metabolism-modulating, autophagy and apoptosis inducing, immune modulating, antioxidant, antinociceptive, antimicrobial, and anti-inflammatory activities (25). It was reported that γ-sitosterol increased insulin secretion in response to glucose in rats and thus, could be used as an antidiabetic agent (26). This compound has also been reported to inhibit the proliferation of MCF-7 and rA549 cells (27).

Besides acetone, other solvents have been used for chemical analysis of different *Curcuma* species, which led to the identification of a vast and diverse repertoire of bioactive compounds. For example, xanthorrhizol; 1H-3a, 7-methanoazulene, and curcumene were the major compounds in the essential oils and hexane extract obtained from *C. aromatica* (28). The ethanolic extract of *C. longa* rhizome was found to contain 11 major components such as alkaloids, glycosides, carbohydrates, saponins, proteins, steroids, terpenoids, flavonoids, phlobotannins, anthraquinones and tannins (29). Out of these 11 compounds, the hydroalcoholic extract contained only 10, except phlobotannins while the choloroform extract possessed only 7, except terpenoids, proteins, flavonoids and tannins (29). In addition, the chemical constituents of methanolic extract isolated from the rhizomes of 3 *Curcuma* plants have been investigated, in which the *C. decipiens* extract predominantly contained 4, 4-di methyl-2, 4, 5, 6-tetrahydro-1H-inden-2-yl) acetic acid, cycloheptyl ethyl methylphosphonate, and trispiro [4.2.4.2.4.2.] hene-

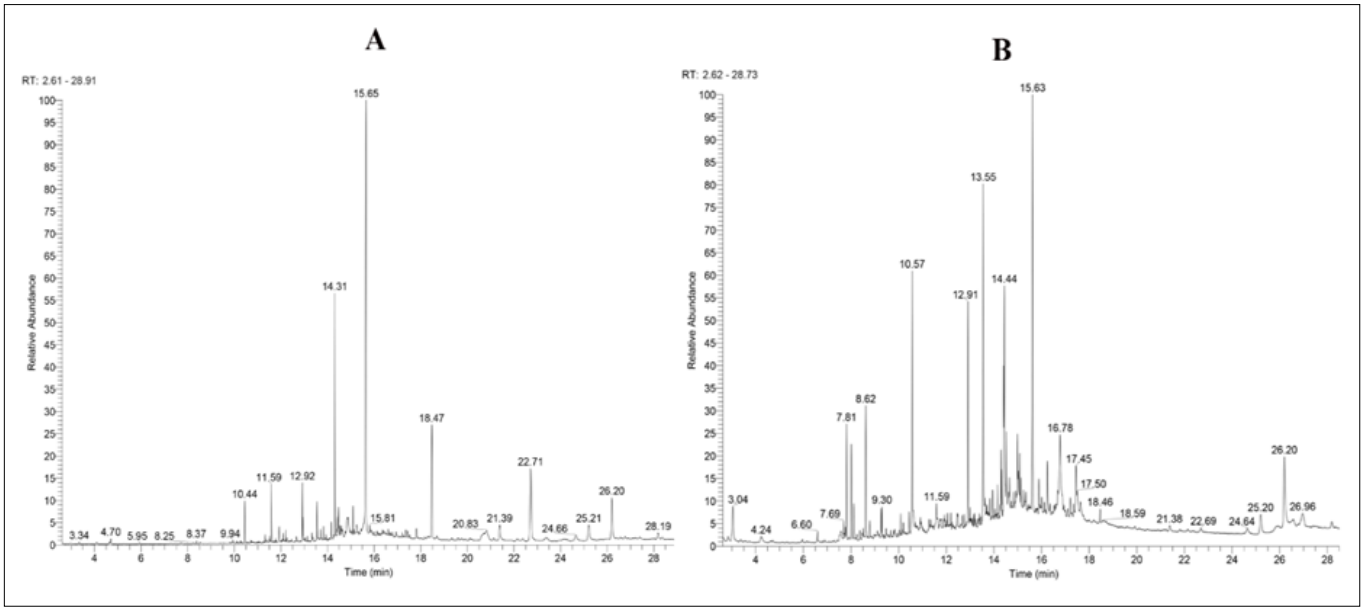

Fig. 2. Gas chromatogram of acetone extracts from *C. thorelii* (A) and *C. cotuana* (B).

icosane, the *C. angustifolia* extract was identified as a mixture of oleic acid, n-hexadecanoic acid, nitrous oxide, and acetic acid while the *C. longa* extract was found to be rich in ar-tumerone, curlone and tumerone (30).

