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**Essential Oils from *Syzygium grande* (Wight) Walp.
and *Syzygium sterrophyllum* Merr. et Perry**

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Abstract: In the present study, essential oils obtained by hydrodistillation from *Syzygium grande* (Wight) Walp. (syn. *Eugenia grandis* Wight) and *Syzygium sterrophyllum* Merrill & L. M. Perry were characterised by gas chromatography (GC) and gas chromatography coupled with mass spectrometry (GC-MS). The main compounds identified in the leaf and stem of *S. grande* 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*. This is the first report on the volatile constituents of the stem of *S. grande* and the leaf of *S. sterrophyllum* from Vietnam.

Keywords: *Syzygium grande*, *Syzygium sterrophyllum*, monoterpenes, sesquiterpenes.

Introduction

Syzygium grande (Wight) Walp. (syn. *Eugenia grandis* Wight) is a species that belongs to the family Myrtaceae. *Syzygium grande* a large tree up to 30 m tall and 80 cm in diameter is cultivated in Vietnam as a roadside ornamental tree. It has simple, opposite, stalked leaves that are broadly elliptic, about 16 cm long and 9 cm wide¹. The leaves are darker green above, lighter green below, and each has a down-turned leaf tip. The flowers are up to 3 cm across, white, and sessile. Each flower has many stamens about 10 mm long. It produces fleshy fruits that are ellipsoid, about 4 cm long and 3 cm wide that are green when ripe. Each fruit has a prominent 6 mm diameter apical

calyx rim and one seed. Traditionally, *S. grande* was used to treat diabetic related complications². Extracts from *S. grande* were shown to possess antimicrobial and antioxidant potentials³. The phytochemical compounds isolated from *S. grande* include myricetin, 4'-methylether-3-*O*- β -d-xylopyranoside, grandoside⁴, 2- α ,3- β -dihydroxylup-12-en-28-oic acid, 3- β -hydroxylup-12-en-28-oic acid, friedelin, 3- β -beta-friedelinol, betulinic acid, oleanolic acid, β -sitosterol⁵ and 1-*O*-galloyl castalagin⁶. Compounds previously isolated from *S. grande* were known to exhibit biological activities such as antifungal, antibacterial, anti-leishmania, DPPH radical-scavenging and cytotoxic activities⁴. The main chemical compounds iden-

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tified in the only report on the essential oil of the leaves of *S. grande* were β -caryophyllene (18.38 %), 10s,11s-himachala-3(12),4-diene (12.06 %), (+)-aromadendrene (10.51 %), α -caryophyllene (10.22 %) and α -selinene (8.94 %) ⁷. The essential oils of *S. grande* possessed antimicrobial ⁷ and antioxidant ⁸ activities.

Syzygium sterrophyllum Merrill & L. M. Perry is a shrub or tree 1 m high with sticky and angular twigs which turns dark brown when dry and old. The leaf blade is thin and leathery 8-12 cm long and 3-5 cm wide. The flowers are white, apex acute and acute, pointed long less than 1 cm, base broadly cuneate or obtuse, above dry after the gray, not shiny, Below the brown, lateral veins separated by about 1.5 mm, 80 degrees open angle slowly oblique to the edge, edge from the edge of about 1 mm, both in the upper and lower sides are slightly raised; petiole 4-6 mm long. The bluish black fruit line axillary and covered with 1-1.5 mm calyx. Flowering occurs in June to October while fruiting takes place between November and January ⁹. No information is readily available on chemical constituents and biological activity of *S. sterrophyllum*. The aim of this paper was to describe the chemical compounds identified in the essential oils of *S. grande* and *S. sterrophyllum* grown in Vietnam as done previously for other plant samples as they are made available ¹⁰⁻¹².

Materials and methods

Collection and authentication of plants

The leaves and stems of *S. grande* and leaf of *S. sterrophyllum* were collected from Pù Mát National Park, Nghe An Province, Vietnam, in May 2013. Dr. Dai carried out the botanical identification of the plants. Voucher specimens NVH 316 and NVH 289 respectively have been deposited at the Botany Museum, Vinh University, Vietnam.

Preparation of plant samples

Prior to hydrodistillation process, plant samples were air-dried under laboratory shade (36°C) for few weeks to reduce the moisture contents. In addition, sediments and other unwanted materials were separated from the samples. Afterwards, samples were pulverized to coarse powder.

