DOI 10.1007/s10600-023-04167-5
Chemistry of Natural Compounds, Vol. 59, No. 5, September, 2023

ANTIMICROBIAL ACTIVITY AND CONSTITUENTS OF *Lindera annamensis* ESSENTIAL OIL FROM VIETNAM

Le Thi Huong,[1*] Le Duy Linh,[1] Nguyen Anh Dung,[1] Dao Thi Minh Chau,[1] Do Ngoc Dai,[2] and Isiaka Ajani Ogunwande[3*]

Lindera is a genus of about 80–100 [1] species of flowering plants in the family Lauraceae, mostly native to eastern Asia. In Vietnam, *Lindera* is represented by over 20 species spread throughout the country. Almost all of the species are aromatic with fragrance principles [1]. In our previous communications, the chemical compositions and antimicrobial activity of essential oils from *L. glauca* [2] and *L. rufa* [3] were reported. *Lindera annamensis* H. Liu is a tree that grows up to 5 m tall. It is known in Vietnamese as O duroc trung bo and has lenticellate twigs that are reddish brown when dry. It is similar to *L. spirei* in leaf shape and size but differs in having longer peduncles and longer staminate flowers [4]. There is no record of phytochemical studies on both the volatile and nonvolatile constituents of this species.

This study was performed with the leaves of *L. annamensis* harvested by handpicking and collected from Pu Huong Natural Reserve (Binh Chuan Commune), Con Cuong District, located in Vietnam (GPS: 19°35′19″N, 104°43′7″E). The plants were identified by Prof. Dr. L. T. Huong, and voucher specimen, LTH 896, which was deposited in the plant specimen room of Vinh University. The amount of sample collected was over 2.0 kg. The method of hydrodistillation follows the procedure normally employed in our laboratory [5–8]. The distillation time was 4 h and was conducted at normal pressure. The volatile oils were collected separately into clean weighed sample bottles. The oils were kept under refrigeration (4°C) until the moment of analysis. The instrumental analysis of the essential oil was performed by using gas chromatography (GC) on an Agilent Technologies HP 7890 Plus Gas chromatograph equipped with an FID and fitted with HP-5MS column (30 m × 0.25 mm, film thickness 0.25 μm, Agilent Technology) [2, 5–11]. The MS conditions were as follows: ionization voltage 70 eV; emission current 40 mA; acquisitions scan mass range of 35–350 amu at a sampling rate of 1.0 scan per second. The identification of constituents from the GC/MS spectra of *L. annamensis* was performed based on retention indices (RI) determined with reference to a homologous series of *n*-alkanes (C_4–C_{40}) under identical experimental conditions. The mass spectral (MS) fragmentation patterns were checked with those of other essential oils of known composition [12].

The minimum inhibitory concentration (MIC) and median inhibitory concentration (IC_{50}) values were measured by the microdilution broth susceptibility assay [2, 5–11]. Streptomycin was used as the antibacterial standard while nystatin and cycloheximide were used as antifungal standards. All experiments were performed in triplicate.

The average yield of essential oil was 0.17%. Table 1 presents the compounds as identified by GC/MS. The leaves of *L. annamensis* afforded essential oil dominated by sesquiterpene hydrocarbons (76.1%) and monoterpene hydrocarbons (14.9%). The other classes of compounds include oxygenated monoterpenes (2.3%) and oxygenated sesquiterpenes (2.4%). Non-terpene constituents make up to 2.6% of the oil contents. From the GC/MS analysis, α-zingiberene (66.3%) was the most singly abundant compound of *L. annamensis* leaf oil, with a significant quantity of (*E*)-β-ocimene (8.4%) and (*E*,*E*)-α-farnesene (5.8%). Other compounds that were identified at amounts up to 1% includes decanal (2.1%), *trans*-β-elemenone (1.9%), α-pinene (1.3%), camphene (1.3%), selina-4(15),7(11)-diene (1.3%), and bornyl acetate (1.0%). This is the first report on the chemical constituents of the studied essential oils of *L. annamensis*.

1) School of Natural Science Education, Vinh University, 182 Le Duan, Vinh City 4300, Nghe An Province, Vietnam, e-mail: lehuong223@gmail.com; 2) Faculty of Agriculture, Forestry and Fishery, NgheAn College of Economics, 51-Ly Tu Trong, Vinh City, NgheAn Province, Vietnam; 3) Institute of Research and Translation, Ibadan, Nigeria, e-mail: isiakaogunwande@gmail.com. Published in *Khimiya Prirodnykh Soedinenii*, No. 5, September–October, 2023, pp. 815–816. Original article submitted October 5, 2022.

 0009-3130/23/5905-0968

TABLE 1. Chemical Constituents of Essential Oil from the Leaves of *L. annamensis*

Compound	RI[a]	Concentration[b]	Compound	RI[a]	Concentration[b]
α-Pinene	939	1.3	(*Z*)-β-Farnesene	1460	0.6
Camphene	956	1.3	α-Humulene	1470	0.3
Sabinene	978	0.5	α-Zingiberene	1500	66.3
Myrcene	992	0.4	(*E*,*E*)-α-Farnesene	1512	5.8
α-Phellandrene	1011	0.9	δ-Cadinene	1537	0.2
Limonene	1034	0.7	β-Sesquiphellandrene	1540	0.4
(*Z*)-β-Ocimene	1037	0.8	Selina-4(15),7(11)-diene	1554	1.3
(*E*)-β-Ocimene	1049	8.4	Selina-3,7(11)-diene	1560	0.8
Terpinolene	1093	0.6	(*E*)- Nerolidol	1569	0.5
Nonanal	1102	0.2	*trans*-β-Elemenone	1601	1.9
p-Cymen-8-ol	1190	0.2	Monoterpene hydrocarbons		14.9
Estragole	1204	0.9	Oxygenated monoterpene		2.3
Decanal	1207	2.1	Sesquiterpene hydrocarbons		76.1
Bornyl acetate	1294	1.0	Oxygenated sesquiterpenes		2.4
Dodecanal	1410	0.3	Non-terpenes		2.6
β-Caryophyllene	1437	0.4	Total		98.3
Geranyl acetone	1456	0.2			

[a] Retention indices on HP-5MS column; [b] mean of three replicates.