Antibacterial activity of acetone extract from C. thorelii and C. cotuana

The acetone extract of *C. thorelii* was found to be effective against all five Gram positive bacterial strains, including *B. cereus, L. monocytogenes*, *S. aureus* (ATTC 25923), *S. aureus* (ATTC 29213) and *S. saprophyticus* (Table 2 and Fig. 3). Accordingly, the antibacterial effect of *C. thorelii* extract was the most potent against *B. cereus* with the inhibition zone of 16.49 mm, followed by *S. aureus* (11.32 mm), *S. aureus* (9.94 mm), *S. saprophyticus* (9.12 mm) and *L. monocytogenes* (8.13 mm). Meanwhile, the *C. cotuana* extract was found to inhibit only *B. cereus* but with stronger effect (19.83 mm) compared to *C. thorelii*.

Table 2. Inhibition zone of acetone extracts obtained from *C. thorelii* and *C. cotuana*

Bacterial strains	Growth inhibition zone (mm)		
	C. thorelii	***C. cotuana***	**Gentamycin**
Bacillus cereus	16.49±0.11[a]	19.83 ±1.26[b]	27.05±0.14[c]
Listeria monocytogenes	8.13±0.05[a]	-	26.99±0.04[b]
Staphylococcus aureus (ATTC 25923)	11.32±0.10[a]	-	27.68±0.12[b]
Staphylococcus aureus (ATTC 29213)	9.94±0.25[a]	-	25.76±0.10[b]
Staphylococcus saprophyticus	9.12±0.09[a]	-	28.74±0.18[b]
Escherichia coli	-	-	28.33±1.15
Klebsiella pneumoniae	-	-	27.50±0.5
Salmonella enteritidis	-	-	28.16±1.26
Salmonella typhimurium	-	-	28.17±1.26
Shigella flexneri	-	-	27.83±1.44

The chemical constituents found in the acetone extracts of *C. thorelii* and *C. cotuana* could be the main factors contributing to the antibacterial activity. For instance, vitamin E was proved to have an inhibitory effect on *K. pneumoniae, S. aureus, E. coli* and *Staphylococcus epidermidis* (31). *Stigmasterol* was found to be effective against many pathogenic bacteria such as *Streptococcus faecalis, E. coli, S. aureus, Pseudomonas fluorescens, K. pneumoniae,* and *S. typhimurium* (32). Phytol has been reported to possess antibacterial effects against *E. coli, Pseudomonas aeruginosa, Mycobacterium tuberculosis, S. aureus, Clostridium sporogenes, Sarcina lutea* and *Enterococcus faecalis* (25). Reports states that squalene could inhibit the virulence of *S. aureus* (33).

The other solvent extracts of several *Curcuma* species have also been shown to have antibacterial properties. For instance, the aqueous and ethanolic extracts of *C. longa* had antibacterial activity against *S. aureus, P. aeruginosa, E. coli* and *Salmonella enterica* (34). The ethanolic extracts of three *Curcuma* species, including *C. aromatica, C. zedoaria* and *C. longa* had inhibitory effects on some bacterial strains such as *Salmonella typhosa, Listeria monocytogenes, P. aeruginosa, B. cereus* and *S. aureus* (35) whereas the aqueous and methanolic extracts of *C. aromatica* was found to be effective against *E. coli* and *S. aureus* (36).

