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Le Thi Huong, Hoang Vinh Phu, Le Duc Giang, Dao Thi Minh Chau & Isiaka Ajani Ogunwande

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Article

Antimicrobial Activity and Constituents of Essential Oils from the Leaves of *Syzygium szemaoense* Merrill & L.M. Perry and *Syzygium corticosum* (Lour.) Merr. & L.M. Perry grown in Vietnam**Le Thi Huong^{1*}, Hoang Vinh Phu¹, Le Duc Giang², Dao Thi Minh Chau¹, Isiaka Ajani Ogunwande^{3*}**¹ Faculty of Biology, College of Education, Vinh University, 182 Le Duan, Vinh City, Nghe An Province 4300, Vietnam² Faculty of Chemistry, College of Education, Vinh University, 182 Le Duan, Vinh City, Nghe An Province 4300, Vietnam³ University Road, Aleku Area, Osogbo, Nigeria* Corresponding Authors: Le Thi Huong (lehuong223@gmail.com)Isiaka Ajani Ogunwande (isiakaogunwande@gmail.com)

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Abstract: In this study, the leaves of *Syzygium szemaoense* Merrill & L.M. Perry and *Syzygium corticosum* (Lour.) Merr. & L.M. Perry was screened for their essential oils constituents and antimicrobial activities. The main constituent of *S. szemaoense* was *cis*- β -elemene (68.0%), while β -caryophyllene (54.0%) was the most abundant constituent of the essential oil of *S. corticosum*. Both essential oils displayed moderate antimicrobial activity against *Enterococcus faecalis* ATCC 29212 and *Staphylococcus aureus* ATCC 25923 with minimum inhibitory concentration (MIC) value of 128.0 μ g/mL, as well as anti-candidal action towards *Candida albicans* ATCC 10231 with MIC value of 64.0 μ g/mL. The essential oil of *S. corticosum* showed antimicrobial activity towards *Bacillus cereus* ATCC 14579 with MIC value of 128.0 μ g/mL. The chemical compositions and antimicrobial activities of both *S. szemaoense* and *S. corticosum* are being reported for the first time. The essential oils of *S. szemaoense* and *S. corticosum* may be considered for further investigation for antimicrobial uses.

Keywords: Antimicrobial activity, *cis*- β -elemene, β -caryophyllene, essential oil composition, *Syzygium*.**Introduction**

In a continued effort to evaluate the chemical constituents and biological activities of unexploited species from Vietnamese flora¹⁻³, we report the results of investigation on the two plants from *Syzygium* species. *Syzygium szemaoense* Merrill & L. M. Perry is a shrub or tree that grows between 4-8 m tall. The leaf blade is elliptic to narrowly elliptic of 4-10 x

1.7-4 cm dimension⁴. The ridged branches are grayish brown when dry but become brown when old. The flower buds are obovoid about 3.5 mm in diameter. The fruits are purple when ripe and ellipsoid-ovoid in shape about 1-1.5 \times 0.8⁻¹ cm. The plant has one seed per fruit. Flowering occurs between July and August, while fruiting takes place from September to December⁴. Previously, antimicrobial compounds such as

syzygiumursanolides A and B, syzygiumone B and syzygiumursanolide D were isolated from *S. szemaoense* leaves ⁵.

Syzygium corticosum (syn *Eugenia corticosa* Lour.) is an evergreen tree that grows up to 15 m tall. The leaves have thinly leathery leaf blades, while the barks are dark-grey. The flowers are white, and occur in clusters. The fleshy fruits are round 1.8-2.2 cm across, and dark red to purplish-black when ripe ⁶. Each seed is oblong to round. *In vitro* showed that *S. corticosum* leaves possess anticancer activity ⁷. Also, ellagic acid derivatives, cyclohexanone, (+)-fouquierol, (+)-ursolic acid and megastigmanes, were characterized from the leaves of *S. corticosum* ⁸. Furthermore, the investigations showed that (+)-ursolic acid was the major cytotoxic component of *S. corticosum* ⁸. A phenol, trimethylellagic acid, was previously isolated and identified from the leaves and twigs of *S. corticosum* ⁹.

