DOI: 10.15625/2615-9023/16983

CHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITY OF THE ESSENTIAL OIL FROM *Magnolia balansae* A. DC. GROWING IN VIETNAM

Chu Thi Thu Ha[1,2,*], Tran Huy Thai[1], Nguyen Thi Hien[1], Le Ngoc Diep[1], Dinh Thi Thu Thuy[3]

[1]Institute of Ecology and Biological Resources, VAST, Vietnam
[2]Graduate University of Science and Technology, VAST, Vietnam
[3]Institute of Natural Product Chemistry, VAST, Vietnam

Received 13 March 2022; accepted 6 September 2022

ABSTRACT

Essential oils of *Magnolia balansae* A. DC. growing wild in Son La (leaf, twig and fruit) and Phu Tho (fruit) provinces of Vietnam were obtained by hydrodistillation and were analyzed using GC/MS-FID. From 30 to 49 compounds were identified accounting for 66.1–80.0% of the oils. Spathulenol (33.9%, 20.7%, and 11.2%), and caryophyllene oxide (10.0%, 10.1%, and 9.3%) were the main components of leaf, twig and fruit oils of *M. balansae,* respectively, from Son La province. Linalool (13.8%) was also the main component of fruit oil. While, in the fruit oil of *M. balansae* from Phu Tho province, *cis-β*-elemene (10.4%) was the sole main component. The antimicrobial activity of the oils was tested against 3 microorganism strains using an agar disk diffusion method with inhibitory zone diameters ranging from 13.5 to 40 mm. IC_{50} and MIC of the *M. balansae* oils were determined using a microdilution broth susceptibility assay against 7 microorganism strains. Among them, *Bacillus subtilis* had the highest sensitivity with IC_{50} values (ranging from 57.0 to 82.0 µg/mL) and MIC value (512 µg/mL). This is the first study on the chemical composition of essential oils of *M. balansae* and their antimicrobial activity.

Keywords: Magnoliaceae, *Magnolia balansae*, essential oil composition, antimicrobial activity.

Citation: Chu Thi Thu Ha, Tran Huy Thai, Nguyen Thi Hien, Le Ngoc Diep, Dinh Thi Thu Thuy, 2022. Chemical composition and antimicrobial activity of the essential oil from *Magnolia balansae* A. DC. growing in Vietnam. *Academia Journal of Biology*, 44(3): 11–21. https://doi.org/10.15625/2615-9023/16983

**Corresponding author email*: hachuthi@yahoo.com

INTRODUCTION

Magnolia balansae A. DC. (syn. *Michelia balansae* (A. DC.) Dandy, *Michelia balansae* var. *appressipubescens* Y.W.Law, *Michelia balansae* var. *brevipes* B.L.Chen, *Michelia baviensis* Finet & Gagnep.) belonging to the family of Magnoliaceae Juss. has Vietnamese names as Gioi ba, Gioi long. *M. balansae* can be up to 10–15 m high and 60 cm d.b.h. The bark is gray to grayish brown and smooth. The young branches, vegetative buds, leaf blade abaxial surfaces, flower buds, and brachy blasts have densely dark reddish-brown tomentose or with appressed fine trichomes. Flowers are fragrant. Flowering in March–July, fruiting in June–October. *M. balansae* is distributed in some provinces of China and Vietnam, usually in humid and fertile soil areas, along rivers, in evergreen broad-leaved forests, at 200–1.100 m a.s.l. (Pham, 1999; Nguyen, 2003; Vu, 2011; Tu et al., 2014). In Vietnam, *M. balansae* was ranked at the level of vulnerable - VU A1c,d (MOST & VAST, 2007).

The previous studies on *M. balansae* focused on pollen morphology and ultrastructure (Xu & Kirchoff, 2008), propagation (Nguyen et al., 2012), influence of leaf extract on the activity of cholinesterase from pig serum (Chen et al., 2013), distribution survey and conservation situation evaluation (Nguyen et al., 2015), conservation carrying out (Nguyen, 2016), and the complete chloroplast genome sequence (Sima et al., 2021). A study on the chemical compositions of flower essential oil of *M. balansae* in China indicated that essential oil consisted of two main constituents: ethyl hexanoate (61.0%) and limonene (15.7%) (Zhu, 1993). Another study on the leaf oil of *M. balansae* collected in Pu Mat National Park, Nghe An province of Vietnam showed that the oil consisted of three main constituents: α-pinene (18.4%), α-phellandrene (17.4%), and germacrene D (18.0%) (Nguyen et al., 2005). The present study reports on the chemical composition and antimicrobial activity of essential oils of *M. balansae* growing in Son La province (leaf, twig and fruit) and Phu Tho province (fruit) of Vietnam.

