

Chemical Composition of Essential Oils from Four Vietnamese Species of *Piper* (Piperaceae)

Le D. Hieu^{1,3}, Tran D. Thang^{2*}, Tran M. Hoi³ and Isiaka A. Ogunwande^{4*}

¹ Vinh Medical University, Vinh City, Nghean Province, Vietnam

² Faculty of Chemistry, Vinh University, 182-Le Duan, Vinh City, Nghe An Province, Vietnam

³ Institute of Ecology and Biological Resources, Vietnam Academy of Science and Technology, 18-Hoang Quoc Viet, Cau Giay, Hanoi, Vietnam

⁴ Natural Products Research Unit, Department of Chemistry, Faculty of Science, Lagos State University, Badagry Expressway Ojo, P. M. B. 0001, Lasu Post Office, Ojo, Lagos, Nigeria

Abstract: The chemical composition of essential oils from four *Piper* species, *Piper retrofractum* Vahl., *P. boehmeriaefolium* (Miq.) C. DC., *P. sarmentosum* Roxb., and *P. maclurei* Merr., were analysed by gas chromatography-flame ionization detector (GC-FID) and gas chromatography-mass spectrometry (GC-MS). Nineteen to sixty-four compounds representing 92.0%-98.4% of the total contents were identified in the oil samples. The major constituents identified in *P. retrofractum* leaf oil were benzyl benzoate (14.4%), myrcene (14.4%), bicycloelemene (9.9%), bicyclogermacrene (7.0%) and β -caryophyllene (5.3%). On the other hand, the main constituents of *P. boehmeriaefolium* were α -copaene (28.3%), α -pinene (7.4%) and 1, 8-cineole (5.7%). *P. sarmentosum* showed a very different chemical profile characterized mainly by aromatic compounds and devoid of monoterpene hydrocarbons. The major constituents were benzyl benzoate (49.1%), benzyl alcohol (17.9%), 2-hydroxy-benzoic acid phenylmethyl ester (10.0%) and 2-butenyl-benzene (7.9%). The leaf of *P. maclurei* was characterized by higher amount of (*E*)-cinnamic acid (37.4%) and (*E*)-nerolidol (19.4%). Moreover, (*Z*)-9-octadecanoic acid methyl ester (28.0%), (*E*)-cinnamyl acetate (17.2%), phytol (12.2%) and (*E*)-cinnamaldehyde (8.8%) were the major compounds identified in the stem oil.

Key words: *Piper retrofractum*, *Piper boehmeriaefolium*, *Piper sarmentosum*, *Piper maclurei*, terpenes, fatty acid, aromatic compounds

1 INTRODUCTION

In continuation of our research on the chemical constituents of the essential oils of poorly studied Vietnamese flora¹⁾, we report herein the compounds identified in four *Piper* species. The genus *Piper* belongs to the family Piperaceae, comprising more than 700 species distributed throughout the tropical and subtropical regions of the world²⁾. Most of the species in this genus are aromatic, woody perennial climbers and rarely shrub. The *Piper* species have high commercial, economical and medicinal importance. *Piper retrofractum* Vahl. (Syn. *Piper chaba* Hunter) is a flowering vine cultivated for its fruit, which is usually dried and used as a spice and seasoning. It also has uses in ethnomedicine to cure rheumatic pain and body pain after childbirth. Extracts of the plant are known to possess larvicidal³⁾, safe aphrodisiac³⁾, insecticidal⁴⁾, antioxidant and anti-malaria potentials⁵⁾. Phytochemical studies revealed the characterization of anti-obesity alkaloids⁶⁾, compounds with neurotrophic activity⁷⁾, anti-leish-

manial amides and lignans⁸⁾, antioxidant compounds⁹⁾ and gastoprotective amides¹⁰⁾. Previous analysis on the fruit essential oil identified few monoterpene hydrocarbons, a moderate content of sesquiterpenes and high content of aliphatic hydrocarbons¹¹⁾. α -Humulene (16.4%), caryophyllene oxide (12.2%), viridiflorol (8.1%), globulol (7.4%), β -selinene (7.1%), spathulenol (6.2%), *trans*-nerolidol (5.1%), linalool (4.5%), 3-pentanol (3.5%), tricyclene (2.2%) and *p*-cymene (1.6%) were previously identified as the major compounds of the leaf oil analysed from Bangladesh^{12, 13)}. The leaf oils also displayed potential anti-listerial effect against the strains of *Listeria monocytogenes* (ATCC 19111, 19116, 19118, 19166 and 15313)¹²⁾ and antifungal activity against *Fusarium oxysporum* (KACC 41083), *Phytophthora capsici* (KACC 40157), *Colletotrichum capsici* (KACC 410978), *Fusarium solani* (KACC 41092) and *Rhizoctonia solani* (KACC 40111)¹³⁾.