Hydrodistillation of essential oils from plants

Aliquots of 500 g of the pulverized sample were carefully introduced into a 5 L flask and distilled water was added until it covers the sample completely. Essential oils were obtained by separate hydrodistillation which was carried out in an all glass Clevenger-type distillation unit designed according to the established specification ¹³. The distillation time was 3 h and conducted at normal pressure. The volatile oils were distilled over water and collected in the receiver arm of the apparatus into clean and previously weighed sample bottles. The oils were kept under refrigeration (4°C) until the moment of analyses as described previously ¹⁰⁻¹².

Gas chromatography (GC) analysis of essential oils

The GC analysis of essential oils was carried out using an Agilent Technologies HP 6890 Plus GC which was equipped with a flame ionization detector and HP-5MS column. The dimension of the column is 30 m x 0.25 mm (film thickness 0.25 μ m). The GC operating parameters based on temperature programming were as follows: a column oven 40°C, injection pot 250°C while the detector temperature was 260°C. Time programming: 40°C for 2 min, temperature and then raise to 220°C (and held isothermally for 10 min) at 4°C/min. The carrier gas used was H₂ at a flow rate of 1 mL/min. The split ratio was 10:1 while 1.0 μ L of the essential oil was injected into the GC at inlet pressure was 6.1 kPa. Each analysis was performed in triplicate. Retention indices (RI) value of each component was determined relative to the retention times of a homologous *n*-alkane series (C₄-C₃₂), under the same operating conditions, with linear interpolation on the HP-5MS column as described previously ¹⁰⁻¹². The relative amounts of individual components were calculated based on the GC peak area (FID response) without using correction factors.

Gas chromatography-Mass spectrometry (GC-MS) analysis of essential oils

GC-MS was performed on HP 5973 MSD Mass spectrometer coupled with HP 6890N Plus GC system fitted with a fused silica capillary HP-5 MS column (30 m x 0.25 mm, film thick-

ness 0.25 μm). The conditions were the same as described above for GC with He (1 mL/min) as carrier gas. The MS conditions were as follows: ionization voltage 70 eV; emission current 40 mA; acquisitions scan mass range of 50-550 amu at a sampling rate of 1.0 scan/s as described previously¹⁰⁻¹².

The spectra were scanned from m/z 50 to 550. Most constituents were identified by gas chromatography by comparison of their retention indices with those in the literature or with those of available authentic compounds. The retention indices were determined in relation to a homologous series of *n*-alkanes (C_8 - C_{24}) obtained under the same operating conditions. Further identification was made by comparison of their mass spectra with those stored in the library¹⁴, peak enrichment on co-injection with authentic standard wherever possible as described previously¹⁰⁻¹².

Results and discussion

The yield of essential oils were 0.15 % (leaf), 0.12 % (stem) (v/w, *S. grande*) and 0.16 (v/w *S. sterrophyllum*) calculated on a dry weight basis. The hydrodistillation process afforded light yellow coloured essential oils. The main volatile compounds were displayed in Table 1, along with their percentages and retention indices calculated on HP-5MS column. In *S. grande*, 22 and 44 representing 91.4 % and 88.4 % of the total oil contents were identified respectively in the leaf and stem. Monoterpene hydrocarbons (35.0 % and 28.3 %) and sesquiterpene hydrocarbons (52.9 % and 54.0 %) were the main classes of compounds present in the leaf oil of *S. grande* (Table 1). The major constituents of the leaf oil were β -caryophyllene (25.6 %), sabinene (16.8 % and (*E*)- β -ocimene (11.9 %). Some of the minor compounds identified in the oil were α -copaene (5.0 %), γ -cadinene (4.9 %), α -humulene (4.2 %), (*E,E*)- α -farnesene (3.7 %) and δ -cadinene (3.5 %). Although β -caryophyllene and aromadendrene were identified as main compounds in the oil in agreement with the previous study⁷, some other compounds such as 10s,11s-himachala-3(12),4-diene and α -caryophyllene were conspicuously absent while the amount of α -selinene in the present sample was much lower than previously reported.

Moreover, sabinene and (*E*)- β -ocimene were not reported to be of major significant in the previous study. In addition, β -caryophyllene (29.3 %), sabinene (10.2 %) and (*E*)- β -ocimene (9.5 %) were the main constituents of the stem oil of *S. grande*. There were significant quantity of δ -cadinene (6.6 %) and α -gurjunene (5.1 %). Although there seems to be quantitative similarity in the major compounds of the leaf and stem of *S. grande*, the present study represent the first attempt at characterization of the volatile compounds in the stem oil. The contents of caryophyllene, *cis*- β -ocimene and δ -cadinene confer similarity in compositions of *S. grande* and *S. makul*¹⁵.