MATERIALS AND METHODS

Plant material: Fresh leaves, twigs and mature fruits of *M. balansae* were collected in July 2019 at Chieng Son commune, Moc Chau district, Son La province. Fresh mature fruits of the same species were collected in June 2017 at Xuan Son commune, Thanh Son district, Phu Tho province of Vietnam. Botanical identification was performed individually by Vu Quang Nam, Vietnam National University of Forestry, Ha Noi and Trinh Ngoc Bon, Vietnamese Academy of Forest Sciences, Ha Noi. Voucher specimens (SL1917 & PT1701) were deposited at the Herbarium of the Institute of Ecology and Biological Resources, VAST, Ha Noi, Vietnam.

Hydrodistilation of Essential Oil: 1.0–3.0 kg each sample of fresh leaf, twig, fruit material was shredded and hydrodistilled for 4 hours using a Clevenger type apparatus. The principle of hydrodistilation was based on the guideline of the Ministry of Health (2017). The essential oil was separated and stored at -5 ^{o}C until analysis.

GC/MS-FID analysis: Analysis of the essential oils was carried out by GC/MS-FID using an Agilent GC7890A system with Mass Selective Detector (Agilent 5975C). An HP-5MS fused silica capillary column (60 m × 0.25 mm i.d. × 0.25 μm film thickness) was used. Helium was the carrier gas with a flow rate of 1.0 mL/min. The inlet temperature was 250 ^{o}C and the oven temperature program was as follows: 60 ^{o}C to 240 ^{o}C at 4 ^{o}C/min. The split ratio was 1:100, the detector temperature was 270 ^{o}C, and the injection volume was 1 μL. The MS analysis was carried out at interface temperature 270 ^{o}C, MS mode, E.I. detector voltage 1,258 V, and mass range 35–450 Da at 1.0 scan/s. FID analysis was carried out using the same chromatographic conditions. The FID temperature was 270 ^{o}C. Essential oil constituents were identified by their relative retention indices, determined by co-injection of a homologous series of n-alkanes (C5–C30), as well as by comparison of their mass spectral fragmentation patterns with those stored on the MS library NIST08, Wiley09, HPCH1607 (Adams, 2017; Linstrom & Mallard, 2020). Data processing software was MassFinder 4.0 (König et al., 2020). Component relative concentrations were calculated based on the area peak of FID chromatography without standardization.

Microbial strains: The antimicrobial activity of the essential oils was evaluated using 1 strain each of Gram (+) bacteria *Staphylococcus aureus* (ATCC 13709), Gram (-) bacteria *Escherichia coli* (ATCC 25922) and yeast *Candida albicans* (ATCC 10231). The MIC and IC_{50} values of the oils then were determined using 3 above mentioned strains of microorganisms and 2 other strains of Gram (+) bacteria including *Bacillus subtilis* (ATCC 6633) and *Lactobacillus fermentum* (VTCC N4), 2 other strains of Gram (-) bacteria including *Salmonella enterica* (VTCC) and *Pseudomonas aeruginosa* (ATCC 15442). The ATCC strains were obtained from American Type Culture Collection; the VTCC strains were obtained from the Vietnam Type Culture Collection, Institute of Microbiology and Biotechnology, Vietnam National University, Ha Noi.

Screening of antimicrobial activity: The agar disk diffusion method was used to test the antimicrobial activity of essential oil (Balouiri et al, 2016). A 100 µL inoculum solution was taken and spread evenly over the surface of the agar. Two holes were made on agar plates. 50 µL essential oil was put into each hole. The petri dishes were kept at room temperature for 2–4 hours and then incubated at 37 ^{o}C for 18–24 hours. An inhibition zone of 14 mm or greater was considered as high antibacterial activity (Philip et al., 2009).

MIC and IC_{50} of the essential oils were measured by the microdilution broth susceptibility assay (Hadacek & Greger, 2000; Cos et al., 2006). After incubation at 37 ^{o}C for 24 hours, the MIC values were determined at well with the lowest concentration of agents that completely inhibit the growth of microorganisms. The IC_{50} values were determined by the percentage of microorganisms inhibited growth based on the turbidity measurement data of EPOCH2C spectrophotometer (BioTeK Instruments, Inc Highland Park Winooski, USA) and Rawdata computer software (Belgium) according to the following equations:

$$\%\ \text{inhibition} = \frac{OD_{\text{control}(+)} - OD_{\text{test agent}}}{OD_{\text{control}(+)} - OD_{\text{control}(-)}} \times 100\%$$

$$IC_{50} = High_{Conc} - \frac{\left(High_{Inh\%} - 50\%\right) \times \left(High_{Conc} - Low_{Conc}\right)}{\left(High_{Inh\%} - Low_{Inh\%}\right)}$$

Where: OD: Optical density; control(+): Only cells in medium without an antimicrobial agent; test agent: corresponds to a known concentration of antimicrobial agent; control(-): Culture medium without cells. $High_{Conc}$/Low_{Conc}: Concentration of test agent at high concentration/low concentration; $High_{Inh\%}$/$Low_{Inh\%}$: % inhibition at high concentration/% inhibition at low concentration.