Piper sarmentosum Roxb., a creeping herb with erect, slender branchlets is used in folk medicine as counter-irri-

* Correspondence to: Isiaka Ajani Ogunwande, Natural Product Research Unit, Department of Chemistry, Faculty of Science, Lagos State University, PMB 0001 Lasu Post Office, Ojo, Lagos, Nigeria

E-mail: thangtd@vinhuni.edu.vn; isiaka.ogunwande@lasu.edu.ng

Accepted December 1, 2013 (received for review October 22, 2013)

Journal of Oleo Science ISSN 1345-8957 print / ISSN 1347-3352 online

http://www.jstage.jst.go.jp/browse/jos/ http://mc.manuscriptcentral.com/jjocs

tants in poultices for headaches and pains in bones. Extracts of *P. sarmentosum* are known to have shown potent anti-nociceptive¹⁴⁾, antipyretic¹⁴⁾ anti-inflammatory^{14, 15)}, anti-acetylcholinesterase¹⁶⁾, antioxidant¹⁷⁾, anticarcinogenic¹⁸⁾ and antimicrobial activities¹⁹⁾. *P. sarmentosum* also decreases atherosclerotic lesions in rats²⁰⁾. In addition, the aqueous extract of *P. sarmentosum* restores ultrastructural integrity in the diabetic cardiovascular tissues²¹⁾. Phytochemical screening revealed the isolation of amides with antioxidant²⁰⁾, anti tuberculosis^{22, 23)} and α -glucosidase inhibitory activities²⁴⁾. In addition, flavonoids with cytotoxic, antiplasmodial and antituberculosis activities^{25, 26)} as well as antimicrobial alkenylphenols²⁷⁾ were characterized previously from the plant. The major compounds of the essential oil from the leaf of *P. sarmentosum* were spathulenol (21.0%), myristicin (18.8%), β -caryophyllene (18.2%) and (*E,E*)-farnesol (10.5%). The leaf oil displayed inhibitory activity against the larvae of *Artemia salina* with LC₅₀ value of 35.2 μ g/mL, and mortality against the subterranean termite, *Coptotermes* sp.²⁸⁾. The toxicity and antitermite activities of the essential oil have been attributed to the presence of spathulenol and β -caryophyllene²⁸⁾. Myristicin (65.22%) and *trans*-caryophyllene (13.89%) were also the major components identified in the leaf essential oil of plants from China²⁹⁾. The essential oil and its component myristicin exhibited strong antifeedant and contact toxicity effects on both the 3rd instar larvae and the imagoes of *Brontispa longissima*²⁹⁾.

Literature information is scanty on the phytochemical constituents and biological activities of *Piper boehmeriaefolium* (Miq.) C. DC. and *Piper maclurei* Merr. However, cytotoxic amide alkaloids have been isolated from *P. boehmeriaefolium*³⁰⁾.

2 EXPERIMENTAL PROCEDURES

2.1 Plant Materials

Leaves of *P. boehmeriaefolium*, *P. sarmentosum* and the leaves and stem of *P. maclurei* were collected from Bạch Mã National Park, Thừa Thiên-Huế, on August 2012; *Piper retrofractum* leaves were collected from Kê Gồ Natural Reserve, Hà Tĩnh Province, Vietnam, on August 2012. Voucher specimens LDH 334, LDH 335, LDH 336 and LDH 337 respectively were deposited at the Botany Museum, Vinh University, Vietnam. Plant samples were air-dried prior to extraction.

2.2 Extraction of essential oils

Air-dried and pulverized samples of each plant material (0.5 Kg each) were subjected to hydrodistillation separately for 4 h at normal pressure according to Vietnamese Pharmacopeia³¹⁾.

2.3 Gas Chromatography (GC) Analysis

Gas chromatography (GC) was performed on an Agilent Technologies HP 6890 Plus Gas chromatograph equipped with a Flame ionization detector (FID) and fitted with HP-5MS columns (both 30 m \times 0.25 mm, film thickness 0.25 μ m, Agilent Technology, Berkshire, United Kingdom). The analytical conditions were: carrier gas H₂ (1 mL/min), injector temperature (PTV, programmed temperature vaporization injection) 250°C, detector temperature 260°C, column temperature programmed from 40°C (2 min hold) to 220°C (10 min hold) at 4°C/min. Samples were injected by splitting and the split ratio was 10:1. The volume injected was 1.0 μ L. Inlet pressure was 6.1 kPa. Each sample was analyzed thrice.

2.4 Gas Chromatography/Mass Spectrometry (GC-MS) Analysis

An Agilent Technologies HP 6890N Plus Chromatograph fitted with a fused silica capillary HP-5MST column (30 m \times 0.25 mm, film thickness 0.25 μ m) and interfaced with a Mass spectrometer HP 5973 MSD was used for the GC-MS analysis. The conditions were the same as described above with He (2 mL/min) as carrier gas. 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/s.

