Volatile Compounds from the Different Organs of Houttuynia cordata and Litsea cubeba (L. citriodora)
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Abstract: The volatile compounds obtained from the different organs of Houttuynia cordata (Saururaceae) and Litsea cubeba (Lauraceae) were analyzed by Gas Chromatography/Mass Spectrometry (GC/MS), Headspace Solid Phase Micro Extraction-Gas Chromatography/Mass Spectrometry (HS-SPME-GC/MS), and GC/olfactometry (GC/O). The major component of all parts of H. cordata is assigned as 4-tridecanone. Each organ produces myrcene as the major monoterpenoid. The major monoterpenes in the rhizomes and roots was β-pinene instead of myrcene. 1-Decanal which was responsible for the unpleasant odor of this plant, was the predominant polyketide in both leaves and stems. The presence of 1-decanal was very poor in flowers, stems collected in summer, rhizomes, and roots. GC/MS analyses were very simple in case of the crude extracts of flowers. The content of sesquiterpenoids was extremely poor. (8Z)-Heptadecene, geranial, and neral were detected as the major components in Litsea cubeba. Odor-contributing components by GC/O analysis of the ether extract of the fresh flowers of L. cubeba were neral and geranial which played an important role in sweet-lemon fragrance of the flowers. The role of a high content of (8Z)-heptadecene was still unknown but it might play a significant role in the dispersion of the volatile monoterpenes hydrocarbons and aldehydes. The flower volatiles of the Japanese L. cubeba were chemically quite different from those of the Chinese same species.

Key words: Houttuynia cordata, 1-decanal, myrcene, β-pinene, Litsea cubeba, geranial, neral, (8Z)-heptadecene

1 INTRODUCTION
1.1. The genus Houttuynia belonging to the Saururaceae has two species in Southeast Asia, one is H. cordata which grows everywhere in Japan and the other is H. emeiensis found in China. It is known that H. cordata emits potent unpleasant odor when one crushes fresh leaves and rhizomes. The strong smelling substances are due to 1-decanal and 1-dodecanal which show strong antimicrobial activity1-3. Thus the eczema and pyesis are cured by patching of the crushed fresh leaves4. Kim et al.5 reported that H. cordata water extract showed antibacterial activity against Salmonella species.

The dried H. cordata is well known as “Jyuyaku” produced from its aerial part which shows diuretic, laxative and antihypertensive effect and has been used for prevention of arterial sclerosis6. These biological activity has been known to be quercitrin and isoquercitrin7. The dried leaves don’t show this strong smell and they are available in the market to use as tea. The chemical analysis of H. cordata was carried out by many researchers because this plant shows not only the above mentioned activity but also anti HSV-1, anti-influenza virus, anti-HIV-1, anticancer, antioxidant, anti-allergic and anti-inflammatory activity8, 9, however, the comparison of its chemical constituents of different organs and the geographical and seasonal chemical variation of these plants has not yet fully been studied.

1.2. Litsea cubeba(L. citriodora) belonging to the Lauraceae grows in the warm location such as Shikoku and Kyushu. The yellow flowers stick to the twigs densely and emits potent sweet lemon-like odor when crushed. The essential oils of L. cubeba shows antimicrobial, antioxidant, insecticidal, and anti-inflammatory activity10, 11.
volatile oil obtained from \( L. \) \( \text{cubeba} \) seeds induces apoptosis and causes cell cycle arrest in lung cancer cell (non-small cell lung carcinoma) \(^{12}\). Recently, Wang and Lu\(^2\) reported the distribution of terpenoids of the essential oils obtained from roots, stems, leaves, flowers, and fruits of the Chinese \( L. \) \( \text{cubeba} \) and their antibacterial activity.

The present paper concerns with the comparative study on volatile compounds of different organs and their geographical and seasonal variation of \( H. \) \( \text{cordata} \) and \( L. \) \( \text{cubeba} \), and comparison of the volatile compounds between the Japanese \( L. \) \( \text{cubeba} \) and Chinese same species.

2 MATERIALS AND METHODS

2.1 Plant materials

\( Houttuynia \) \( \text{cordata} \) \( \) (identified by Yoshinori Asakawa (YA)) were collected by YA in Tokushima Bunri University (TBU) and Asakawa’s botanical gardens in Yamashiro-cho, and Hata-cho and Anan, Tokushima, Japan, in April, May, and August, 2013, respectively. Each fresh plant was divided into flowers, leaves, stems, roots and rhizomes by scissors and crushed by pestle in mortar, followed by extraction with diethyl ether for a week. Each extract was filtered through the Pasteur pipette packed with celite to give characteristic \( H. \) \( \text{cordata} \) odorous oils after the solvent was evaporated at room temperature.

The flowers and leaves of \( Litsea \) \( \text{cubeba} \) (L. \( \text{citriodora} \) \( \)) (identified by YA) were collected by YA in Hinomine, Komatsushima, Tokushima, Japan in March and June, 2013, respectively. The flower petals \( (50 \text{ pieces}, \) \( 4.71 \text{ g}) \) of \( L. \) \( \text{cubeba} \) were macerated with redistilled ether \( (50 \text{ mL}) \) for one night. The extract was concentrated on the water bath at \( 42^\circ \text{C} \) using a Widmer condenser, then removed the solvent at \( 80^\circ \text{C} \) in \( 1 \text{ Torr} \) for one hour and condensed it under \( \text{N}_2 \) stream to give the fragrant oil.