Conclusion

In this study, 41 and 31 chemical compounds were successfully identified in the acetone extracts of *C. thorelii* and *C. cotuana* respectively. The *C. thorelii* extract was found to be rich in (E)-labda-8(17),12-diene-15,16-dial (33.37%), vitamin E (12.33%), phytol (9.83%), squalene (9.62%), γ-sitosterol (5.63%) while (E)-labda-8(17),12-diene-15,16-dial (14.58%), n-hexadecanoic acid (10.96%), 3,7,11,15-tetramethylhexadec-2-en-1-yl acetate (8.13%) were the major compounds in the *C. cotuana* extract. None of the Gram negative strains was inhibited by the 2 acetone extracts. The extract of *C. thorelii* was found to be effective against all 5 Gram positive strains including *B. cereus, L. monocytogenes*, *S. aureus* (ATCC 25923), *S. aureus* (ATCC 29213) and *S. saprophyticus* whereas the *C. cotuana* extract was only found to be highly effective against *B. cereus. Curcuma thorelii* and *C. cotuana* are rare species and their relatively small populations are found in very few regions of Vietnam. In order to ensure the development and conservation of these species, a small number of samples were collected and our next work is to propagate these 2 species to obtain more abundant sample sources. This study was only the first step to provide more insight and place a good foundation for future applications of *C. cotuana* and *C. thorelii* in pharmaceutical products and other related fields.

Acknowledgements

This research is funded by University of Science, VNU-HCM, Vietnam under grant number SH-CNSH 2022-03.

Authors contributions

Hong Thien Van designed this study. All authors searched and handled the data. Hong Thien Van and Tan Viet Pham drafted the manuscript and resolved all the queries of editors and reviewers.

Compliance with ethical standards

Conflict of interest: No conflict of interest was declared by the authors.

Ethical issues: None.