Plants are part of our daily life and essential oils have been extracted from over 3000 different species. These essential oils have domestic, industrial and medicinal uses ¹⁰. Essential oils have an important role in the protection of plants and are well known for their various biological and pharmacological effects including antimicrobial, anti-viral, anti-inflammatory, anti-cancer amongst others ¹⁰. These activities are normally related to the chemical substances mostly terpenes that are present in them. Essential oils are generally recognized as environmental friendly, easily biodegradable, minimally toxic to mammals and have toxicity against different pathogens and insect pests. Essential oils have been used for a long time as natural products.

Recently, essential oils have been extracted from several medicinal plants growing in Vietnam. In addition, their chemical compositions of these essential oils and some biological activities have been studied and reported ¹¹⁻¹³. Moreover, the essential oil compositions of some *Syzygium* plants grown in Vietnam and other parts of the world have been documented and published. The main compounds identified in the leaf and stem of *S. grande* ¹⁴ were β -caryophyllene (25.6% and 29.3%), sabinene (16.8% and 10.2%) and (*E*)- β -ocimene (11.9% and 9.5%) respectively. On the

other hand, α -pinene (35.4%) and (*E*)-nerolidol (30.4%) were the major compounds present in the leaf of *S. sterrophyllum* ¹⁴. The main constituents of *S. hancei* were γ -guaiene (11.07 %) and β -caryophyllene (9.11%), while β -caryophyllene (42.53%) and (*E*)- β -ocimene (19.38%) were identified in the leaf of *S. caryophyllatum* ¹⁵. However, β -caryophyllene (64.53%) was the abundant compound in the leaf of *S. lineatum* ¹⁵. The essential oil of *S. bullockii* leaf consists mainly of (*E*)-caryophyllene (49.5%), while *S. tsoongii* had abundant of (*E*)-caryophyllene (23.40%), bicyclogermacrene 2 (21.23%) and (*Z*)- β -ocimene ¹⁶. Eugenol (52.5% and 76.54%) was the main compound of *S. aromaticum* from Brazil ¹⁷ and Vietnam ¹⁸, respectively. The essential oil of *S. cordatum* ¹⁹ had a compositional pattern dominated by hexahydrofarnesyl acetate (14.4%), and a non-terpene, 2,3-butanediol diacetate (13.3%).

However, the chemical compositions and biological activities of essential oils from the plants of *S. szemaoense* and *S. corticosum* have not been the subject of literature discussion. The aim of the present study was to report for the first time on the chemical compositions and antimicrobial activity of essential oils of both *S. szemaoense* and *S. corticosum* from Vietnam. This is in furtherance of the on-going extensive work with the sole aim of characterization of the compositions and biological of essential oils from Vietnamese plants ^{11-14,20,21}.

Materials and method

Plant materials collection

In Vietnam, the leaves of *S. szemaoense* and *S. corticosum* were collected from Pù Hoạt Nature Reserve with GPS location at 19°35'19"N, 104°43'7"E. The Nature Reserve is situated in NgheAn Province of Vietnam. The collection was done by handpicking. The authors collected both plants from the reserve in February 2022, mainly at an elevation of 280 m. After the collection, the identification of the samples was performed by Dr. Huong LT. Thereafter, individual voucher specimen LTH 928 and LTH 930, respectively, were deposited at Vinh City University, Vietnam.

The hydrodistillation of essential oils from Syzygium plants

The individual plant sample was divided into three equal portions. Each portion was separately packed inside a 5 L flask. Following the procedures described earlier^{11-14,20,21}, distilled water was added to submerge the pulverized sample and the flask was connected to the hydrodistiller (Clevenger-type apparatus) and source of heat. The distillation was allowed to run for 3 h at normal pressure when essential oil distilled off. The essential oils were collected and weighed accordingly as described earlier^{11-14,20,21}. The preservation of the essential oils was done inside refrigerator (4°C) before the instrumental and biological analyses were performed. The distillation of essential oils from each of the plant was conducted three times. The mass (g) of the essential oil was divided by the mass (g) of the each plant to obtain the percentage yield of the essential oils.