Reference materials: Ampicillin for Gram (+) bacteria with IC_{50} and MIC values in the ranges of 0.02–3.62 µg/mL and 0.125–32.0 µg/mL, Cefotaxime for Gram (-) bacteria with IC_{50} and MIC values in the range of 0.07–4.34 µg/mL and 0.5–32.0 µg/mL, Nystatine for fungal strains with IC_{50} and MIC values of 1.32 µg/mL and 8.0 µg/mL.

Statistical Analysis: Average and standard deviation values of diameters of microorganism inhibition zone in the test were calculated using software Excel.

RESULTS AND DISCUSSION

Chemical composition of *Magnolia balansae* essential oils

By hydrodistillation, essential oils from leaves, twigs of *M. balansae* obtained were pale yellow liquids, while the oils from its fruits were dark yellow liquids. All of these oils had a lower density than water. The chemical compositions of the essential oils of *M. balansae* from Son La and Phu Tho provinces are summarized in Table 1.

Table 1. Compositions of the essential oils of *Magnolia balansae*

No.	RI	Components	Essential oil of *M. balansae* (%)			
			Leaf SL	Twig SL	Fruit SL	Fruit PT
1	800	Hexanal			2.7	
2	861	*n*-Hexanol			0.6	0.3
3	888	2-Heptanone			0.8	
4	901	Heptanal			0.3	
5	939	*α*-Pinene			0.2	0.2
6	984	*β*-Pinene		0.3	0.7	0.4
7	989	3-*p*-Menthene (= Menthomenthene)			0.2	
8	992	Myrcene				3.8
9	993	2-Pentylfuran			0.5	
10	1009	*p*-Mentha-1(7),8-diene			0.1	
11	1011	*α*-Phellandrene				0.3
12	1028	*p*-methyl-Anisole			1.2	
13	1030	*o*-Cymene				0.3
14	1033	Limonene			0.6	0.4
15	1037	1,8-Cineole		0.3	1.0	0.2
16	1058	2-Octenal			0.3	
17	1076	*trans*-Linalool oxide (furanoid)		0.3	1.2	
18	1092	*cis*-Linalool oxide (furanoid)		0.2	1.0	
19	1101	Linalool		5.3	13.8	3.0
20	1105	Nonanal		0.3		
21	1106	Hotrienol			0.5	
22	1115	*o*-Guiacol			0.3	
23	1118	*β*-Phenyl ethyl alcohol			0.2	
24	1148	*trans*-Sabinol			0.4	
25	1148	*cis*-Sabinol		0.5		
26	1152	*trans*-Verbenol		0.1		
27	1172	Pinocarvone		0.2		
28	1185	Terpinen-4-ol		0.2		
29	1197	*α*-Terpineol		0.2		0.2
30	1204	Myrtenol		0.4	0.3	
31	1206	Myrtenal		0.6	0.5	
32	1264	2-Phenylethyl acetate				0.4
33	1307	Sabinyl acetate		0.1		
34	1320	(*2E,4E*)-Decadienal		0.2	0.6	
35	1349	*δ*-Elemene				1.5
36	1382	*α*-Ylangene		0.2		0.2
37	1389	*α*-Copaene		0.5	0.3	0.2