TABLE 2. Antimicrobial Activity of the Leaf Oil of *L. annamensis*

Microorganism	MIC, µg/mL	IC_{50}, µg/mL	Microorganism	MIC, µg/mL	IC_{50}, µg/mL
Enterococcus faecalis ATCC299212	32.0 ± 0.00	9.34 ± 0.11	*Bacillus cereus* ATCC14579	64.0 ± 0.00	20.51 ± 0.12
Staphylococcus aureus ATCC25923	32.0 ± 0.00	10.34 ± 0.00	*Candida albican*s ATCC10231	32.0 ± 1.00	11.34 ± 0.10

The essential oil from the leaf oil of *L. annamensis* displayed promising antibacterial activity against *Enterococcus faecalis* ATCC299212 and *Staphylococcus aureus* ATCC25923, and anti-candidal action towards *Candida albicans* ATCC10231, with a minimum inhibitory concentration (MIC) value of 32.0 µg/mL (Table 2). The corresponding IC_{50} values (percentage of microorganisms that inhibited growth based on the turbidity measurement data) were 9.34, 10.34, and 11.34 µg/mL, respectively. The essential oil also showed antibacterial properties against *Bacillus cereus* ATCC14579 with an MIC value of 64.0 µg/mL and IC_{50} value of 20.51 µg/mL. Overall, the studied essential oils displayed moderate antimicrobial activity with most MIC values being less than 100 µg/mL, when considering the criteria for determining the antimicrobial efficacy of natural substances. Streptomycin, the standard antimicrobial agent for gram-positive bacteria displayed antimicrobial activity with MIC values in the range of 0.35 µg/mL to 1.29 µg/mL. In addition, nystatine used as a standard antimicrobial agent for gram-negative bacteria had an MIC value of 5.0 µg/mL, with cycloheximide, an anticandidal agent, showing activity at an MIC of 2.20 µg/mL. This is the first report on the antimicrobial activity of the essential oil of *L. annamensis*.

The antimicrobial activities of the essential oil of *L. annamensis* can be related to some compounds or synergy among the constituents present in the essential oils. The antimicrobial potential of some of these compounds has been reported. α-Pinene is known for its antibacterial activity [13], while zingiberene was noted for its antifungal and antibacterial activity against Gram-positive organisms [14].

REFERENCES

1. H. T. T. Do, J. C. Grant, B. N. Trinh, H. C. Zimmera, and J. D. Nichols, *J. Asia-Pacific Biodiv.*, **10**, 472 (2017).
2. D. T. M. Chau, N. T. G. An, L. T. Huong, and I. A. Ogunwande, *J. Essent Oil Bearing Plants*, **25**, 93 (2022).

3. D. N. Dai, T. D. Than, and J. A. Pino, *J. Essent. Oil Bearing Plants*, **16**, 832 (2013).
4. S. Tagane, S. Phetlasy, and T. Yahara, *Acta Phytotax. Geobot*., **72**, 253 (2021).
5. D. N. Dai, L. T. Huong, L. Y. Sam, and I. A. Ogunwande, *Chem. Nat. Compd.,* **58**, 148 (2022).
6. L. T. Huong, L. D. Linh, D. N. Dai, and I. A. Ogunwande, *Chem. Nat. Compd.*, **58**, 1152 (2022).
7. L. T. Huong, T. N. Hoi, N. T. Chung, T. D. Binh, D. H. Son, and I. A. Ogunwande, *Chem. Nat. Compd.*, **58**, 943 (2022).
8. L. T. Huong, D. T. M. Chau, D. N. Dai, and I. A. Ogunwande, *Rec. Nat. Prod.*, **16**, 477 (2022).
9. N. V. Le, L. N. Sam, L. T. Huong, and I. A. Ogunwande, *J. Essent., Oil Bearing Plants*, **25**, 82 (2022).
10. L. T. Huong, D. T. M. Chau, N. T. H. An, D. N. Dai, and I. A. Ogunwande, *J. Essent. Oil Bearing Plants*, **25**, 297 (2022).
11. L. T. Huong, N. T. Chung, D. T. M. Chau, D. N. Dai, and I. A. Ogunwande, *Rec. Nat. Prod.*, **16**, 387 (2022).
12. National Institute of Science and Technology, Chemistry Web Book Data. Data from NIST Standard Reference Database 69 (2011).
13. Y. Asakawa, *Dietary Monoterpenoids*, in: *Handbook of Dietray Phytochemicals*, Vol. 2, J. Xiao, S. D. Sarker, Y. Asakawa (eds.), Springer, Singapore, 2021, p. 607.
14. A. Abdullah, A. Khairulmazmi, S. Yasmeen, I. S. Ismail, A. Norhayu, M. R. Sulaiman, O. H. Ahmed, and M. R. Ismail, *Arabian J. Chem.*, **13**, 8012 (2020).