Procedure for analysis of essential oil constituents

Conveniently, the instruments of Gas chromatography (GC) and Gas chromatography-mass spectrometry (GC/MS) were used. For both the GC and GC/MS analyses, the GC operating conditions were the same. The Gas chromatograph was a HP 7890A Plus manufactured by Agilent Technologies, USA. Essential oil samples (1.0 µL) were separately injected by splitting method at Inlet pressure of 6.1 kPa. The components and operating conditions are listed as Column: HP-5MS; Column dimension: of 30 m x 0.25 mm, and with film thickness of 0.25; Detector: Flame ionization (FID); Carrier gas and flow rate: He 1 mL/min; Injector temperature: 250°C; and Detector temperature: 260°C. The essential oil was injected into GC column at a split ratio of 10:1. Thereafter, the column temperature programmed from 40°C (held 2 min isothermally) to 220°C (10 min hold) at 4°C/min. Each of the essential oils was analyzed three times. The relative amounts of individual components were calculated based on the GC peak area (FID response) as described in previous studies^{11-14,20,21}.

However, for the GC/MS analysis, the GC

chromatograph was connected with a mass spectrometer HP 5973 MSD. The GC components and conditions were similar as described above. However, the MS conditions were: Ionization voltage: 70 eV; Emission current 40 mA; Acquisitions scan mass range: 45-350 amu; and Sampling rate: 1.0 scan/s.

Modalities for identification of the constituents of the essential oils

In order to identify the components present in the studied essential oils of *S. szemaense* and *S. corticosum*, the procedures described were used and consists of (i) comparison of GC retention indices with reference to a homologous series of *n*-alkanes (C₆-C₄₀); (ii) co-injection with known compounds under the same GC conditions; and (iii) checking the MS fragmentation patterns of the the individual GC/MS spectra with known essential oil composition in literature²² as described recently^{1-3, 11-14,20,21}.

Study of the antimicrobial activity of the essential oils

In the study of the antimicrobial activity of the essential oils of *Syzygium* plants, the following microorganisms were used, namely, *Enterococcus faecalis* ATCC 29212, *Staphylococcus aureus* ATCC 25923, *Bacillus cereus* ATCC 14579, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *Salmonella enterica* ATCC 13076 and *Candida albicans* ATCC 10231. The study involves the measurement of the minimum inhibitory concentration (MIC) and the evaluation of the median inhibitory concentration (IC₅₀) values by using the microdilution broth susceptibility assay. The concentration of the essential oils were prepared by two-fold dilution from 1.6384 x 10⁴ µg/mL to 2 µg/mL. Strains of Gram-positive test bacteria (3), Gram-negative test bacteria (3), and fungi (1) were used in the study of the antimicrobial potential of the essential oils. The bacteria used in the experiment maintained in double-strength Mueller-Hinton broth were standardized to 5 × 10⁵ CFU/mL while the fungi strength was 1 × 10³ CFU/mL, and grown in double-strength Sabouraud dextrose broth as described earlier^{1-3, 11-14,20,21}. Dilute

solutions of essential oils in sterile distilled water and microorganisms were transferred to 96-well microtiter plates. The solutions were allowed to incubate for 24 h, and at temperature of at 37°C. Afterwards, the MIC values were evaluated from the well with the lowest concentration of essential oils which completely inhibited the growth of microorganisms. On the other hand, the IC₅₀ values were measured by considering the percentage of microorganisms that inhibited growth based on the turbidity measurement data of EPOCH2C spectrophotometer. For positive control, standard antimicrobial drugs of Streptomycin (antibacterial), as well as while nystatin and cycloheximide (anticandidal) were used. The last row of the microtiter plates containing only the serial dilutions of the essential oils without microorganisms was used as the negative (no growth) control. The data obtained were calculated as described previously ^{1-3, 11-14,20,21}.

Statistical analysis

The obtained percentages of each of the components of the essential oils, the MIC values and the IC₉₀ values were based on the consideration of the statistical analysis (ANOVA) of the differences between mean values obtained for experimental groups and were calculated as a mean of standard deviation (SD, ±) of three independent measurements using Microsoft excel program 2003.