On the other hand, 38 compounds amounting to 97.1 % of the oil contents were identified in the leaf of *S. sterrophyllum*. The main classes of compounds present in the oil were monoterpene hydrocarbons (42.0 %), sesquiterpene hydrocarbons (10.4 %) and oxygenated sesquiterpenes (41.7 %). The oil contained large quantity of α -pinene (35.4 %) and (*E*)-nerolidol (30.4 %) while the minor compounds were found to be globulol (4.8 %), limonene (3.8 %) α -cedrol (3.2 %) and farnesol (2.0 %). This is the first report on the volatile constituents of the stem of *S. grande* and the leaf of *S. sterrophyllum*.

The chemical compositions and biological activities of several *Syzygium* essential oils have been published. It was reported previously that 3-caryophyllene (20.31 %), *cis*- β -ocimene (11.23 %) and δ -cadinene (11.02 %) were the major constituents of the leaf of *S. makul*¹⁵. The buds of *S. aromaticum* was characterized by myrtenone (49.1 %) and eugenol (27.1 %) ¹⁶ while the leaf essential was dominated by eugenol (83.02 %) which contributed positively to its antimicrobial activity¹⁷. A previous study revealed that limonene (21.8 %) and globulol (16.0 %) were the major components of *Syzygium cordatum*¹⁸. Caryophyllene oxide (49.6 %) was the major constituent in *S. gardneri* which displayed antimicrobial activity¹⁹ and *S. densiflorum* contained abundance of β -maaliene (17.43 %), isodene (12.46 %) and α -gurjunene (10.44 %) ²⁰. The leaf oil of *S. benthamianum* however showed compositional pattern in which sitosterol acetate (11.83 %), stigmastan-3,5,22-triene (7.0 %) and

Table 1. Chemical constituents of essential oils of *S. grande* and *S. sterrophyllum*

Compounds ^b	RI ^c	RI ^d	Percent composition ^a		
			<i>S. grande</i> L	<i>S. sterrophyllum</i> S	<i>S. sterrophyllum</i> L
α -Pinene	939	932	0.5	0.5	35.4
Camphene	953	946	-	-	0.6
Sabinene	976	962	16.8	10.2	-
β -Pinene	980	978	-	-	0.2
β -Myrcene	990	988	0.6	1.2	0.1
α -Phellandrene	1006	1004	2.4	0.2	-
α -Terpinene	1017	1014	-	0.3	-
<i>p</i> -Cymene	1026	1022	-	0.3	0.7
Limonene	1032	1030	-	2.3	3.8
(<i>Z</i>)- β -Ocimene	1043	1034	0.5	0.7	-
(<i>E</i>)- β -Ocimene	1052	1044	11.9	9.5	0.4
γ -Terpinene	1061	1056	-	0.7	0.5
α -Terpinolene	1090	1089	2.3	1.9	0.3
Linalool	1100	1100	-	-	1.6
Fenchyl alcohol	1120	1121	-	-	0.1
<i>allo</i> -Ocimene	1128	1128	-	0.5	-
Pinocarvone	1165	1161	-	-	0.1
Citronella	1143	1148	-	0.5	-
Borneol	1167	1167	-	-	0.2
Terpinene-4-ol	1177	1177	-	0.2	0.1
α -Terpineol	1189	1187	-	-	0.6
<i>cis</i> -Verbenone	1205	1206	-	-	0.1
Nerol	1234	1229	-	-	0.1
(<i>Z</i>)-Citral (Neral)	1250	1249	-	0.2	-
(<i>E</i>)-Citral (Geranial)	1270	1273	-	0.2	-
Bicycloelemene	1327	1337	0.2	0.4	-
<i>cis</i> -Jasmone	1369	1372	-	-	0.1
α -Copaene	1377	1374	5.0	-	0.6
β -Elemene	1391	1389	-	-	0.1
α -Gurjunene	1403	1408	4.0	5.1	-
β -Caryophyllene	1419	1417	25.6	29.3	0.4
β -Gurjunene	1434	1432	-	0.4	-
γ -Elemene	1437	1437	0.8	0.3	-
Aromadendrene	1441	1439	-	1.4	0.4
α -Humulene	1454	1452	4.2	0.5	0.4
α -Patchoulene	1457	1457	-	0.4	-
γ -Gurjunene	1477	1477	-	0.5	-
α -Curcumene	1482	1483	-	-	0.1
Germacrene D	1485	1484	-	0.4	-
α -Amorphene	1485	1485	-	0.4	0.3
β -Selinene	1486	1486	-	-	0.4
<i>epi</i> -Bicyclosesqui- phellandrene	1489	1482	-	-	0.7

table 1. (continued).

