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Fonenol, the main constituent of the essential oil of the leaf of *Piper longum* L

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Abstract
In this paper, the compounds identified from the essential oil of the leaf of *Piper longum* L. are described. The essential oil which was hydrodistilled from the air-dried leaf of *P. longum* was characterized for the constituents by means of gas chromatography (GC) and gas chromatography coupled to mass spectrometry GC-MS). The yield of the colorless essential oil was 0.12% (v/w) calculated on a dry weight basis. The main constituent of the oil was fonenol (40.5%). The chemical compositions of the studied oil sample were considerably different from data observed from other samples around the world.

Keywords: *Piper longum*, essential oil, sesquiterpene, fonenol

Introduction
In our previous communications [1, 2], we reported the chemical compounds identified in the essential oils from the leaves of some *Piper* plants grown in Vietnam. It is well known that researches into the volatile components of several *Piper* plants were focused mainly on the fruits. *Piper longum* is a perennial, dioecious herb with woody rootstock and slender prostrate or ascending shoots. The leaves are thinly membranous and longest in lower leaves. The plant has erect branches about 30-60 cm tall, bearing erect inflorescences. The flowers are crowded, each with 3-4 stigmas. The berries are concrescent, forming a thin, fleshy, slender, cylindrical infructescence. Extracts from various parts of *P. longum* were reported to possesses anti-inflammatory [3], anti-fertility [4], contraceptive spermicidal [5], antidiabetic and antihyperlipidemic [6], antioxidant [7], antifungal [8], anti-rheumatoid [9], antiasthmatic [10] and analgesic [11] activities among several others [12]. Some of the phytochemical compounds isolated from *P. longum* were piperlongimin A and piperlongimin B which inhibited cell proliferation of human leukemia, HL-60 cell lines, and displayed major apoptosis-inducing effects [13], bakuchiol, bavachin, and isobavachalcone which have suppressive effects against pigmentation by melanin in the skin [14], piperine and piperidine with neuroprotective effects [15] as well as pipernonaline, a mosquito larvicidal agent [16].
A number of reports on the chemical constituents of essential oils from the fruits of *P. longum* have been published. The content of β-caryophyllene in *P. longum* fruit was found to be 10.2% [17]. The inflorescences oil was rich in eugenol (33.11%) and caryophyllene (9.29%) [18], while another authors reported the main components in *P. longum* to be β-caryophyllene (33.44%), 3-carene (7.58%) and eugenol (7.39%) [19]. A large quantity of pentadecane (17.8%), β-caryophyllene (17.0%) and β-bisabolene (11.2%) were also described from the oil [20]. Also, α-pinene (11.8%-15.3%), β-pinene (26.4%-43.1%), limonene (6.3%-10.6%) and β-caryophylene (5.6%-9.3%) were the main compounds in the root, stem and fruit oils of *P. longum* while camphene (13.9%) and bornyl acetate (10.4%) were additional compounds in the root [21]. However, the authors are aware of only two reports on the leaf oil of *P. longum* in which (*E*)-nerolidol (19.08%) and β-caryophyllene (12.25%) [18] as well as (*E*)-nerolidol (22.5%) and β-caryophyllene (16.88%) were the main compounds [21]. The essential oils of various parts of *P. longum* were known to have exhibited biological activities such as anti depressant [20] and inhibitory action on muscular activity of liver fluke, *Fasciola gigantica* [22].
The aim of the present paper was to report the chemical compounds identified in the essential oil from the leaf of *P. longum* grown in Vietnam.

2. Materials and methods

2.1 Collection of plant

The mature leaves of *P. longum* were collected from Pù Huống Nature Reserve, Nghệ An Province, Vietnam, in May 2013. Botanical identification was carried out by Dr. Hieu. A voucher specimen LDH 342 was deposited at the Botany Museum, Vinh University, Vietnam.

2.2 Preparation of plant sample

Prior to hydrodisitillation process the plant sample was air-dried under laboratory shade 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.

2.3 Hydodistillation of the essential oil

In this process 500 g of air-dried and pulverized leaves of *P. longum* were introduced into a 5 L flask and distilled water (5 L) was added until it covers the sample completely. Hydrodistillation was carried out with a Clevenger-type distillation unit designed according to the specification [23]. The distillation time was 3 h and conducted at normal pressure. The volatile oil distilled over water and was collected into clean weighed sample bottle. The oil was kept under refrigeration (4 ºC) until the moment of analyses.