2.5 Identification of the Constituents

The identification of constituents was performed on the basis of retention indices (RI) determined by co-injection with reference to a homologous series of *n*-alkanes (C₉-C₂₄), under identical experimental conditions. Mass spectra of our test oils constituents were compared with known spectra of molecules in NIST 08 Libraries (on ChemStation HP) and Wiley 9th Version and the home-made MS library built up from pure substances and components of known essential oils. The retention indices of all the compounds of the essential oils were also compared with literature values^{32, 33)}. The relative amounts of individual components were calculated based on the GC peak area (FID response) without using correction factors.

3 RESULTS

3.1 Yields of the oils

The plant samples yielded low content of essential oils: 0.2% (v/w; *P. retrofractum*), 0.2% (v/w; *P. boehmeriaefolium*), 0.25% (v/w; *P. sarmentosum*) and 0.25% and 0.20% (v/w; respectively for *P. maclurei* leaf and stem). Oil samples were light yellow coloured.

3.2 Chemical compositions of the oils

Table 1 gives the chemical profiles of volatiles oils of the

Table 1 Chemical compositions of *Piper* oils from Vietnam.

Compounds ^a	RI ^b	RI ^c	Percent composition (% \pm SD)				
			<i>P. r</i>	<i>P. b</i>	<i>P. s</i>	<i>P. ml</i>	<i>P. ms</i>
α -Pinene	939	932	2.3 \pm 0.01	7.4 \pm 0.01	—	2.1 \pm 0.01	3.7 \pm 0.01
Camphene	953	946	1.5 \pm 0.01	0.6 \pm 0.01	—	0.1	0.6 \pm 0.01
Verbenene	962	961	—	0.5 \pm 0.01	—	—	—
Sabinene	976	969	Tr	0.4	—	—	—
β -Pinene	980	974	1.6 \pm 0.01	0.9 \pm 0.01	—	0.4 \pm 0.01	0.7 \pm 0.01
Myrcene	990	988	14.4 \pm 0.02	0.3	—	0.1	—
α -Phellandrene	1006	1002	0.2	1.6 \pm 0.01	—	—	—
δ -3-Carene	1011	1008	0.2	—	—	—	—
α -Terpinene	1017	1014	0.1	0.1	—	—	—
<i>p</i> -Cymene	1022	1020	0.1	1.3 \pm 0.01	—	—	—
Benzyl alcohol	1026	1026	—	—	17.9 \pm 0.02	—	—
Limonene	1032	1024	4.1 \pm 0.01	4.4 \pm 0.01	—	0.9 \pm 0.01	0.7 \pm 0.01
1,8-Cineole	1034	1026	—	5.7 \pm 0.02	—	—	—
(<i>Z</i>)- β -Ocimene	1043	1044	2.0 \pm 0.01	—	—	—	—
(<i>E</i>)- β -Ocimene	1052	1052	3.5 \pm 0.01	0.8 \pm 0.01	—	0.1	—
γ -Terpinene	1061	1054	0.1	0.2	—	—	—
<i>trans</i> -2-Butenylbenzene	1064	1064	—	—	7.9 \pm 0.02	—	—
α -Terpinolene	1090	1086	2.1 \pm 0.01	0.2	—	—	—
Linalool	1100	1095	1.5 \pm 0.01	0.2	0.5 \pm 0.01	1.1 \pm 0.01	3.3 \pm 0.01
<i>allo</i> -Ocimene	1128	1128	2.1 \pm 0.01	—	—	—	—
<i>neo</i> -alloocimene	1140	1140	0.1	—	—	—	—
<i>trans</i> -Pinocarveol	1141	1135	—	1.1 \pm 0.01	—	—	—
<i>trans</i> -Verbenol	1145	1140	—	1.0 \pm 0.01	—	—	—
Camphor	1145	1141	—	1.0 \pm 0.01	—	—	—
Isoborneol	1154	1155	0.1	—	—	—	—
5-Methylundecane	1154	1159	—	—	0.2	—	—
Borneol	1167	1165	—	1.4 \pm 0.01	—	—	—
Terpinen-4-ol	1177	1174	Tr	0.4 \pm 0.01	—	—	—
α -Thujenal	1182	1182	—	1.3 \pm 0.01	—	—	—
<i>p</i> -Cymene-8-ol	1183	1179	—	0.2	—	—	—
α -Terpineol	1189	1186	Tr	1.2 \pm 0.01	—	—	—
Verbenone	1205	1204	—	0.5 \pm 0.01	—	—	—
<i>trans</i> -Carveol	1217	1215	—	0.2	—	—	—
Neral	1238	1235	—	0.2	—	—	—
2-Decenal ^d	1259	1259	0.2	—	—	—	—
2-Phenyl ethyl acetate	1260	1260	—	—	—	0.6 \pm 0.01	—
(<i>E</i>)-Cinnamaldehyde	1266	1270	—	—	—	4.4 \pm 0.01	8.8 \pm 0.02
(<i>E</i>)-Citral	1272	1272	—	0.4 \pm 0.01	—	—	—
Bornyl acetate	1289	1287	0.9 \pm 0.01	0.3 \pm 0.01	—	—	—
Isobornyl acetate	1290	1283	0.3	—	—	—	—
2-Undecanone	1291	1293	0.1	—	—	—	—
Tridecane	1300	1300	0.1	—	—	—	—
(<i>E</i>)-Cinnamyl alcohol	1303	1303	—	—	—	0.1	—
Bicycloelemene	1337	1338	9.9 \pm 0.02	—	0.6 \pm 0.01	4.2 \pm 0.01	—
Eugenol	1356	1356	—	1.3 \pm 0.01	—	0.2	0.6 \pm 0.02