No.	RI	Components	Essential oil of *M. balansae* (%)			
			Leaf SL	Twig SL	Fruit SL	Fruit PT
38	1403	*cis-β*-Elemene	2.1	4.8	3.4	10.4
39	1420	Ylanga-2,4(15)-diene		0.1		
40	1434	β-Cedrene			0.3	
41	1437	(*E*)-*β*-Caryophyllene	1.7	0.5	0.3	2.5
42	1441	*β*-Copaene			0.2	
43	1447	*γ*-Elemene				0.7
44	1465	(*E*)-*β*-Farnesene		0.2		
45	1471	*α*-Humulene	0.6	0.2		1.1
46	1479	9-*epi*-(*E*)-*β*-Caryophyllene	0.2			
47	1490	*γ*-Muurolene		0.4	0.1	0.4
48	1496	(*E*)-*β*-Ionone	0.5	0.2		
49	1497	*α*-Amorphene				0.6
50	1501	Germacrene D				8.6
51	1508	*trans-β*-Guaiene				0.7
52	1510	*trans*-Muurola-4(14),5-diene		0.4		
53	1513	Bicyclogermacrene	0.9		0.5	5.9
54	1513	*α*-Muurolene		0.5		
55	1525	*δ*-Amorphene				0.3
56	1531	*γ*-Cadinene	0.4	1.0	0.5	0.2
57	1536	*δ*-Cadinene	0.2	0.4	0.1	1.1
58	1538	*cis*-Calamenene	0.3	0.2		
59	1559	*α*-Calacorene	0.3	0.4	0.2	0.4
60	1563	Elemol	2.4	2.4	0.5	0.7
61	1570	(*E*)-Nerolidol	3.2	1.1	1.4	0.8
62	1582	Germacrene B				1.6
63	1587	Mintoxide	1.0	1.8	1.4	
64	1600	Germacrene D-4-ol				0.3
65	1599	Spathulenol	33.9	20.7	11.2	5.6
66	1606	Caryophyllene oxide	10.0	10.1	9.3	4.1
67	1613	Guaiol (=Champacol)	0.7			
68	1614	Cubeban-11-ol		0.5		
69	1619	Humulene epoxide I		0.3	0.2	
70	1620	Viridiflorol				1.1
71	1625	Ledol	0.9			
72	1626	*epi*-Cedrol	0.7	0.8		
73	1633	Cedrol				0.6
74	1631	Humulene Epoxide II	2.6	2.7	2.8	1.4
75	1646	1-*epi*-Cubenol		2.5	1.4	
76	1646	Alismol	2.3			
77	1650	Phenyl ethyl hexanoate		0.6	0.2	1.0
78	1650	*γ*-Eudesmol	0.3			
79	1658	2-Phenylethyl tiglate				1.0
80	1663	*α*-Muurolol (= δ-Cadinol)	0.7	2.5	0.6	0.8
81	1667	*epi*-*α*-Cadinol (= τ-Cadinol)				1.1

No.	RI	Components	Essential oil of *M. balansae* (%)			
			Leaf SL	Twig SL	Fruit SL	Fruit PT
82	1668	*epi*-*α*-Muurolol (= τ-Muurolol)				1.2
83	1668	Copaen-15-ol		0.5		
84	1670	*β*-Eudesmol	6.9	4.3	0.7	
85	1673	*α*-Cadinol			0.7	7.2
86	1676	*neo*-Intermedeol	2.7	2.7	1.8	1.6
87	1693	Cadalene		0.8		
88	1772	Cyclocolorenone		0.9		
89	1847	6,10,14-Trimethylpentadecan-2-one	0.6	0.3		
90	2117	Phytol	1.7	0.3		
Total			80.1	75.0	66.1	73.4
Monoterpene hydrocarbons			0.0	0.3	1.8	5.4
Oxygenated monoterpenes			1.7	8.4	18.7	3.4
Sesquiterpene hydrocarbons			6.1	9.2	5.7	36.0
Oxygenated sesquiterpenes			68.3	53.8	32.0	26.5
Oxygenated diterpenes			2.3	0.6	0.0	0.0
Benzenoids			0.6	2.0	2.1	2.1
Other compounds			1.0	0.7	5.8	0.3

Note: RI = Retention indices, SL = Collected in Son La province, PT = Collected in Phu Tho province.

Essential oils of *M. balansae* yielded 0.043, 0.023, 0.064 and 0.060% (v/w), calculated on a dry weight basis (the leaves, twigs, fruits collected in Son La and the fruits collected in Phu Tho). A total of 90 essential oil components were identified. Among them, 30, 49, 47 and 42 compounds were identified representing 80.0, 75.0, 66.1 and 73.4% of the respective oil compositions. Oxygenated sesquiterpenes were predominant (68.3, 53.8 and 32.0%) in the oils from Son La. While sesquiterpene hydrocarbons (36.0%) were predominant in the fruit oil from Phu Tho.

The common feature of 3 oil samples of *M. balansae* in Son La was that they contained 17 same compounds. In which, there were 2 same main components: spathulenol (33.9, 20.7 and 11.2%) and caryophyllene oxide (10.0, 10.1 and 9.3%). On the other hand, linalool (13.8%) was the third main constituent of the fruit oil. The most abundant minor components identified were: (*E*)-Nerolidol (3.2%) and *β*-eudesmol (6.9%) (Leaf oil), linalool (5.3%), *cis*-*β*-elemene (4.8%) and *β*-eudesmol (4.3%) (Twig oil), *cis*-*β*-elemene (3.4%) and humulene epoxide II (2.8%) (Fruit oil).

There was a big difference in the main compounds of fruit oil from *M. balansae* in Phu Tho to the one in Son La. In the former oil, *cis*-*β*-elemene (10.4%) was the sole main constituent. While, in the later oil, there were 3 main constituents mentioned above. Besides, germacrene D (8.6%), bicyclogermacrene (5.9%), spathulenol (5.6%), caryophyllene oxide (4.1%) and *α*-cadinol (7.2%) had significant contents in the fruit oil of *M. balansae* in Phu Tho (Table 1).