Results and discussion

Analysis of the compounds identified in the essential oils from the leaves of S. szemaoense and S. corticosum

The average yields of the essential oil of *S. szemaoense* was 0.16% (± 0.01%, v/w), while *S. corticosum* was obtained in yield of 0.12% (± 0.01%, v/w). The essential oils of *S. szemaoense* and *S. corticosum* were light yellow coloured. A consideration of the GC-MS analysis showed that twenty-four and twenty-six compounds with total essential oil contents of 97.6% and 90.4%, respectively, were identified in the the leaves of *S. szemaoense* and *S. corticosum* (Table 1). Both essential oils of the leaves of *S. szemaoense* and *S. corticosum* contained a

large proportions of sesquiterpene hydrocarbons (91.9% and 80.2%, respectively). Oxygenated monoterpene compounds were not identified in the leaves of *S. szemaoense*, while the monoterpene hydrocarbons were conspicuously absent in *S. corticosum*. From the analysed essential oil component, *cis*-β-elemene (68.0%) was the most singly abundant compound of *S. szemaoense*. There are significant amount of bicyclogermacrene (7.7%), β-caryophyllene (4.9%), β-selinene (2.8%) and α-humulene (2.2%). However, in the essential oil of *S. corticosum*, β-caryophyllene (54.0%) was identified as the most abundant constituent. The essential oil of *S. corticosum* also features sizeable quantity of aromadendrene (7.8%), viridiflorene (6.3%), caryophyllene oxide (4.2%), α-humulene (3.8%) and chrysanthenone epoxide (3.0%).

The compositional pattern of the leaf essential oil of *S. corticosum* under investigation looks similar to the composition of essential oils from the leaves of *S. lineatum* ¹⁵, *S. bullock* ¹⁶ and *S. tsoongii* ¹⁶ from Vietnam due to high contents of β-caryophyllene. However, *cis*-β-elemene as seen in *S. szemaoense* essential oil has not featured prominently as the main compound of any of *Syzygium* essential oils as documented in Table 1 and 2.

A variety of terpenoids and non-terpenoid compounds were identified previously from the essential oils of *Syzygium* species reported from all over the world (Table 2) ^{13-19,23-33}. Moreover, majority of *Syzygium* essential oils contained larger amount of sesquiterpenes over monoterpenes. The monoterpene, α-pinene was identified in the leaf of *S. sterrophyllum* from Vietnam ¹⁴, wax of *S. samarangense* ²⁴ from Vietnam and *S. cumini* from Egypt ³⁰. Limonene and γ-terpinene were identified in the leaves of *S. malaccense* from Nigeria ²³. β-Ocimene could be seen prominent in the leaves of *S. nervosum* ¹³, *S. grande* ¹⁴ and *S. caryophyllatum* ¹⁵ from Vietnam. Sabinene in *S. grande* ¹⁴, α-terpineol and allo-ocimene in *S. cumini* ³⁰, were the monoterpene of notable quantities. Among the non-terpenes, the composition of essential oil from the leaves of *S. benthamianum* analysed from India was dominated by sitosterol acetate, stigmastan-

Table 1. The identified compounds of the leaf essential oils of *S. szemaoense* and *S. corticosum*