Table 1 Continued.

Compounds ^a	RI ^b	RI ^c	Percent composition (% \pm SD)				
			<i>P. r</i>	<i>P. b</i>	<i>P. s</i>	<i>P. ml</i>	<i>P. ms</i>
Cyclosativene	1371	1369	0.1	—	—	—	—
α -Ylangene	1375	1373	—	—	0.3 \pm 0.01	—	—
Isoledene	1376	1374	0.1	0.6 \pm 0.01	—	—	—
α -Copaene	1377	1374	0.2	28.3 \pm 0.03	0.4 \pm 0.01	0.2	—
β -Patchoulene	1379	1379	0.1	—	—	—	—
Geranyl acetate	1381	1379	0.3	—	—	—	—
β -Bourbonene	1385	1387	0.4 \pm 0.01	—	—	—	—
β -Cubebene	1388	1387	0.9 \pm 0.01	—	—	—	—
β -Elemene	1397	1398	0.6 \pm 0.01	—	—	1.2 \pm 0.01	—
Dodecanal	1408	1408	0.3	—	—	—	—
Methyl eugenol	1407	1410	—	0.2	—	—	—
α -Gurjunene	1412	1409	—	—	—	0.1	—
β -Caryophyllene	1419	1417	5.3 \pm 0.01	0.2	—	2.4 \pm 0.01	0.8 \pm 0.01
β -Gurjunene	1431	1431	0.1	—	—	—	—
γ -Elemene	1437	1434	0.4 \pm 0.01	—	—	—	—
α -Guaiene	1440	1437	—	0.4 \pm 0.01	—	—	—
Aromadendrene	1441	1439	0.4 \pm 0.01	—	0.5 \pm 0.01	—	—
(<i>E</i>)-Cinnamyl acetate	1443	1443	—	—	—	—	17.2 \pm 0.03
α -Humulene	1454	1452	0.7 \pm 0.02	—	0.6 \pm 0.01	—	1.7 \pm 0.01
(<i>E</i>)-Cinnamic acid	1455	1452	—	—	3.6 \pm 0.01	37.4 \pm 0.02	—
1-Dodecanol	1469	1469	0.1	—	—	—	—
γ -Gurjunene	1477	1475	0.1	0.5 \pm 0.01	—	—	1.1 \pm 0.01
α -Amorphene	1485	1483	—	2.8 \pm 0.01	—	—	—
Germacrene D	1485	1484	3.3 \pm 0.01	—	—	—	—
β -Selinene	1486	1489	0.1	—	—	3.1 \pm 0.01	—
<i>n</i> -Pentadecane	1500	1500	—	—	0.9 \pm 0.01	—	0.6 \pm 0.01
Bicyclogermacrene	1500	1500	7.0 \pm 0.01	—	0.8 \pm 0.01	3.4 \pm 0.01	2.4 \pm 0.01
Epizonarene	1505	1501	0.1	1.6 \pm 0.01	—	—	—
β -Bisabolene	1506	1505	0.1	—	—	0.2	—
γ -Cadinene	1514	1513	0.2	0.2	—	0.1	—
<i>cis</i> - Z - α -Bisabolene epoxide	1515	1515	—	—	—	0.4	—
δ -Cadinene	1525	1522	0.5 \pm 0.01	2.1 \pm 0.01	—	0.1	—
14- <i>nor</i> -Cadin-5-en-4-one isomer A	1526	1524	—	2.9 \pm 0.01	—	—	—
<i>cis</i> -Calamenene	1527	1528	—	3.6 \pm 0.01	—	—	—
Guaia-3,9-diene	1556	1556	—	—	—	0.7 \pm 0.01	—
Germacrene B	1561	1559	0.7 \pm 0.01	—	—	0.2	—
(<i>E</i>)-Nerolidol	1563	1561	0.6 \pm 0.01	—	0.7 \pm 0.01	19.4 \pm 0.02	2.8 \pm 0.01
Spathulenol	1578	1577	1.1 \pm 0.01	0.7 \pm 0.01	—	3.2 \pm 0.01	0.9 \pm 0.01
Caryophyllene oxide	1583	1582	—	0.8 \pm 0.01	—	—	—
Viridiflorol	1593	1592	0.9 \pm 0.01	—	—	0.5 \pm 0.01	—
Widdrol	1597	1599	—	1.4 \pm 0.01	—	—	—
β -Oplophenone	1608	1607	—	0.3	—	—	—
Isospathulenol	1625	1631	0.9 \pm 0.01	—	—	0.7 \pm 0.01	—
<i>allo</i> -Aromadendrene epoxide	1623	1639	—	0.3	—	—	—
α -Cadinol	1654	1653	0.3	3.3 \pm 0.01	—	—	—