In comparison to the previous studies, only a few species of genus *Magnolia*, for example, *Magnolia gloriensis* (Haber et al., 2008), *Magnolia mediocris* (Do et al., 2016), *Magnolia macclurei* (Chu et al., 2020a), *Magnolia coriacea* (Chu et al., 2020b), the essential oil composition was rich in sesquiterpenoids similar to the current study. Many studied species of genus *Magnolia* had contents of monoterpenes that account for the majority of essential oils including *Magnolia sieboldii* (Sun et al., 2014), *Magnolia acuminata*, *Magnolia calophylla*, *Magnolia virginiana* (Farag et al., 2015), *Magnolia hypolampra* (Liu et al., 2007; Chu et al., 2019), *Magnolia kwangsiensis* (Huang et al., 2010; Zheng et al., 2015; Zheng et al., 2019; Chu et al., 2020c), and *Magnolia insignis* (Chu et al., 2021).

A previous study indicated that flower essential oil of *M. balansae* in China consists of 2 main constituents: Ethyl hexanoate (61.0%) and limonene (15.7%) (Zhu, 1993). The leaf oil of *M. balansae* collected in Pu Mat National Park, Nghe An province of Vietnam consisted of 54 compounds representing more than 95.0% of the oil, with 3 main constituents: *α*-pinene (18.4%), *α*-phellandrene (17.4%) and germacrene D (18.0%) (Nguyen et al., 2005), while these 3 compounds were absent in the present study. Some minor compounds were present in the leaf oil such as *β*-myrcene (3.9%), *β*-phellandrene (7.4%), *δ*-elemene (5.1%) and bicyclogermacrene (7.6%) (Nguyen et al., 2005). Whereas in the present study, *β*-myrcene, *β*-phellandrene and *δ*-elemene were absent, and bicyclogermacrene was present at a low concentration in leaf oil.

Antimicrobial activity of *Magnolia balansae* essential oils

The antimicrobial activity of the *M. balansae* oils was assessed using the standard agar disk diffusion method against three test microorganisms. Results obtained after 18–24 hours of incubation are presented in Table 2.

Table 2. Anti-yeast and antibacterial activity of essential oils of *Magnolia balansae* (average ± standard deviation, n = 2)

Samples	Inhibition zones (mm)		
	Staphylococcus aureus	*Escherichia coli*	*Candida albicans*
Leaf SL	31.8 ± 0.4	18.0 ± 0.7	> 40.0
Twig SL	25.5 ± 0.7	15.8 ± 0.4	38.8 ± 0.4
Fruit SL	30.5 ± 0.7	17.5 ± 0.7	> 40.0
Fruit PT	19.0 ± 1.4	13.5 ± 0.7	20.5 ± 0.7

Note: SL = Collected in Son La province, PT = Collected in Phu Tho province.

All oils of *M. balansae* (Son La) had a strong inhibitory activity (Philip et al., 2009) against 3 tested microorganism strains with inhibitory zone diameters from 15.8 to 40.0 mm. *M. balansae* fruit oil (Phu Tho) exhibited moderate inhibitory activity against *Escherichia coli*, and strong activity against *Staphylococcus aureus* and *Candida albicans* with inhibitory zone diameters from 13.5 mm to 20.5 mm. Of the 3 microorganism strains tested, *E. coli* was more tolerant to the *M. balansae* essential oils than the other two strains. The diameter range of the microbiological inhibition zones was 13.5–18.0 mm (for *E. coli*) compared to 19.0−31.8 mm (for *S. aureus*) and 20.5 - more than 40.0 mm (for *C. albicans*).

The essential oil samples were then determined MIC and IC_{50} values using 7 strains of microorganisms. The results obtained after 16–24 hours are presented in Table 3. In general, the leaf oil of *M. balansae* from Son La province had stronger antimicrobial activity than the other oils as indicated by the ranges of IC_{50} and MIC values from 57.0 µg/mL to 3,072 µg/mL and from 512 µg/mL to 8,192 µg/mL. While the IC_{50} and MIC values of the other three oil samples ranged from 60.0 µg/mL to 7,680 µg/mL and from 512 µg/mL to 16,384 µg/mL. Out of 7 strains of microorganisms tested, *Bacillus subtilis* was the most sensitive to the *M. balansae* oils (Table 3).

The antimicrobial activity of essential oils extracted from different species of genus *Magnolia* has been reported. *Magnolia liliflora* essential oil inhibited the growth of tested strains of fungi with MIC and MFC from 125 µg/mL to 500 µg/mL and from 125 µg/mL to 1,000 µg/mL, respectively (Bajpai & Kang, 2012). *Magnolia grandiflora* leaf oil had MIC values for *S. aureus* and *Streptococcus pyogenes* bacteria of 500 µg/mL and 125 µg/mL (Guerra-Boone et al., 2013). Another study reported that the antimicrobial activity of essential oils of the same plant may vary seasonally throughout the year, as was the case for *Magnolia ovata* (syn. *Talauma ovata*) (Stefanello et al., 2008).