Compounds ^b	RI ^c	RI ^d	Percent composition ^a		
			<i>S. grande</i> L	<i>S. sterrophyllum</i> S	<i>S. grande</i> L
Zingiberene	1494	1493	-	0.7	-
Cadina-1,4-diene	1496	1496	-	0.5	-
α -Selinene	1498	1494	1.0	0.4	-
Bicyclogermacrene	1500	1500	-	-	1.5
α -Muurolene	1500	1501	-	0.5	-
β -Bisabolene	1506	1506	-	0.9	0.6
(<i>E,E</i>)- α -Farnesene	1508	1505	3.7	0.3	-
γ -Cadinene	1514	1513	4.9	2.3	-
Cadina-4,9-diene	1523	1522	-	1.6	-
δ -Cadinene	1525	1525	3.5	6.6	0.9
Calacorene	1546	1541	-	1.1	0.2
Germacrene B	1561	1556	-	-	0.8
(<i>E</i>)-Nerolidol	1563	1561	1.0	1.4	30.4
Spathulenol	1578	1577	0.4	1.0	1.1
Caryophyllene oxide	1583	1581	-	0.7	-
Globulol	1585	1586	0.4	-	4.8
Viridiflorol	1593	1591	-	0.8	-
Guaiol	1601	1600	-	0.2	0.2
α -Cedrol	1601	1602	-	-	3.2
Bulnesol	1672	1666	-	0.9	-
Farnesol	1748	17147	-	-	2.0
Phytol	2125	2119	0.8	-	-
Octadecanoic acid	2200	2188	0.9	-	-
Total			91.4	88.3	97.1
Monoterpene hydrocarbons			35.0	28.3	42.0
Oxygenated monoterpenes			-	1.0	3.0
Sesquiterpene hydrocarbons			52.9	54.0	10.4
Oxygenated sesquiterpenes			1.8	3.0	41.7
Diterpenes			0.8	-	-
Non-terpenes			0.9	-	-

^aStandard deviation (SD \pm) were insignificant and were excluded from the Table

^bElution order on HP-5MS column

^cRetention indices on HP-5MS column

^dLiterature retention indices

- =Not identified

L= leaf

S = stem

2,6-dimethyl-2-octene (6.99 %) predominates ²¹. The analysis of the leaf oil of *S. malaccense* showed p-cymene (13.5 %), β -caryophyllene (9.0 %) and β -pinene (8.0 %) as the major compo-

nent ²². It has been observed that non-terpene compounds of 6,10,14-trimethylpentadecane-2-one (14.4 %) and 2,3-butanediol diacetate (13.3 %) were identified in *S. cordatum* ²³. Eugenol

(74.3 %) in the leaf as well as eugenol (49.7 %), caryophyllene (18.9 %) and benzene,1-ethyl-3-nitro (11.1 %) in the bud were the significant compounds of *S. caryophyllatum*²⁴. The leaf oil of *S. cumini* contained α -pinene (32.32 %), β -pinene (12.44 %) and *trans*-caryophyllene (11.19 %)²⁵ while eugenol (23.52 %), γ -eudesmol (14.85 %), β -pinene (15.12 %), β -caryophyllene (12.03 %), and linalyl acetate (10.23 %) were present in antimicrobial *S. mundagam* buds²⁶.

It could be seen each plant has its own compositional pattern that are different from each other, although ubiquitous terpene compounds predominate in most cases. This variation may be explained in view of ecological and climatic conditions between the various places where plant samples were collected for analysis as well

as the age of the plant and the handling procedures.

Conclusions

The chemical composition of essential from the leaf and stem of *S. grande* and the leaf of *S. sterrophyllum* were characterised and reported for the first time. The dominant compounds identified in the essential oils were β -caryophyllene, sabinene, (*E*)- β -ocimene, α -pinene and (*E*)-nerolidol. The compositional patterns of the essential oils were found to vary from those of other species in the genus. However, the contents of caryophyllene, *cis*- β -ocimene and δ -cadinene may confer similarity in compositions of *S. grande* to those of *S. makul*.

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