Table 1 Continued.

Compounds ^a	RI ^b	RI ^c	Percent composition (% \pm SD)				
			<i>P. r</i>	<i>P. b</i>	<i>P. s</i>	<i>P. ml</i>	<i>P. ms</i>
Apiole	1674	1677	0.2	—	—	—	—
Germacrene epoxide	1700	—	—	4.1 \pm 0.01	—	—	—
Farnesol ^d	1718	—	—	—	—	0.2	—
Mint sulfide	1741	1740	0.1	—	—	—	—
Benzyl benzoate	1760	1760	14.4 \pm 0.01	—	49.1 \pm 0.03	3.4 \pm 0.01	—
Tetradecanoic acid	1770	1769	—	—	—	0.2	—
2-hydroxy-Benzoic acid phenyl methyl ester (Benzyl salicylate)	1863	1869	3.8 \pm 0.01	—	10.0 \pm 0.01	—	—
Tetradecanolide ^d	1866	1864	—	—	—	0.4	—
Eicosane	2000	2000	—	—	0.6 \pm 0.01	—	—
(Z)-9-Octadecanoic acid methyl ester	2085	2100	—	—	—	0.1	28.0 \pm 0.04
Heneicosane	2100	2100	—	—	—	—	5.1 \pm 0.02
Phytol	2125	1942	0.1	—	—	0.1	12.2 \pm 0.02
Hexadecanamide	2182	2182	—	—	2.4 \pm 0.01	0.4 \pm 0.01	—
Docosane	2200	2200	—	—	1.6 \pm 0.01	—	2.5 \pm 0.01
Octadecanamide	2349	2349	—	—	—	0.3	—
(Z)-9-Octadecenamide	2398	2398	—	0.2	—	1.9 \pm 0.01	—
(Z)-13-Docosenamide	2499	2499	—	2.6 \pm 0.01	—	2.1 \pm 0.01	—
Pentacosane	2500	2500	—	—	—	0.1	1.0 \pm 0.01
Heptacosane	2700	2700	—	—	0.8 \pm 0.01	—	1.6 \pm 0.01
Total			92.0	92.2	98.4	97.1	98.4
Monoterpene hydrocarbons			34.4	18.7	—	3.8	5.7
Oxygenated monoterpenes			3.1	15.1	4.1	43.0	29.3
Sesquiterpene hydrocarbons			31.4	40.3	3.2	16.1	6.0
Oxygenated sesquiterpenes			3.8	13.8	0.7	24.4	3.7
Diterpenes			0.1	—	—	0.1	12.2
Fatty acids			0.1	—	3.9	0.4	38.8
Aromatic compounds			18.4	—	84.9	4.0	2.1
Phenylpropanoids			—	1.5	—	0.2	0.6
Others			0.7	2.8	2.6	5.1	—

^aElution order on HP-MS capillary column; ^bRetention indices on HP-5 MS column; ^cLiterature retention indices; ^dCorrect isomer not identified; SD Standard Deviation (not determined where value was not indicated); Tr; Trace amount < 0.1%; — Not identified; *P. r* = *P. retrofractum*; *P. b* = *P. boehmeriaefolium*; *P. s* = *P. sarmentosum*; *P. ml* = *P. maclueri* leaf; *P. ms* = *P. maclurei* stem

four *Piper* species analyzed by GC-FID and GC-MS. The main classes of compounds present in the leaf oil of *P. retrofractum* were monoterpene hydrocarbons (34.4%), sesquiterpene hydrocarbons (31.4%) and aromatic compounds (18.4%). The major constituents of the oil were benzyl benzoate (14.4%), myrcene (14.4%), bicycloelemene (9.9%), bicyclogermacrene (7.0%) and β -caryophyllene (5.3%).