Table 3. MIC and IC_{50} concentrations of essential oils of *M. balansae*

Essential oil samples		Leaf SL		Twig SL		Fruit SL		Fruit PT	
Value (µg/mL)		IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC
Gram (+) bacteria	*Staphylococcus aureus*	405	1,024	931	2,048	467	1,024	1,210	2,048
	Bacillus subtilis	57.0	512	63.0	512	60.0	512	82.0	512
	Lactobacillus fermentum	3,072	8,192	5,301	16,384	3,456	8,192	6,599	16,384
Gram (-) bacteria	*Salmonella enterica*	2,348	4,096	5,461	16,384	3,413	8,192	7,680	16,384
	Escherichia coli	2,355	8,192	2,498	8,192	2,448	8,192	2,765	8,192
	Pseudomonas aeruginosa	1,984	8,192	4,411	16,384	3,755	16,384	5,559	16,384
Yeast	*Candida albicans*	768	2,048	1,317	4,096	853	4,096	2,650	4,096

Note: SL = Collected in Son La province, PT = Collected in Phu Tho province.

The various antimicrobial activity of essential oils on microorganisms can be derived from their main compounds or the synergism of many of the components in the oils. Related to the antimicrobial activity role of four main compounds of *M. balansae* oils in the present study, a previous study indicated that spathulenol inhibited *in vitro* growth and had bactericidal activity against *Mycobacterium tuberculosis* (Dzul-Beh et al., 2019). In another study, *β*-elemene was reported to have anti-inflammatory and anti-cancer effects (Zhang et al., 2011). Linalool exhibited antibacterial against *S. aureus* and *E. coli* with MIC values of 5.0 µg/mL and 6.0 µg/mL, respectively (Soković et al., 2010) and methicillin-resistant *S. aureus* isolates at concentration ≤ 90 µg/mL (Taechowisan et al., 2018). Whereas caryophyllen oxide showed weak antibacterial activity against *S. aureus*, *E. coli* and *P. aeruginosa* with MIC values of 500 µg/mL (Kiran et al., 2010).

CONCLUSION

The contents of essential oils obtained from *M. balansae* leaves, twigs, fruits in Son La and fruits in Phu Tho were 0.043, 0.023, 0.064% and 0.060% (v/w), calculated on a dry weight basis. All oils of *M. balansae* (Son La) had the same main components: spathulenol (33.9, 20.7 and 11.2%) and caryophyllene oxide (10.0, 10.1 and 9.3%). On the other hand, linalool (13.8%) was the third main constituent of the fruit oil. The difference between fruit oil in Phu Tho and Son La province was *cis*-β-elemene (10.4%) as its sole main compound.

The essential oils of *M. balansae* exhibited moderate and strong inhibitory activity against 3 microorganisms using the standard agar disk diffusion method. The microdilution broth susceptibility assay for 7 strains of microorganisms tested showed that *B. subtilis* is the most sensitive bacteria to the oils. The antimicrobial activity test results of the present study can be the basis for future research in the field of the food and beverage industry as flavoring and preservative agents.

Acknowledgements: This research was funded by Vietnam National Foundation for Science and Technology Development (NAFOSTED) under Grant number 106.03-2019.16.

REFERENCES

Adams R. P., 2017. Identification of essential oil components by gas chromatography/mass spectrometry, ed. 4.1. Allured Publishing Corporation. ISBN: 978-1-932633-21-4, pp. 804.

Bajpai V. K., Kang S. C., 2012. *In vitro* and *in vivo* inhibition of plant pathogenic fungi by essential oil and extracts of *Magnolia liliflora* Desr. *Journal of Agricultural Science and Technology*, 14: 845–856.

Balouiri M., Sadiki M., Ibnsouda S. K., 2016. Methods for *in vitro* evaluating antimicrobial activity: A review. *Journal of Pharmaceutical Analysis*, 6: 71–79.

Chen K., Zhao G., Wu S., Zhuang Q., Qian F., 2013. Preliminary researches on influence of several kinds of Magnoliaceae plants on activity of cholinesterase from pig serum. *Advanced Materials Research*, 791–793: 163–166.

Chu T. T. H., Bui V. T., Dinh T. T. T., 2020b. Chemical composition and antimicrobial activity of the essential oil from leaves of *Magnolia coriacea* (Hung T. Chang & B. L. Chen) Figlar growing in Vietnam. *Academia Journal of Biology*, 42(3): 135–144.

