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# COMPOSITION OF ESSENTIAL OILS FROM *Litsea firma* var. *austroannamensis* FROM VIETNAM

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*Litsea* is a genus of evergreen or deciduous trees or shrubs belonging to the family Lauraceae. The genus includes 136 accepted species in tropical and subtropical areas of both hemispheres. They are either dioecious trees or shrubs [1]. The leaves can be either deciduous or evergreen depending on species and are aromatic. The flowers are from greenish to white, greenish-yellow, or yellowish. Our investigations into the volatile composition of Vietnamese species of *Litsea* have yielded several results which have been published [2]. The leaf oil of *L. helferi* was rich in limonene (17.5%), $\beta$-caryophyllene (14.2%), bicyclogermacrene (13.1%), bicycloelemene (12.4%), and $\alpha$-phellandrene (8.0%). The main constituents of *L. ferruginea* leaf oil were sabinene (34.5%), $\alpha$-pinene (10.1%), $\gamma$-terpinene (7.8%), limonene (6.9%), and terpinen-4-ol (6.6%). The significant constituents of the leaf oil of *L. verticillata* included linalool (23.4%), $\alpha$-pinene (26.1%), and $\beta$-pinene (11.7%). In addition, (*E*)-$\beta$-ocimene (57.4%), along with $\alpha$-pinene (7.8%) and $\beta$-pinene (7.3%), were the main constituents in the leaf oil of *L. glutinosa.* The main compounds in the leaf, stem, fruits, and roots oils of *L. cubeba* were (*Z*)-citral (32.9–66.1%), sabinene (1.4–14.2%), limonene (7.0–13.6%), and linalool (1.9–9.5%).

In continuation of our extensive research on the essential oils from Vietnamese flora [3–5], this paper reports the volatile constituents of *Litsea firma* Hook. f. var. *austroannamensis* Liou Ho leaf oil. Literature information is relatively scarce on the volatile and nonvolatile constituents of *L. firma* var. *austroannamensis*.

The leaves of *L. firma* Hook. f. var. *austroannamensis* Liou Ho were collected from Pu Huong Natural Reserve, Nghe An Province (19°20′N 104°50′E), Vietnam, in August 2014. Botanical identification was made by Dr. Nguyen Lam. A voucher specimen, NTL376, was deposited at the Botany Museum, Vinh University, Vietnam. A total of 500 g of the pulverized plant samples was used for the experiment at different times. Essential oils were obtained by hydrodistillation, which was carried out in an all-glass Clevenger-type distillation unit designed according to the Vietnamese Pharmacopoeia [6] as described previously [2–5]. All experiments were done in triplicate.

Gas chromatographic (GC) analysis was performed on an Agilent Technologies HP 6890 Plus Gas chromatograph equipped with an FID and fitted with an HP-5MS column (30 m × 0.25 mm, film thickness 0.25 μm, Agilent Technology). The analytical conditions were: carrier gas He (1 $mL^{-1}$ min), injector temperature (PTV) 250°C, detector temperature 260°C, and column temperature programmed from 40°C (2 min hold) to 220°C (10 min hold) at $4°C^{-1}$ 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). An Agilent Technologies HP 6890N Plus chromatograph fitted with a fused silica capillary HP-5 MS column (30 m × 0.25 mm, film thickness 0.25 μm) and interfaced with a mass spectrometer HP 5973 MSD was used for this experiment, under the same conditions as those used for gas chromatography analysis as described previously [2–5].

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TABLE 1. Volatile Constituents of *L. firma* Hook. f. var. *austroannamensis* from Vietnam