The leaf oil of *P. boehmeriaefolium* comprised mainly of α -copaene (28.3%), α -pinene (7.4%) and 1, 8-cineole (5.7%). Quantitatively, the terpene hydrocarbons (18.7% vs. 40.3%) predominated over their oxygenated counterparts (15.1% vs. 13.8%) respectively for monoterpene and

sesquiterpenes, in this oil. This is the first report on the volatile *P. boehmeriaefolium* leaf.

Aromatic compounds (84.9%) represented by benzyl benzoate (49.1%), benzyl alcohol (17.9%), 2-hydroxy-benzoic acid phenylmethyl ester (10.0%) and 2-butenyl-benzene (7.9%) were the major constituents of *P. sarmentosum* leaf oil. The leaf oil of *P. sarmentosum* was devoid of oxygen containing monoterpene compounds.

We have identified (*E*)-cinnamic acid (37.4%) and (*E*)-nerolidol (19.4%) as the major compounds in the leaf oil of *P. maclurei*. Oxygenated monoterpenes (43.0%), sesquiterpene hydrocarbons (15.9%) and oxygenated sesquiterpenes (24.4%) were the main classes of compound present

in the oil. However, the main compounds in the stem oil were mixture of fatty acids represented by (*Z*)-9-octadecanoic acid methyl ester (28.0%), oxygenated monoterpenes namely (*E*)-cinnamyl acetate (17.2%) and (*E*)-cinnamaldehyde (8.8%), and the diterpene, phytol (12.2%). It is noted that fatty acids (38.8%), oxygen containing monoterpenes (29.3%) and diterpenes (12.2%) represents the major class of compounds identified in the oil.

4 DISCUSSION

The compositional patterns of the leaf oil of *P. retrofractum* from Vietnam were entirely different from those of previous analysis elsewhere. The major compounds identified in the previous studies^{12, 13)} were either absent (tricycane, 3-pentanol, caryophyllene oxide and globulol) or detected in low quantities (*p*-cymene, linalool, α -humulene, viridiflorol, β -selinene, spathulenol and (*E*)-nerolidol) in the present report. Moreover, all the major compounds identified in the present analysis were conspicuously absent in the Bangladesh oil samples^{12, 13)}. Also, the chemical profile of *P. sarmentosum* differs both qualitative and quantitatively from previous studies^{28, 29)} because myristicin, β -caryophyllene and (*E, E*)-farnesol, the major compounds of the oils two reports were conspicuously absent in the present study. The variations between the present oil compositions and previous studies may be attributed to factors such as the place of collection, age and nature of the plant, climatic conditions, handling procedures etc.

The compositions of the leaf oils of *P. boehmeriaefolium*, *P. retrofractum*, *P. sarmentosum* and the leaf and stem essential oils of *P. maclurei* from Vietnamese were elucidated and each sample has its compositional pattern different from others. The main compounds identified in the oil samples consisted of ubiquitous terpenes, fatty acids and benzenoid compounds. For example, benzyl alcohol and trans-2-butenylbenzene were identified only in *P. sarmentosum*. A high content of myrcene could be identified only in the leaf oil of *P. retrofractum*. α -Pinene, β -pinene, limonene and β -caryophyllene the common constituents of essential oils were conspicuously absent in the leaf oil of *P. sarmentosum*. Also, (*E*)-cinnamaldehyde, (*E*)-cinnamyl acetate, (*E*)-nerolidol, (*Z*)-9-octadecanoic acid methyl ester, heneicosane and phytol were characteristics of the oils of *P. maclurei*. Benzyl benzoate was identified only in the leaf oil of *P. retrofractum* and *P. sarmentosum*. In addition, 1, 8-cineole and high content of α -copaene were present only in *P. boehmeriaefolium*. It could be seen that linalool was the only compound identified in all the oil samples.

5 CONCLUSION

For the first time, the compositions of the leaf essential oils of the Vietnamese grown *P. retrofractum*, *P. boehmeriaefolium* and *P. sarmentosum* as well as the leaf and stem essential oils of *P. maclurei* were being reported. It is well noted that each sample has its own compositional pattern different from other species.

ACKNOWLEDGMENT

We are grateful to Mrs. Ogunwande Muslimat for the type setting of the manuscript.