Chu T. T. H., Dinh T. T. T., Vu Q. N., Nguyen K. B. T. and Setzer W. N., 2021. Composition and antimicrobial activity of essential oils from leaves and twigs of *Magnolia hookeri* var. *longirostrata* D.X.Li & R. Z. Zhou and *Magnolia insignis* Wall. in Ha Giang province of Vietnam. *Record of Natural Product*, 15(3): 207–212.

Chu T. T. H., Tran H. T., Le N. D., Dinh T. T. T., Nguyen D. K., Ha M. T., 2020a. Chemical composition and antimicrobial activity of the essential oil from twigs and leaves of *Magnolia macclurei* (Dandy) Figlar from Ha Giang province, Vietnam. *Academia Journal of Biology*, 42(1): 41–49.

Chu T. T. H., Tran H. T., Nguyen T. H., Dinh T. T. T., Bui V. T., Nguyen V. D., Vu Q. N., Setzer W. N., 2020c. Chemical composition and antimicrobial activity of the leaf essential oils of *Magnolia kwangsiensis* Figlar & Noot growing in Vietnam. *American Journal of Essential Oils and Natural Products*, 8(3): 13–19.

Chu T. T. H., Tran H. T., Nguyen T. H.n, Ha T. V. A., Le N. D., Dinh T. T. T., Do D. N., Setzer W. N., 2019. Chemical composition and antimicrobial activity of the leaf and twig essential oils of *Magnolia hypolampra* (Dandy) Figlar growing in Na Hang Nature Reserve, Tuyen Quang province of Vietnam. *Natural Product Communications*, 14(6): 1–7.

Do N. D., Tran D. T. & Isiaka A. O., 2016. Essential oil composition of four Magnoliaceae species cultivated in Vietnam. *Journal of Herbs, Spices & Medicinal Plants*, 22(3): 279–287.

Dzul-Beh A. de J., García-Sosa K., Uc-Cachón A. H., Bórquez J., Loyola L. A., Barrios-García H. B., Pe˜na-Rodríguez L. M., Molina-Salinas G. M., 2019. *In vitro* growth inhibition and bactericidal activity of spathulenolagainst drug-resistant clinical isolates of *Mycobacterium tuberculosis*. *Revista Brasileira de Farmacognosia* (*Brazilian Journal of Pharmacognosy*), 29: 798–800.

Farag M. A., El Din R. S., Fahmy S., 2015. Headspace analysis of volatile compounds coupled to chemometrics in leaves from the Magnoliaceae family. *Records of Natural Products*, 9(1): 153–158.

Guerra-Boone L., Álvarez-Román R., Salazar-Aranda R., Torres-Cirio A., Rivas-Galindo V. M., Waksman de Torres N., González G. M., Pérez-López L.A., 2013. Chemical compositions and antimicrobial and antioxidant activities of the essential oils from *Magnolia grandiflora*, *Chrysactinia mexicana*, and *Schinus molle* found in Northeast Mexico. *Natural Product Communications*, 8(1): 135–138.

Haber W. A., Agius B. R., Stokes S. L., Setzer W. N., 2008. Bioactivity and chemical composition of the leaf essential oil of *Talauma gloriensis* Pittier (Magnoliaceae) from Monteverde, Costa Rica. *Records of Natural Products*, 2(1): 1–5.

Huang P. X., Zhou Y. H., Lai J. Y., Li W. G., Liu X. M., 2010. Extraction and analysis of volatile constituents from testa of rare and endangered plant *Kmeria septentrionalis*. *Guihaia*, 30(5): 691–695.

Kiran I., Durceylan Z., Kirimer N., Başer K. H. C., Noma Y., & Demirci F., 2010. Biotransformation of α-cedrol and caryophyllene oxide by the fungus Neurospora crassa. *Natural product communications*, 5(4): 515–518.

König W. A., Joulain D., Hochmuth D. H., 2020. Terpenoids library - Terpenoids and related constituents of essential oils. https://massfinder.com/wiki/Terpenoids_Library. Retrieved on 01 August 2020.

Liu J-F, Huang M, Tan, L-Q, Liang J-M, Wu X-G., 2007. GC/MS analysis of chemical constituents of volatile oil of *Michelia hedyosperma* Lew fruits. *Chinese Journal of Pharmaceutical Analysis*, 27(9): 1481–1483.

Ministry of Health, 2017. Vietnamese Pharmacopoeia V. Medical Publishing House, Ha Noi, Vietnam (in Vietnamese).

MOST and VAST (Ministry of Science and Technology, Vietnam Academy of Science and Technology), 2007. Vietnam’s Red Data Book. Part II. Plants. Publishing House of Natural Sciences and Technology, Hanoi (in Vietnamese).