No.	RT (min)	Compounds ^a	RI ^b	RI ^c	<i>S. szemaoense</i> ^d	<i>S. corticosum</i> ^d
1	10.49	α -Pinene	937	932	0.6	-
2	11.88	β -Pinene	983	978	0.4	-
3	22.89	Chrysanthenone epoxide	1304	1301	-	0.3
4	24.33	δ -Elemene	1347	1351	0.8	-
5	24.74	α -Cubebene	1359	1353	0.2	-
6	25.60	Isoledene	1386	1388	-	0.2
7	25.71	α -Copaene	1388	1389	0.9	-
8	26.26	<i>cis</i> - β -Elemene	1404	1407	68.0	0.2
9	26.95	α -Gurjunene	1429	1431	-	0.1
10	27.24	β -Caryophyllene	1437	1437	4.9	54.0
11	27.74	α -Maaliene	1454	1456	-	0.3
12	27.86	Aromadendrene	1455	1461	-	7.8
13	27.96	Selina-5,11-diene	1458	1458	-	0.2
14	28.31	α -Humulene	1471	1478	2.2	3.8
15	28.54	9- <i>epi</i> -(<i>E</i>)-Caryophyllene	1477	1479	-	1.9
16	28.88	β -Chamigrene	1490	1491	1.1	0.5
17	29.13	Germacrene D	1498	1499	1.3	-
18	29.38	β -Selinene	1503	1501	2.8	1.5
19	29.41	<i>allo</i> -Aromadendr-9-ene	1507	1510	-	0.7
20	29.56	Viridiflorene	1512	1513	-	6.3
21	29.58	α -Selinene	1513	1515	1.1	2.3
22	29.62	Bicyclogermacrene	1514	1517	7.7	-
23	30.08	γ -Cadinene	1528	1534	-	0.4
24	30.29	δ -Cadinene	1532	1536	0.7	0.7
25	30.33	<i>trans</i> -Calamenene	1538	1536	0.2	0.3
26	31.06	Elemol	1567	1571	0.3	-
27	31.66	Dendrolasin	1583	1585	0.7	-
28	32.07	Spathulenol	1596	1597	0.7	0.5
29	32.30	Caryophyllene oxide	1605	1610	0.4	4.2
30	32.54	Cubean-11-ol	1613	1616	-	1.0
31	32.79	Rosifoliol	1622	1626	-	0.2
32	32.88	Ledol	1625	1630	-	0.4
33	33.38	5-Guaioene-11-ol	1643	1644	-	0.4
34	33.46	1- <i>epi</i> -Cubenol	1644	1646	0.4	-
35	33.80	<i>epi</i> - α -Cadinol	1656	1656	-	0.5
36	34.21	α -Cadinol	1671	1672	0.4	0.2
37	34.30	(<i>E</i>)-Bisabol-11-ol	1675	1675	-	0.7
38	34.32	<i>neo</i> -Intermedeol	1676	1678	1.3	-
39	34.65	<i>trans</i> - α -(<i>Z</i>)-Bergamotol	1678	1690	0.3	-
40	34.73	14-Hydroxy-9- <i>epi</i> - α -caryophyllene	1691	1700	-	0.2
		Total			97.6	90.4
		Monoterpene hydrocarbons (Sr. No. 1,2)			1.0	-
		Oxygenated monoterpenes (Sr. No. 3)			-	3.0
		Sesquiterpene hydrocarbons (Sr. No. 4-25)			91.9	80.2
		Oxygenatedsesquiterpenes (Sr. No. 26-40)			4.7	7.2

^a Elution order on HP-5MS column; ^b Retention indices on HP-5MS column; ^c Literature retention indices on HP-5MS column NIST [16]; ^d Standard deviation were insignificant and excluded; RT, Retention times; - Not identified

Table 2. Compositional patterns of essential oils from some *Syzygium* plants around the world