References

- 1) Thang, T. D.; Dai, D. N.; Hoi, T. M.; Ogunwande, I. A. Essential oils from five species of Annonaceae from Vietnam. *Nat. Prodt. Commun.* **8**, 239-242 (2013).
- 2) Chansang, U.; Zahiri, N. S.; Bansiddhi, J.; Boonruad, T.; Thongsirak, P.; Mingmuang, J.; Benjapong, N.; Mulla, M.S. Mosquito larvicidal activity of aqueous extracts of long pepper (*Piper retrofractum* vahl) from Thailand. *J. Vector Ecol.* **30**, 195-200 (2005).
- 3) Rahmawati, N.; Bachri, M. S. The aphrodisiac effect and toxicity of combination *Piper retrofractum* L., *Centella asiatica* and *Curcuma domestica* infusion. *Health Sci. J. (Indonesia)* **3**, 19-22 (2012).
- 4) Tripathi, A. K.; Sharma, S.; Kumar, S.; Edison, S.; Ramana, K. V.; Sasikumar, B.; Babu, K. N.; Eapen, S. J. Repellent and insecticidal properties of *Piper retrofractum* against insect pests of crops and stored grain. *Biotechnology of spices, medicinal & aromatic plants. Proceedings of the national seminar on biotechnology of spices and aromatic plants, Calicut, India*, pp. 134-138 (1997).
- 5) Nidhi, R. S.; Mittal, S. V.; Menghani, E. Antioxidant agents alternative source for malaria disease. *Int. J. Appl. Phar.* **4**, 14-16 (2012).
- 6) Kim, K. J.; Lee, M. S.; Jo, K.; Hwang, J. K. Piperidine alkaloids from *Piper retrofractum* Vahl. protect against high-fat diet-induced obesity by regulating lipid metabolism and activating AMP-activated protein kinase. *Biochem. Biophys. Res. Comm.* **411**, 219-225 (2011).
- 7) Kubo, M.; Ishii, R.; Ishino, Y.; Harada, K.; Matsui, N.; Akagi, M.; Kato, E.; Hosoda, S.; Fukuyama, Y. Evaluation of constituents of *Piper retrofractum* fruits on neurotrophic activity. *J. Nat. Prod.* **76**, 769-773 (2013).
- 8) Badiwala, H. S.; Singh, G.; Singh, R.; Dey, C. S.; Sharma, S.S.; Bhutani, K. K.; Singh, I. P. Antileishmanial amides and lignans from *Piper cubeba* and *Piper retrofractum*. *J. Nat. Med.* **61**, 418-420 (2007).