Nguyen A. D., Tran D. T. & Nguyen X. D., 2005. Chemical composition of the leaf oil of Michelia balansae (A.DC.) Dandy from Vietnam. *Journal of Essential Oil Bearing Plants*, 8(1): 11–14.

Nguyen Q. H., Nguyen T. H., Tu B. N., Nguyen S. K., 2015. Preliminary studies on the diversity of Magnolias of Ha Giang province and their conservation status. Proceedings of the 6th National Conference on ecology and biological resources: 130−136 (*in Vietnamese*).

Nguyen T. B., 2003. Checklist of plant species of Vietnam. Vol. II. Agriculture Publishing House, Hanoi (in Vietnamese).

Nguyen T. H., Nguyen S. K., Nguyen T. V., Nguyen T. S. and Nguyen Q. H., 2012. Seed collection and test propagation of Conifer and Magnolia from the Sinh Long Forest Area. Report supported to People Resources and Conservation Foundation, 12 pp.

Nguyen T. P., 2016. Study on conservation of *Michelia balansae* (DC.) Dandy 1927 in Tam Dao National Park, Vinh Phuc province. Thesis submitted for the Degree of Bachelor of Science. Vietnam National University of Forestry (in Vietnamese).

Pham H. H., 1999. An Illustrated Flora of Vietnam. Vol.I. Tre Publishing House. p236. (in Vietnamese).

Philip K., Malek S. N. A., Sani W., Shin S. K., Kumar S., Lai H. S., Serm L. G., and Rahman S. N. S. A., 2009. Antimicrobial activity of some medicinal plants from Malaysia. *American Journal of Applied Sciences*, 6(8): 1613–1617.

Sima Y. K., Wu T., Chen S. Y., Ma H. F., Hao J. B., Fu Y. P., & Zhu Y. F., 2021. The complete chloroplast genome sequence of *Michelia balansae* var. *balansae* (Aug. Candolle) Dandy, a timber and spices species in Magnoliaceae. *Mitochondrial DNA Part B*, 6(2): 465–467.

Soković M., Glamočlija J., Marin P. D., Brkić D., & Van Griensven L. J., 2010. Antibacterial effects of the essential oils of commonly consumed medicinal herbs using an in vitro model. *Molecules*, 15(11): 75327546.

Stefanello M. É. A., Salvador M. J., Ito I. Y., Wisniewski Jr. A., Simionatto E. L., Mello-Silva de R., 2008. Chemical composition, seasonal variation and evaluation of antimicrobial activity of essential oils of *Talauma ovata* A. St. Hil. (Magnoliaceae). *Journal of Essential Oil Research*, 20(6): 565–569.

Sun G., Du F., Wan R., 2014. Comparison of biomaterials from essential oils in five parts of *Magnolia sieboldii*. *Applied Mechanics and Materials*, 442: 142–146.

Taechowisan T., Jantiya J., Mungchukeatsakul N., & Phutdhawong W. S., 2018. Major compounds from *Ocimum basilicum* L. and their antimicrobial activity against methicillin-resistant Staphylococcus aureus. *Biomedical Journal of Scientific & Technical Research*, 3(3): 3315–3323.

Tu B. N., Nguyen T. H., Nguyen T. T., 2014. Some species of genera *Michelia* L. at Bat Dai Son Natural Reserve, Quan Ba District, Ha Giang province. *VNU Journal of Science: Natural Sciences and Technology*, 30(2): 61–70 (in Vietnamese).

Vu Q. N., 2011. Taxonomic revision of the family Magnoliaceae from Vietnam. A thesis submitted for the Degree of Doctorate in Botany at the Graduate University of The Chinese Academy of Sciences, Quangzhou.

Xu F. X., and Kirchoff B. K., 2008. Pollen morphology and ultrastructure of selected species of Magnoliaceae. *Review of Palaeobotany and Palynology*, 150: 140–153.

Zhang R., Tian A., Zhang H., Zhou Z., Yu H., Chen L., 2011. Amelioration of experimental autoimmune encephalomyelitis by β-elemene treatment is associated with Th17 and Treg cell balance. *Journal of Molecular Neuroscience*, 44: 31–40.

Zheng Y. F., Liu X. M., Zhang Q., Lai F, Ma L., 2019. Constituents of the essential oil and fatty acid from rare and endangered plant *Magnolia kwangsiensis* Figlar & Noot. *Journal of Essential Oil Bearing Plants*, 22(1): 141–150.

Zheng Y. F., Ren F., Liu X. M., Lai F., Ma L., 2015. Comparative analysis of essential oil composition from flower and leaf of *Magnolia kwangsiensis* Figlar & Noot. *Natural Product Research*, 30(13): 1552–6.

Zhu L., 1993. Aromatic Plants and Essential Constituents. South China Institute of Botany, Chinese Academy of Sciences: Hai Feng Publishing Co., Hong Kong.