Species	Parts	Origin	Main constituents	References
<i>S. nervosum</i>	Leaves	Vietnam	(Z)- β -ocimene (20.3%), caryophyllene oxide (13.2%) and (E)-caryophyllene (12.1%)	13
<i>S. grande</i>	Leaves/ Stem	Vietnam	β -caryophyllene (25.6% and 29.3%), sabinene (16.8% and 10.2%) and (E)- β -ocimene (11.9% and 9.5%)	14
<i>S. sterrophyllum</i>	Leaves	“	α -pinene (35.4%) and (E)-nerolidol (30.4%)	14
<i>S. hancei</i>	“	“	γ -guaiaene (11.07 %) and β -caryophyllene (9.11%)	15
<i>S. caryophyllatum</i>	“	“	β -caryophyllene (42.53%) and (E)- β -ocimene (19.38%)	15
<i>S. lineatum</i>	“	“	β -caryophyllene (64.53%)	15
<i>S. bullockii</i>	“	“	(E)-caryophyllene (49.5%)	16
<i>S. tsoongii</i>	“	“	(E)-caryophyllene (23.40%), bicyclogermacrene (21.23%) and (Z)- β -ocimene (10.61%)	16
<i>S. aromaticum</i>	Bud	Brazil	eugenol (52.5%)	17
“	“	Vietnam	eugenol (76.54%)	18
<i>S. cordatum</i>	Leaves	“	hexahydrofarnesyl acetate (14.4%), 2,3-butanediol diacetate (13.3%)	19
<i>S. samarangense</i>	Leaves	Nigeria	α -cadinol (12.7%), juniper camphor (8.2%)	23
<i>S. aromaticum</i>	“	“	eugenol (76.5%)	23
<i>S. malaccense</i>	“	“	limonene (48.8%) and γ -terpinene (26.2%)	23
<i>S. samarangense</i>	Wax	Vietnam	α -pinene (14.02%), o-cymene (13.47%), α -cubebene (21.49%), epizonarene (13.10%) ^a β -gurjunene (10.73%), α -selinene (20.11%) and caryophyllene oxide (15.02%) ^a	24
<i>S. malaccense</i>	Leaves	Brazil	aristolochene (20.06%), γ -himachalene (16.5%), δ -amoprhene (11.79%) ^a , myltayl-4(12)-ene (11.43%), viridiflorol (10.67%) and (2E,6E)-farnesyl acetate (10.10%) ^a	25
<i>S. jambos</i>	“	“	β -caryophyllene (17.67%), (E,E)- α -farnesene (16.10%), caryophyllene alcohol (11.45%), and α -humulene (10.66%)	25
<i>S. cumini</i>	“	“	β -caryophyllene (37.65%) and α -humulene (18.37%)	25

table 2. (continued).

Species	Parts	Origin	Main constituents	References
<i>S. benthamianum</i>	''	India	sitosteryl acetate (11.83%), stigmastan-3,5,22-triene (7.0%), 2,6-dimethyl-2-octene (6.99%)	26
<i>S. myrtifolium</i>	''	''	δ-cadinol (29.53 %), caryophyllene oxide (26.25 %)	27
<i>S. kararensis</i>	''	''	seychellene (7.3%), α-muurolol (5.4%), <i>cis</i> -cadinene ether (5.3%), β-vetivenene (5.1%)	28
<i>S. polyanthum</i>	''	Malaysia	<i>cis</i> -4-decanal (43.489%), 1-decyl aldehyde (19.752%), and capryl aldehyde (14.092%)	29
<i>S. aromaticum</i>	''	''	p-eugenol (75.190%) and β-caryophyllene (18.364%)	29
<i>S. cumini</i>	''	Egypt	α-pinene (17.53%), α-terpineol30 (16.67%) and <i>allo</i> -ocimene (13.55%) ³⁰	30
<i>S. aromaticum</i>	Seeds	Mexico	eugenol (77.322%) and caryophyllene (16.77%)	31
<i>S. guineense</i> var. <i>guineense</i>	Leaves	Benin	α-cadinol (12.7%), <i>cis</i> -calamenen-10-ol (14%), citronellyl pentanoate (15.2%) ^a β-caryophyllene (20.1%) and α-humulene (39.5%) ^a	32
<i>S. aromaticum</i>	''	Brazil	eugenol (82.4%) and β-caryophyllene (12.6%)	33

^a, multiple analysis

3,5,22-triene and 2,6-dimethyl-2-octene ²⁶, while *S. polyanthum* leaf oil from Malaysia had abundance of *cis*-4-decanal, 1-decyl aldehyde, and capryl aldehyde ²⁹. 2,3-Butanediol diacetate was the main compound in the leaf of *S. cordatum* from Vietnam ¹⁹.