References

1. Rahaman M, Rakib A, Mitra S, Tareq A, Emran T, Shahid-Ud-Daula A et al. The Genus *Curcuma* and Inflammation: Overview of the Pharmacological Perspectives. Plants 2021;63(10):2020. https://doi.org/10.3390/plants10010063
2. Leong-Škorničková J, Ly NS, Nguyen QB. *Curcuma arida* and *C. sahuynhensis*, 2 new species from subgenus Ecomata (Zingiberaceae) from Vietnam. Phytotaxa. 2015;192(3):181-9. https://doi.org/10.11646/phytotaxa.192.3.4
3. Pham HH. An Illustrated Flora of Vietnam. Vol. 3. Araceae. Young Publishing House, Ho Chi Minh City; 2000.
4. Nguyen QB. Zingiberaceae-Flora of Vietnam. Science and Technology Publishing, Ha Noi; 2017.
5. Akarchariya N, Sirilun S, Julsrigival J, Chansakaowa S. Chemical profiling and antimicrobial activity of essential oil from *Curcuma aeruginosa* Roxb., *Curcuma glans* K. Larsen & J. Mood and *Curcuma* cf. *xanthorrhiza* Roxb. collected in Thailand. Asian Pacific Journal of Tropical Biomedicine. 2017;7(10):881-85. https://doi.org/10.1016/j.apjtb.2017.-09.009
6. Rukayadi Y, Hwang JK. *In vitro* activity of xanthorrhizol isolated from the rhizome of Javanese turmeric (*Curcuma xanthorrhiza* Roxb.) against *Candida albicans* biofilms. Phytotherapy Research. 2013;27(7):1061-66. https://doi.org/10.1002/ptr.4834
7. Udin Z. Sitotoksisitas Xanthorrhizol Dari Minyak Atsiri Rimpang *Curcuma Xanthorrhiza* Roxb. Terhadap Sel Kanker Payudara YBM-1. Jurnal Kimia Terapan Indonesia. 2013;15(1):23-19. https://doi.org/10.14203/jkti.v15i1.101
8. Araujo C, Leon L. Biological activities of *Curcuma longa* L. Memórias do Instituto Oswaldo Cruz. 2001;96:723-28. https://doi.org/10.1590/S0074-02762001000500026
9. Van HT, Phan UTX, Doan VD, Le VS. Chemical Constituents of Essential Oil from Rhizomes and Aerial Parts of *Curcuma gracillima*. Chemistry of Natural Compounds. 2021;57(3):569-71. https://doi.org/10.1007/s10600-021-03419-6
10. Azeez TB, Lunghar J. Antiinflammatory effects of Turmeric (*Curcuma longa*) and Ginger (*Zingiber officinale*). Inflammation and Natural Products. 2021:127-46. https://doi.org/10.1016/B978-0-12-819218-4.00011-0
11. Leong-Škorničková J, Šída O, Tran HD. *Curcuma pygmaea* sp. nov. (Zingiberaceae) from Vietnam and notes on two related species *C. parviflora* and *C. thorelii*. Nordic Journal of Botany. 2014;32(1):119-27. https://doi.org/10.1111/njb.91749
12. Luu HT, Tran HD, Nguyen TQT, Leong-Škorničková J. *Curcuma cotuana* sp. nov. (Zingiberaceae: Zingibereae) from central Vietnam. Nordic Journal of Botany. 2017;35(5):-552-56. https://doi.org/10.1111/njb.01594
13. Van HT, Luu LT, Pham TV, Tran MD, Nguyen TKN, Le VS, Nguyen QH, Le TT, Nguyen PN. Chemical constituents and antibacterial activity of essential oils of *Curcuma thorelii* Gagnep. (Zingiberaceae). Chem Nat Compd. 2023;59(1):176-179. https://doi.org/10.1007/s10600-023-03947-3
14. Le HT, Luu TN, Nguyen HMT, Nguyen DH, Le PTQ, Trinh NN et al. Antibacterial, antioxidant and cytotoxic activities of different fractions of acetone extract from flowers of *Dipterocarpus intricatus* Dyer. (Dipterocarpaceae). Plant Science Today. 2021;8(2):273-77. https://doi.org/0.14719/pst.2021.8.2.1086
15. Van HT, Le NT, Nguyen DL, Tran GB, Huynh NTA, Vo HS et al. Chemical profile and antibacterial activity of acetone extract of *Homalomena cochinchinensis* Engl. (Araceae). Plant Science Today. 2021;8(1):58-65. https://doi.org/10.14719/pst.2021.8.1.971
16. Clinical, Institute LS. Methods for antimicrobial dilution and disk susceptibility testing of infrequently isolated or fastidious bacteria; approved guideline. Clinical and Laboratory Standards Institute Wayne, PA; 2010.
17. Lognay G, Marlier M, Severin M, Haubruge E, Gibon V, Trevejo E. On the characterization of some terpenes from *Renealmia alpinia* Rott. (Maas) oleoresin. Flavour and Fragrance Journal. 1991;6(1):87-91. https://doi.org/10.1002/ffj.2730060113
18. Itokawa H, Morita M, Mihashi S. Phenolic compounds from the rhizomes of *Alpinia speciosa*. Phytochemistry. 1981;20(11):2503-06. https://doi.org/10.1016/0031-9422(81)830828
19. Abreu PM, Noronha RG. Volatile Constituents of the Rhizomes of *Aframomum alboviolaceum* (Ridley) K. Schum. from Guinea-Bissau. Flavour and Fragrance Journal. 1997;12(2):79-83. https://doi.org/10.1002/(SICI)1099-1026(199703)12:2<79::AID-FFJ617>-3.0.CO;2-7
20. Ahmed Hamdi OA, Syed Abdul Rahman SN, Awang K, Abdul Wahab N, Looi CY, Thomas NF et al. Cytotoxic constituents from the rhizomes of *Curcuma zedoaria*. The Scientific World Journal. 2014;2014. https://doi.org/10.1155/2014/321943
21. Yamauchi R. Vitamin E: mechanism of its antioxidant activity. Food Science and Technology International, Tokyo. 1997;3(4):301-19. https://doi.org/10.3136/fsti9596t9798.3.-301
22. Knekt P, Aromaa A, Maatela J, Aaran R-K, Nikkari T, Hakama M et al. Vitamin E and cancer prevention. The American Journal of Clinical Nutrition. 1991;53(1):283S-6S. https://doi.org/10.1093/ajcn/53.1.283S
23. Kline K, Yu W, Sanders BG. Vitamin E and breast cancer. The Journal of Nutrition. 2004;134(12):3458S-62S. https://doi.org/10.1093/jn/134.12.3458S
24. Kim YS, Li XF, Kang KH, Ryu B, Kim SK. Stigmasterol isolated from marine microalgae *Navicula incerta* induces apoptosis in human hepatoma HepG2 cells. BMB reports. 2014;47(8):433. https://doi.org/10.5483/BMBRep.2014.47.8.153
25. Islam MT, Ali ES, Uddin SJ, Shaw S, Islam MA, Ahmed MI, et al. Phytol: A review of biomedical activities. Food and Chemical Toxicology. 2018;121:82-94. https://doi.org/10.1016/j.fct.2018.08.032
26. Balamurugan R, Duraipandiyan V, Ignacimuthu S. Antidiabetic activity of γ-sitosterol isolated from *Lippia nodiflora* L. in streptozotocin induced diabetic rats. European Journal of Pharmacology. 2011;667(1-3):410-48. https://doi.org/10.1016/j.ejphar.2011.05.025
27. Sundarraj S, Thangam R, Sreevani V, Kaveri K, Gunasekaran P, Achiraman S et al. γ-Sitosterol from *Acacia nilotica* L. induces