2.4 Gas chromatography analysis (GC) of the oil

Gas chromatography (GC) analysis was performed on an Agilent Technologies HP 6890 Plus Gas chromatograph equipped with a FID and fitted with HP-5MS column (30 m X 0.25 mm, film thickness 0.25 μm, Agilent Technology). The analytical conditions were: carrier gas H_2 (1 mL/min), injector temperature (PTV) 250ºC, detector temperature 260 ºC, column temperature programmed from 40 ºC (2 min hold) to 220 ºC (10 min hold) at 4 ºC/min. Samples were injected by splitting and the split ratio was 10:1. The volume injected was 1.0 μL. Inlet pressure was 6.1 kPa. Each analysis was performed in triplicate. The relative amounts of individual components were calculated based on the GC peak area (FID response) without using correction factors as previously described [1, 2].

2.4 Gas chromatography-mass spectrometry (GC-MS) analysis of the oil

An Agilent Technologies HP 6890N Plus Chromatograph fitted with a fused silica capillary HP-5 MS column (30 m X 0.25 mm, film thickness 0.25 μm) and interfaced with Mass spectrometer HP 5973 MSD, was used for this experiment, under the same conditions as those used for gas chromatography analysis as described previously [1,2]. The conditions were the same as described above with He (1 mL/min) as carrier gas. The MS conditions were as follows: ionization voltage 70eV; emission current 40 mA; acquisitions scan mass range of 35-350 amu at a sampling rate of 1.0 scan/s.

2.4.1 Identification of the constituents of the oil

The identification of constituents was performed on the basis of retention indices (RI) determined with reference to a homologous series of *n*-alkanes (C_4-C_{40}), under identical experimental conditions. In some cases, co-injection with known compounds or standards (Sigma-Aldrich, St. Louis, MO, USA) under the same GC conditions was employed. The mass spectral (MS) fragmentation patterns were checked with those of other essential oils of known composition and with those in the literature [24].

3. Results & Discussion

The yield of essential oils was 0.12% (v/w) calculated on a dry weight basis. The hydrodistillation process afforded light yellow essential oil. The volatile compounds were displayed in Table 1, along with their percentages and retention indices calculated on HP-5MS column. Forty-three compounds representing 98.5% of the total volatile compounds were identified in the leaf essential oil. This consisted of 9 monoterpene hydrocarbons (5.0%), 6 oxygenated monoterpenes (6.0%), 2 aromatic ester (2.0%), 18 sesquiterpene hydrocarbons (32.8%) and 9 oxygenated sesquiterpenes (56.9%). The major compound identified in the leaf oil was fonenol (40.5%). There are significant quantities of elemol (8.2%), calamenene (4.1%), α-cadinol (3.9%), aromadendrene (3.6%), bicycloelemene (3.4%), γ-elemene (3.2%), germacrene D (3.0%) and bicyclogermacrene (3.0%).

A comparison of the present data with previous studies on the leaf oils of *P. longum* [18, 21] revealed some quantitative and qualitative variations. Firstly, nerolidol which was described as a major constituent of the oil of *P. longum* in the previous studies was not detected in the present oil sample. In addition, the amount of β-caryophyllene (1.6%) in the present data was lower when compared with previous samples (12.28% and 16.88%). Moreover, fonenol, the main compound in the present study was not mentioned previously to be present in *P. longum* oil samples. More importantly, several other compounds, such as eugenol, β-bisabolene, pentadecane that are characteristics of essential oils from other parts of the plant were not identified in the present study. These observations may be attributed to factors such as the different environmental and ecological between the points of collection and analysis of the various samples.

The present data is deemed to be normal because literature survey showed contradicting reports on the composition of volatile oils from several species of *Piper* plants. For example, α-humulene (16.4%) and β-caryophyllene (12.2%) were present in the leaf oil of *P. retrofractum* from Bangladesh [25], while the sample from Vietnam contained benzyl benzoate (14.4%), myrcene (14.4%) and bicycloelemene (9.9%). Previously, benzyl benzoate (49.1%) and benzyl acohol (17.9%) were described in the leaf oil of *P. sarmentosum* from Vietnam [2], the sample from Iran [26] consist of spathulenol (21.0%), myristicin (18.8%), β-caryophyllene (18.2%) and (*E,E*)-farnesol (10.5%). The leaf oil of *P. maclurei* [2] was characterized by large quantity of (*E*)-cinnami acid (37.4%) and (*E*)-nerolidol (19.4%) while the stem was rich in (*Z*)-9- octadecenoic acid methyl ester (28.0%), (*E*)-cinnamyl acetate (17.2%) and phytol (12.2%).

However, situation where the different parts of the plant contained the similar compositions in varying quantity have been observed. The main compounds of *P. harmandii* were sabinene (leaves, 14.5%; stems, 16.2%), benzyl benzoate (leaves, 20.0%; stems, 29.40%) and benzyl salicylate (leaves, 14.1%; stems, 24.3%) while sabinene (leaves, 17.9%; stems, 13.5%), benzyl benzoate (leaves, 20.5%; stems, 32.5%) and β-eudesmol (leaves, 13.8%; stems, 8.4%) were the main constituents of *P. brevicaule* [1]. It could be seen that each *Piper* oil sample has compositional pattern which are quite different from other species.