It was well observed that β -caryophyllene among the sesquiterpenes has featured prominently in the compositional pattern of several essential oils of several *Syzygium* plants. For example, the compositional pattern of essential oils of *S. aromaticum* from all over the world consists mainly of eugenol and caryophyllene ^{17,18, 23,29,31,33}. A mixture of β -caryophyllene and α -humulene were observed in *S. cumini*²⁵ and *S. jambos* from Brazil, as well as *S. guineense* var. *guineense* from Benin ³². It also occurred in large quantity along with β -ocimene in the leaves of *S. nervosum* ¹³, the leaves and stems of *S. grande* ¹⁴ and the leaves of *S. caryophyllatum* ¹⁵ from Vietnam. This sesquiterpene also occurred along with γ -guaiene in *S. hancei* from Vietnam ¹⁵.

The analysis of the constituents showed that the essential oils from *Syzygium* plants consist mainly of terpenes of different structural patterns and percentages. This may be considered as the existence of chemical variability in the essential oils of the studied *Syzygium* plants. It is well known that factors such as the age and morphology of the plant, plant parts, time of collection and handling procedures

may contribute greatly to the observed variations and differences in chemical contents of the essential oils of *S. szemaoense* and *S. corticosum* and invariably, the *Syzygium* plants. This will invariably affects the compositions and biological activity of the same species due to differences in the environmental and ecological conditions at the site of the collection of the plants.

Observed antimicrobial activity of the essential oils of *S. szemaoense* and *S. corticosum*

The analysis of results of the antimicrobial test on the leaf essential oils of *S. szemaoense* and *S. corticosum* indicate that both essential oils displayed moderate antimicrobial activity against three Gram-positive tested microorganisms and anti-candidal activity towards the fungi, with almost similar minimum inhibitory concentration (MIC) values (Table 3). The essential oils of *S. szemaoense* and *S. corticosum* showed antimicrobial activity against *E. faecalis* ATCC 29212 and *S. aureus* ATCC 25923, with MIC value of 128.0 μ g/mL. However, *S. corticosum* exhibited activity towards *Bacillus cereus* ATCC 14579 at lower MIC value (128.0 μ g/mL) than *S. corticosum* (MIC, 256.0 μ g/mL) indicating a greater activity. In addition, both essential oils showed similar anti-candidal activity *C. albicans* ATCC 10231 with MIC value of 64.0 μ g/mL. The observed IC₅₀ values falls within the reported range of 10-220 μ g/mL reported previously for essential oils from other plant species ^{1-3, 17,18}.

Table 3. Data on the antimicrobial activity of the leaf essential oils of *S. szemaoense* and *S. corticosum*

Microorganisms	MIC (μ g/mL) ^a		IC ₅₀ (μ g/mL) ^a	
	<i>S. szemaoense</i>	<i>S. corticosum</i>	<i>S. szemaoense</i>	<i>S. corticosum</i>
<i>Enterococcus faecalis</i> ATCC 29212	128.0 \pm 0.01 ^b	128.0 \pm 0.50 ^c	42.8 \pm 0.21	45.2 \pm 0.16
<i>Staphylococcus aureus</i> ATCC 25923	128.0 \pm 0.00 ^d	128.0 \pm 0.61 ^c	39.6 \pm 0.50	45.7 \pm 0.32
<i>Bacillus cereus</i> ATCC 14579	256.0 \pm 0.10 ^f	128.0 \pm 0.50 ^f	109.0 \pm 1.00	45.6 \pm 0.50
<i>Pseudomonas aeruginosa</i> ATCC 27853	na	na	nt	nt
<i>Escherichia coli</i> ATCC 25922	na	na	nt	nt
<i>Salmonella enterica</i> ATCC 13076	na	na	nt	nt
<i>Candida albicans</i> ATCC 10231	64.0 \pm 0.21 ^g	64.0 \pm 0.12 ^g	23.1 \pm 0.01	21.3 \pm 0.00

^a Mean value of three replicate assays; ^{b,c,d,e,f} Streptomycin, MIC values of 0.21 μ g/mL, 1.07 μ g/mL, 1.16 μ g/mL, 2.20 μ g/mL, and 0.18 μ g/mL, respectively; ^g Cycloheximide, MIC of 0.46 μ g/mL; na, No activity; nt, Not tested