| Compound* | RI | % | Compound | RI | % |
|---|---|---|---|---|---|
| $\alpha$-Pinene | 939 | 2.8 | Aromadendrene | 1441 | 0.5 |
| Sabinene | 976 | 1.1 | $\alpha$-Humulene | 1454 | 1.8 |
| $\beta$-Pinene | 980 | 1.2 | $\gamma$-Muurolene | 1480 | 0.4 |
| $\alpha$-Myrcene | 990 | 0.4 | Germacrene D | 1485 | 1.7 |
| $\alpha$-Phellandrene | 1006 | 2.5 | $\beta$-Selinane | 1486 | 3.7 |
| *p*-Cymene | 1024 | 0.3 | Guaia-1(10),11-diene | 1490 | 4.4 |
| $\beta$-Phellandrene | 1028 | 1.3 | (*E*,*E*)-$\alpha$-Farnesene | 1508 | 1.8 |
| Limonene | 1032 | 0.6 | $\delta$-Cadinene | 1525 | 3.1 |
| 1,8-Cineole | 1034 | 0.8 | (*E*)-Nerolidol | 1563 | 8.9 |
| (*Z*)-$\beta$-Ocimene | 1043 | 2.0 | Ledol | 1569 | 0.6 |
| (*E*)-$\beta$-Ocimene | 1052 | 10.2 | $\alpha$-Guaiol | 1600 | 0.1 |
| Citronellal | 1173 | 5.9 | $\beta$-Acorenol | 1637 | 0.5 |
| $\alpha$-Terpineol | 1189 | 0.7 | $\alpha$-Cadinol | 1654 | 1.4 |
| Nerol | 1232 | 14.4 | 7(11)-Selinen-4$\alpha$-ol | 1693 | 2.5 |
| *cis*-Geraniol | 1253 | 10.2 | Zerumbone | 1756 | 0.8 |
| Linalyl acetate | 1257 | 0.3 | Monoterpene hydrocarbons | | 22.4 |
| (*Z*)-Citral | 1318 | 0.9 | Oxygenated monoterpenes | | 33.2 |
| $\beta$-Ylangene | 1375 | 0.2 | Sesquiterpene hydrocarbons | | 24.8 |
| $\alpha$-Copaene | 1377 | 0.5 | Oxygenated sesquiterpenes | | 14.8 |
| Dodecanal | 1412 | 4.5 | Non-terpenes | | 4.5 |
| $\beta$-Caryophyllene | 1419 | 6.7 | Total | | 99.7 |

*Elution order on HP-5 MS column. RI: Retention indices on HP-5 MS column.

The GC conditions were the same as described above with He (1 $mL^{-1}$ 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 $s^{-1}$.

The identification of constituents from the GC/MS spectra 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. The mass spectral (MS) fragmentation patterns were checked with those of other essential oils of known composition (NIST 08 Libraries) [7] and with those in the literature as described previously [2–5].

The average yield of the essential oils was 0.28% ± 0.01 (v/w, *L. firma* var. *austroannamensis*) calculated on a dry weight basis. The oil sample was light yellow in color. All compounds are listed in order of their elution from the HP-5MS column (Table 1). A total of 36 compounds was identified from the hydrodistilled leaf oil of *L. firma* var. *austroannamensis*. This represents 99.7% of the total oil content. Oxygenated monoterpenes (33.2%) predominate, followed by sesquiterpene hydrocarbons (24.8%), monoterpene hydrocarbons (22.4%), and oxygenated sesquiterpenes (14.8%). The major constituents of the oil were nerol (14.4%), (*E*)-$\beta$-ocimene (10.2%), *cis*-geraniol (10.2%), (*E*)-nerolidol (8.9%), $\beta$-caryophyllene (6.7%), and citronellal (5.9%). The constituents of the essential oil from *L. firma* var. *austroannamensis* are being reported for the first time.

Recent information showed that the compositions of the oils of *L. ferruginea*, *L. verticillata*, *L. cubeba*, and *L. glutinosa* were dominated by monoterpene compounds [2] like the oil of *L. firma* var. *austroannamensis* in the present study. The essential oils of *L. cubeba* from China [8] and several other parts of the world [2] contained monoterpene compounds. The essential oils of *L. akoensis* [9] and *Litsea pungens* [10] also contained monoterpene compounds. However, the main components of the individual species differed. The leaf oils of *L. helferi* [2], *L. acutivena* [11], *L. glutinosa* [12], *L. nakaii* [13], *L. kostermansii* [14], *L. acuminata* [15], *L. linii* [16], and *L. mushaensis* [16] contained sesquiterpene compounds. Moreover, the main components of the individual species differed. On the other hand, mixture of sesquiterpene compounds and fatty acids occurred in *L. coreana* [17]. It can be postulated that both intra and inter species variation could be observed in the essential oils of *Litsea* plants.

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