Table 1: Chemical constituents of essential oil of *P. longum*

Compounds [a]	RI (Cal.)	RI (Lit.)	Percentage [b]
α-Pinene	939	932	1.5
Camphene	953	946	0.2
β-Pinene	980	976	0.2
β-Myrcene	990	988	0.1
Limonene	1032	1024	2.2
(*Z*)-β-Ocimene	1043	1032	0.1
(*E*)-β-Ocimene	1052	1044	0.1
α-Terpinolene	1090	1089	0.5
Linalool	1100	1095	0.8
allo-Ocimene	1128	1128	0.1
(*E*)-Cinnamaldehyde	1266	1266	0.3
Bornyl acetate	1289	1287	0.2
Bicycloelemene	1327	1338	3.4
Cyclosativene	1371	1363	0.2
α-Copaene	1377	1374	0.2
β-Maaliene	1380	1382	0.6
Geranyl acetate	1381	1386	0.1
β-Panasinsene	1385	1388	0.5
β-Elemene	1391	1389	2.8
β-Caryophyllene	1419	1417	1.6
(*E*)-Cinnamyl acetate	1430	1422	0.5
γ-Elemene	1437	1435	3.2
α-Guaiene	1440	1433	0.1
Aromadendrene	1441	1439	3.6
Germacrene D	1485	1484	3.0
Ledene	1485	1489	0.2
δ-Selinene	1493	1493	0.6
α-Selinene	1493	1496	1.2
Bicyclogermacrene	1500	1500	3.0
δ-Cadinene	1525	1522	3.9
Elemol	1550	1548	8.2
Guaia-3,9-diene	1556	1556	0.5
p-Methoxy cinnamaldehyde	1564	1568	0.5
Spathulenol	1578	1577	1.0
Globulol	1585	1583	0.8
Guaiol	1601	1602	0.2
Rosifoliol	1615	1615	0.1
Fonenol	1621	1627	40.5
τ-Muurolol	1646	1640	2.1
α-Cadinol	1654	1652	3.9
Calamenene	1702	1702	4.1
Benzyl benzoate	1760	1760	0.2
Benzyl salicylate	1866	1876	1.8
Total			**98.9**
Monoterpene hydrocarbons			5.0
Oxygenated monoterpenes			2.3
Sesquiterpene hydrocarbons			32.8
Oxygenated sesquiterpenes			56.8
Aromatic esters			2.0

[a] Elution order on HP-5MS column; [b] Standard deviation (SD ±) were insignificant
and were excluded from the Table; RI (Cal.) Retention indices on HP-5MS column;
RI (Lit.) Literature retention indices

4. Conclusions

In conclusion, this paper reports the essential oil composition of leaf of *P. longum*. Though a quantitative difference between volatile oil from various parts of the plant was apparent, the root, stem, and fruit oils showed more similarity in their chemical composition. Fonenol the main constituent of the leaf oil was an uncommon compound identified in *P. longum*.