The obtained MIC and IC₅₀ values are indication that the leaf essential oils of *S. szemaoense* and *S. corticosum* exhibited moderate antimicrobial and anti-candidal activities against the tested microorganisms. The essential oils, however, did not inhibit the growth of Gram-negative microorganisms of *P. aeruginosa* ATCC 27853, *S. enterica* ATCC 13076 and *E. coli* ATCC25922. This is an indication that the studied essential oils of *S. szemaoense* and *S. corticosum* specifically inhibited the growth of Gram-positive bacteria. This showed that the studied essential oils possessed moderate antimicrobial activity against the tested microorganisms when compared with the standard. Further purification may improve the antimicrobial potentials of the essential oils. The results in this study validate the medicinal potentials and especially the antimicrobial action of *Syzygium* species from all over the world. Previously, the leaf essential of *S. aromaticum* from Vietnam was active against *B. cereus*, *S. aureus*, *E. coli*, *S. typhimurium* and *P. aeruginosa* among other¹⁸. The essential oil of *S. nervosum* only showed antibacterial activity to *E. faecalis* with (MIC value 32 µg/mL) and anti-candidal property towards *C. albicans* (MIC 128 µg/mL)¹³, while *S. samarangense* only showed pronounced activity towards *S. aureus*¹⁸. Also, *S. aromaticum* oil from Brazil exhibited potential antibacterial activity¹⁷.

In addition, leaf oils of *S. aromaticum*, *S. malaccense* and *S. samarangense* from Nigeria displayed antibacterial potentials against several microbes, but not *Kiebsiella* spp., *Proteus* spp., *Pseudomonas* spp. and *Mucor mucelo*²³. Both essential oils of *S. aromaticum* and *S. polyanthum* from Malaysia strongly inhibited *B. subtilis* growth, while the essential oil of *S. aromaticum* showed a stronger inhibitory activity against *S. aureus*, *Salmonella typhimurium* and *Vibrio cholera* than that of *S. polyanthum*. Both essential oils did not inhibit the growth of *E. coli*²⁹. The oil of *S. cumini* from Egypt demonstrated strong inhibition activity against the some tested bacterial strains³⁰.

The major and minor constituents of essential oil *S. szemaoense* and *S. corticosum* have shown antimicrobial activities against panels of

pathogens. This shows that some compounds of the essential are likely to be responsible for the observed seemingly antimicrobial activity of *S. szemaoense* and *S. corticosum*. The sesquiterpene, β-caryophyllene demonstrated selective antibacterial activity against *S. aureus*³⁴ and antifungal effect³⁵. In addition, some other compounds identified in the essential oils, including δ-cadinene³⁶ and germacrene D^{37,38} were previously reported to exhibit some antibacterial potency. *cis*-β-Ocimene was once described as the component responsible for the inhibitory activity of an essential oil against *S. aureus*³⁹. In light of this, essential oils may represent a valid alternative to avoid the multidrug resistance of many pathogens, or they could be used in combination with antimicrobials to improve their effectiveness against different infectious diseases.

Conclusion

This study showed that the hydrodistilled essential oils of *S. szemaoense* and *S. corticosum* harvested from Pù Hoạt Nature Reserve, NgheAn Province, Vietnam exhibited antimicrobial activity against Gram-positive microorganisms, and anti-candidal activity towards a fungi. The sesquiterpenes, *cis*-β-elemene (68.0%) and β-caryophyllene (54.0%) were respectively the most abundant constituents in *S. szemaoense* and *S. corticosum*. Both essential oils showed similar anti-candidal activity *C. albicans* ATCC10231 with MIC value of 64.0 µg/mL. The essential oils of *S. szemaoense* and *S. corticosum* showed antimicrobial activity against *E. faecalis* ATCC29212 and *S. aureus* ATCC25923, with MIC value of 128.0 µg/mL, while *S. corticosum* exhibited activity towards *B. cereus* ATCC14579 at MIC value of 128.0 µg/mL. These results give significant information about the volatile constituents of the essential and pharmacological activity of the essential oil, which suggest the benefits o human health, having the potential to be used as antimicrobial agents.

Competing interests

The authors declare that no competing interest exists.

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