G2/M cell cycle arrest and apoptosis through c-Myc suppression in MCF-7 and A549 cells. Journal of Ethnopharmacology. 2012;141(3):803-39. https://doi.org/10.1016/j.jep.2012.03.014

28. Choochote W, Chaiyasit D, Kanjanapothi D, Rattanachanpichai E, Jitpakdi A, Tuetun B et al. Chemical composition and anti-mosquito potential of rhizome extract and volatile oil derived from *Curcuma aromatica* against *Aedes aegypti* (Diptera: Culicidae). Journal of Vector Ecology. 2005;30(2):302.

29. Grover M, Behl T, Sehgal A, Singh S, Sharma N, Virmani T et al. *In vitro* phytochemical screening, cytotoxicity studies of *Curcuma longa* extracts with isolation and characterisation of their isolated compounds. Molecules. 2021;26(24):7509. https://doi.org/10.3390/molecules26247509

30. Mehra N, kumar Jain N. Comparative phytochemical screening of *Curcuma angustifolia*, *Curcuma decipiens* and *Curcuma longa* by using GC-MS. Journal of Pharmacognosy and Phytochemistry. 2019;8(2):1227-34.

31. Al-Salih DAAK, Aziz FM, Mshimesh BAR, Jehad MT. Antibacterial effects of vitamin E: *In vitro* study. Journal of Biotechnology Research Center. 2013;7(2):17-23. https://doi.org/10.24126/jobrc.2013.7.2.260

32. Mailafiya MM, Yusuf AJ, Abdullahi MI, Aleku GA, Ibrahim IA, Yahaya M et al. Antimicrobial activity of stigmasterol from the stem bark of *Neocarya macrophylla*. Journal of Medicinal Plants for Economic Development. 2018;2(1):1-5. https://doi.org/10.4102/jomped.v2i1.38

33. Bindu BSC, Mishra DP, Narayan B. Inhibition of virulence of *Staphylococcus aureus*–a food borne pathogen–by squalene, a functional lipid. Journal of functional foods. 2015;18:224-34. https://doi.org/10.1016/j.jff.2015.07.008

34. Ukaegbu-Obi KM, Ifediora AC, Ifediora NH, Chukwu B. *In vitro* combine d antibacterial effect of Turmeric (*Curcuma longa*) and Ginger (*Zingiber officinale*) on some pathogenic organisms. Analele Universităţii din Oradea, Fascicula Biologie. 2016;23 (1):32-36.

35. Kim HJ, Lee JW, Kim YD. Antimicrobial activity and antioxidant effect of *Curcuma longa*, *Curcuma aromatica* and *Curcuma zedoaria*. Korean Journal of Food Preservation. 2011;18(2):219-25. https://doi.org/10.11002/kjfp.2011.18.2.219

36. Chandrasekaran K, Senthilkumar M. Synergic antibacterial effect of *Curcuma aromatica* Salisb. and *Ocimum tenuiflorum* Linn herbal extract combinations on treated cotton knitted fabrics against selective bacterial strains. Indian Journal of Fibre and Textile Research (IJFTR). 2019;44(3):344-51. https://doi.org/10.56042/ijftr.v44i3.21168