References

1. Hieu LD, Hoi TM, Thang TD, Ogunwande IA. Volatile constituents of three *Piper* species from Vietnam.' Natural Product Communications 2015; 10(11): 1997-1998.
2. Hieu LD, Hoi TM, Thang TD, Ogunwande IA. Chemical composition of essential oils from four Vietnamese species of *Piper* (Piperaceae). Journal of Oleo Science 2014; 63(3): 211-217.
3. Kumar A, Panghal S, Mallapur SS, Kumar M, Veerma R, Singh BK. Antiinflammatory Activity of *Piper longum* fruit oil Indian Journal of Pharmaceutical Science 2009; 71(4): 454-456.
4. Lakshmi V, Kumar R, Agarwal SK, Dhar JD. Antifertility activity of *Piper longum* Linn. in female rats. Natural Product Research. 2006; 20(3):235-239.
5. Abu Hasnath Md, Golam S, Ranjeet KN, Mohammed A, Beena K, Thakur SC. Spermicidal activity of the hexane extract of *Piper longum*: an *in vitro* study. Natural Product Research. 2015; 29(12):1166-1169.
6. Shaikh NB, Ramesh BK, Swapna S, Thandaiah KT, Malaka V, Kumar J, Rao CA. Antidiabetic and antihyperlipidemic activity of *Piper longum* root aqueous extract in STZ induced diabetic rats. BMC Complementary and Alternative Medicine. 2013; (13**)**:13-37.
7. Rameshkumar KB, Aravind APA, Mathew PJ. Comparative phytochemical evaluation and antioxidant assay of *Piper longum* L. and *Piper chaba* Hunter Used in Indian traditional systems of medicine. Journal of Herbs, Spices and Medicinal Plants. 2011; 17(4):351-360.
8. Ounchokdee U, Rueangrit S, Dethoup T. Antifungal activity profile of *Piper longum* fruit extract against plant pathogenic fungi. Journal of Biopest. 2016; 9(2):97-103.
9. Yende SR, Sannapuri VD, Vyawahare NS, Harle UN. Anti-rheumatoid activity of aqueous extract of *Piper longum* on Freunds adjuvant-induced arthritis in rats. International Journal of Pharmaceutical Science and Research. 2010; 1(9):129-133.
10. Kaushik D, Rani R, Kaushik P, Sacher D, Yadav J. *In vivo* and *in vitro* anti-asthmatic studies of *plant Piper longum* L. International Journal of Pharmacology. 2012; 8(3):192-197.
11. Ashajyothi V, Jabeen S. Phytochemical evaluation and pharmacological screening for analgesic activity of ethanolic extract of *Piper longum*. Research Journal of Pharmacy. 2014; 2(1):1-6.
12. Suresh K, Jitpal K, Suman SS. Overview for various aspects of the health benefits of *Piper Longum* Linn. fruit. Journal of Acupuncture Meridian Studies. 2011; 4(2):134-140.
13. Priyanka M, Sadhna S, Santosh KG, Shashi B, Vishwakarma RA, Sabari G. Two new amides with cytotoxic activity from the fruits of *Piper longum*. Journal of Asian Natural Products Research. 2011; 13(2):143-148.
14. Ohno O, Watabe T, Nakamura K, Kawagoshi M, Uotsu N, Chiba T *et al*. Inhibitory effects of bakuchiol, bavachin, and isobavachalcone isolated from *Piper longum* on melanin production in B16 mouse melanoma cells. Bioscience, Biotechnology and Biochemistry. 2010; 74(7):1504-1506.
15. Bi Y, Qu PC, Wang QS, Zheng L, Liu HL, Luo R *et al*. Neuroprotective effects of alkaloids from *Piper longum*

in a MPTP-induced mouse model of Parkinson's disease. Pharmaceutical Biology. 2015; 53(10):1516-1524.
16. Lee SE. Mosquito larvicidal activity of Pipernonaline, a piperidine alkaloid derived from long pepper, *Piper longum*. Journal of the American Mosquito Control Association. 2000; 16(3):245-247.
17. Tewtrakul S, Hase K, Kadota S, Namba T, Komatsu K, Tanaka K. Fruit oil composition of *Piper chaba* Hunt., *P. longum* L. and *P. nigrum* L. Journal of Essential Oil Research. 2000; 12(5):603-608.
18. Md Nazrul IB, Jaripa B, Anwar MN. Volatile constituents of essential oils isolated from leaf and inflorescences of *Piper longum* Linn. The Chittagong University Journal of Biological Science 2008; 3(1, 2):77-85.
19. Ling L, Song GX, Hu YM. GC-MS analysis of the essential oils of *Piper nigrum* L. and *Piper longum* L. Chromatographia. 2007; 66(9, 10):785-790.
20. Khan M. Comparative physicochemical evaluation of fruits and anti depressant potential of volatile oils of fruits of local *Piper* species. Oriental Journal of Chemistry. 2015; 31(1):21-29.
21. Varughese T, Kumar PU, Deepak M, Balachandran I, Rema AB. Chemical composition of the essential oils from stem, root, fruit and leaf of *Piper longum* Linn. Journal of Essential Oil Bearing Plants. 2016; 19(1):52-58.
22. Thakur US, Kumar D, Kumar S, Santosh T, Kumar M. Inhibitory effect of essential oils of *Allium sativum* and *Piper longum* on spontaneous muscular activity of liver fluke, *Fasciola gigantic*. Experimental Parasitology. 2009; 123(4): 302-308.
23. Vietnamese Pharmacopoeia. Medical Publishing House, Hanoi, Vietnam, 1997.
24. National Institute of Science and Technology. Chemistry Web Book. Data from NIST Standard Reference Database, 2011, 69.
25. Rahman A, Al-Reza SM, Sattar MA, Kang SC. Potential roles of essential oil and extracts of *Piper chaba* Hunter to inhibit *Listeria monocytogenes*. Records of Natural Product. 2011; 5(4):228-237.
26. Chieng TC, Assim ZB, Fasihudin BA. Toxicity and antitermite activities of the essential oils from *Piper sarmentosum*. The Malaysian Journal of Analytical Science. 2008; 12(4):234-239.