- 9) Saeda, S.; Kikuzaki, H.; Honzawa, M.; Nakatani, N. Chemical constituents of *Piper retrofractum* Vahl and their antioxidant and radical scavenging activities. *ITE Lett. Batter New Tech. Med.* **6**, 566-573 (2005).
- 10) Morikawa, T.; Hisashi, M.; Itadaki, Y.; Yutana, P.; Masayuki, Y. New amides and gastroprotective constituents from the fruit of *Piper chaba*. *Planta Med.* **70**, 152-159 (2004).
- 11) Tewtrakul, S.; Koji, H.; Shigetoshi, K.; Tsuneo, N.; Katsuko, K.; Ken, T. Fruit oil composition of *Piper chaba* Hunt., *P. longum* L. and *P. nigrum* L. *J. Essent. Oil Res.* **12**, 603-608 (2000).
- 12) Rahman, A.; Al-Reza, S. M.; Sattar, M. A.; Kang, S. C. Potential roles of essential oil and extracts of *Piper chaba* Hunter to inhibit *Listeria monocytogenes*. *Rec. Nat. Prod.* **5**, 228-237 (2011).
- 13) Rahman, A.; Al-Reza, S. M.; Kang, S. C. Antifungal activity of essential oil and extracts of *Piper chaba* Hunter against phytopathogenic fungi. *J. Amer. Oil Chem. Soc.* **88**, 573-579 (2011).
- 14) Zakaria, Z. A.; Patahuddin, H.; Mohamad, A. S.; Israf, D.A.; Sulaiman, M. R. *In vivo* anti-nociceptive and anti-inflammatory activities of the aqueous extract of the leaves of *Piper sarmentosum*. *J. Ethnopharm.* **128**, 42-48 (2010).
- 15) Sireeratawong, S.; Supaporn, V.; Suphachai, S.; Arunporn, I.; Kanjana, J. Anti-Inflammatory, anti-nociceptive and antipyretic effects of the ethanol extract from root of *Piper sarmentosum* Roxb. *J. Med. Assoc. Thai.* **93**, S1-S6 (2010).
- 16) Khan, M.; Salah, A. A. E.; Muhammad, M. K.; Nawsher, K. Anti-acetylcholinesterase activity of *Piper sarmentosum* by a continuous immobilized-enzyme Assay. *APCBEE Procedia.* **2**, 199-204 (2012).
- 17) Hafizah, A. H.; Zakaria, Z.; Amom, Z.; Adenan, M. I.; Megat, M.; Nordin, N. A.; Abdullah, M. Z. Piper sarmentosum as an antioxidant on oxidative stress in human umbilical vein endothelial cells induced by hydrogen peroxide. *J. Zhejiang University of Science B*, **11**, 357-365 (2009) doi: 10.1631/jzus.B0900397.
- 18) Ariffin, S. H. Z.; Haryani, W. H.; Wan, O.; Zaidah, Z.; Muhammad, F.; Sahidan, S.; Rohaya, M. A. Intrinsic anticarcinogenic effects of *Piper sarmentosum* ethanolic extract on a human hepatoma cell line. *Cancer Cell Int.* **9**, 6-9 (2009).
- 19) Hanisah, S. P.; Samah, A. O.; Abubakar, S. Selective antimicrobial activity of *Piper sarmentosum* against *Pseudomonas aeruginosa*. *J. Curr. Topics in Nutr. Res.* **9**, 31-34 (2011).
- 20) Amran, A. A.; Zaiton, Z.; Faizah, O.; Srijit, D.; Santhana, R.; Nor-Anita, M. M. N. Aqueous extract of *Piper sarmentosum* decreases atherosclerotic lesions in high cholesterolemic experimental rabbits. *Lipids in Heal. Dis.* **9**, 44-49 (2010).
- 21) Thent, Z. C.; Teoh, S.L.; Srijit, D.; Zaiton, Z. Effect of *Piper sarmentosum* extract on the cardiovascular system of diabetic Sprague-Dawley Rats: Electron microscopic study. *Evid. Based Compl. Alter. Med.* (2012) Article ID 628750. <http://dx.doi.org/10.1155/2012/628750>.
- 22) Kooi-Mow, S.; Choon-Ngo, M.; Li-Ping, H. A new amide alkaloid from the leaves of *Piper sarmentosum*. *J. Asian Nat. Prod. Res.* **11**, 757-760 (2009).
- 23) Hussain, K.; Ismail, Z.; Sadikun, A.; Ibrahim, P. Antioxidant, anti-TB activities, phenolic and amide contents of standardised extracts of *Piper sarmentosum* Roxb. *Nat. Prod. Res.* **23**, 238-249 (2009).
- 24) Damsud, T.; Sirichai, A.; Preecha, P. Three new phenylpropanoyl amides from the leaves of *Piper sarmentosum* and their α -glucosidase inhibitory activities. *Phytochem. Lett.* **6**, 350-354 (2013).
- 25) Ugusman, U.; Zaiton, Z.; Chua, K. H.; Nor-Anita, M. M. N.; Zaleha, A. M. Flavonoids of *Piper sarmentosum* and its cytoprotective effects against oxidative stress. *EXCLI J.* **11**, 705-714 (2012).
- 26) Rukachaisiriku, T.; Siriwananakit, P.; Sukcharoenpho, K.; Wongvein, C.; Ruttanaweang, P.; Wongwattana-vuch, P.; Suksamrarn, A. Chemical constituents and bioactivity of *Piper sarmentosum*. *J. Ethnopharm.* **93**, 173-176 (2004).
- 27) Yang, S. X.; Sun, Q. Y.; Yang, F. M.; Hu, G. W.; Luo, J. F.; Wang, Y. H.; Long, C. L. Sarmentosumols A to F, new mono- and dimeric alkenylphenols from *Piper sarmentosum*. *Planta Med.* **79**, 693-696 (2013).
- 28) Chieng, T. C.; Assim, Z. B.; Fasihuddin, B. A. Toxicity and antitermite activities of the essential oils from *Piper sarmentosum*. *The Malaysian J. Analyt. Sci.* **12**, 234-239 (2008).
- 29) Qin, W.; Shanchun, H.; Chaoxu, L.; Siting, Ch.; Zhengqiang, P. Biological activity of the essential oil from the leaves of *Piper sarmentosum* Roxb. (Piperaceae) and its chemical constituents on *Brontispa longissima* (Gestro) (Coleoptera: Hispididae). *Pest. Biochem. Physiol.* **96**, 132-139 (2010).
- 30) Tang, G. H.; Chen, D. M.; Qiu, B. Y.; Sheng, L.; Wang, Y. H.; Hu, G. W.; Zhao, F. W.; Ma, L. J.; Wang, H.; Huang, Q. Q.; Xu, J. J.; Long, C. L.; Li, J. Cytotoxic amide alkaloids from *Piper boehmeriaefolium*. *J. Nat. Prodt.* **74**, 45-49 (2011).
- 31) Vietnamese Pharmacopoeia. Medical Publishing House, Hanoi, Vietnam, 1-134 (1997).
- 32) Adams, R. P. Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectrometry. 4th Edition, Carol Stream, IL: Allured Publishing (2007).
- 33) Joulain, D.; Koenig, W. A. The Atlas of Spectral Data of Sesquiterpene Hydrocarbons. E. B. Verlag, Hamburg, Germany (1989).