The same flowers \( (0.20 \text{ g}) \) was kept in vial \( (20 \text{ mL}) \) and closed tightly and adsorbed volatile on Divinylbenzene/ Polydimethylsiloxane type fiber (SPME method) at \( 40^\circ \text{C} \) for \( 30 \text{ min} \).

2.2.1 GC/MS condition for \( H. \) \( \text{cordata} \)

GC/MS analysis was carried out by using a 6890N gas chromatograph coupled with a 5973 mass selective detector (Agilent Technologies) on an HP-5MS column \( (30 \text{ m} \times 0.25 \text{ mm i.d., } 0.26 \text{ µm film thickness}) \). The oven temperature program was set at \( 50^\circ \text{C} \) with \( 3 \text{ min initial hold} \) and then raised to \( 250^\circ \text{C} \) at a rate of \( 5^\circ \text{C/min} \). The injector and the ion source temperatures were set at \( 250^\circ \text{C} \) and \( 200^\circ \text{C} \), respectively. The carrier gas was helium with an initial column flow rate of \( 1.0 \text{ mL/min} \). The oven temperature program was set from \( 70^\circ \text{C} \) to \( 220^\circ \text{C} \) at a rate of \( 4^\circ \text{C/min} \).

GC/MS analysis was carried out by using a GCMS-QP2010 Ultra (Shimadzu) equipped with the same column as GC/FID. Electron impact ionization was employed for the mass spectrometry at an ionization energy of \( 70 \text{ eV} \) in scan mode. The injector and the ion source temperatures were kept at \( 250^\circ \text{C} \) and \( 200^\circ \text{C} \), respectively. The carrier gas was helium with a constant pressure of \( 110 \text{ kPa} \). The oven temperature program was the same as described for the GC/FID.

HS-SPME-GC/MS analysis was carried out by using a 6890N Network GC system (Agilent Technologies) equipped with the same column and the same condition as mentioned above. The retention indices \( (RI) \) were calculated relative to \( \text{C}_\text{n}-\text{n}-\text{alkanes} \). Compounds were identified using computer supported spectral library, mass spectra of reference compounds, as well as mass spectra from the literature and our own library databases. NMR spectra were obtained by using a Varian unity-500 (TMS/CDCl\(_3\)).

3 Results and Discussion

3.1 \( Houttuynia \) \( \text{cordata} \)

The NMR spectra of the crude extracts of the leaves and stems of \( H. \) \( \text{cordata} \) showed the presence of a characteristic aldehyde signal. Table 1 shows the distribution of volatile terpenoids and polyketides of each organ of \( H. \) \( \text{cordata} \). The major compound of all parts was assigned as 4-tridecanone. Each organ produced myrcene as the major monoterpenoid. The major monoterpenes in the rhizomes and roots was \( \beta \)-pinene instead of myrcene. 1-Decanal which was responsible for the potent odor of this plant, was the predominant polyketides in both leaves and stems collected in May. On the other hand, the presence of 1-decanal was very poor in flowers, stems collected in summer, rhizomes, and roots. GC/MS analyses are very simple in case of the crude extracts of both flowers and
buds. The major compounds were myrcene and 4-tridecanone. The content of sesquiterpenoids in all of the organs was extremely poor. Only β-elemene, β-caryophyllene, and α-humulene were identified as very minor components. The GC/MS profiles of leaves and stems collected in the different seasons were not significantly changed except for the content of 4-tridecanone.

The leaves collected in May were dried for 2 months at room temperature and then extracted with diethyl ether. The GC/MS profile of the extract was changed. Only 23% decrease of 1-decanal from the crude extract of the leaves collected in May was seen in GC/MS. 1-Nonanal, 1-nonanol, and two sesquiterpenes, spathulenol and β-caryophyllene oxide which might be products newly appeared on GC/MS. H. cordata were collected several locations: TBU and YA’s botanical gardens, Yamashiro-cho, Hata-cho, and Anan, Tokushima, Japan. The volatile compounds of aerial parts were analyzed by HS-SPME-GC/MS found to be the same GC/MS profiles. Kang et al.\textsuperscript{1} reported that the essential oil of H. cordata contained eight monoterpenes and four sesquiterpenes as well as seven polyketides, and 2-undecanone and decanoyl acetaldehyde could be the compounds which play an important role in the flavoring of H. cordata. Lu et al.\textsuperscript{2} identified 2-undecanone, bornyl acetate, and myrcene as three major volatile components of the Chinese H. cordata. Xu et al.\textsuperscript{3} also reported the distributions of monoterpenoids in three different organs, flowers, bracts, and leaves of H. cordata collected in three different locations in China and identified 15 monoterpenoids of which myrcene was the most predominant component and followed by β-phellandrene. Neither n-alkanols nor n-alkanones were detected in all parts of the plant.

The underground parts of 17 wild H. cordata were collected in different valleys and altitudes in Chinese mountains and cultivated for one year and then each aerial part was steam-distilled to give essential oils which were ana-

| Compounds                 | Organs | RI (HP-5MS) | Flowers\textsuperscript{a} | Leaves\textsuperscript{b} | Leaves\textsuperscript{c} | Leaves\textsuperscript{d} | Stems\textsuperscript{b} | Stems\textsuperscript{c} | Rhizomes\textsuperscript{a} | Roots\textsuperscript{a} |
|---------------------------|--------|-------------|-----------------------------|---------------------------|---------------------------|---------------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| α-Pinene                  | 936    | 1.5         | 3.9                         | 1.2                       | 3.4                       | 3.5                       | 0.8                       | 10.3                      |                           |
| Camphene                  | 950    | 0.5         |                             |                           |                           |                           |                           |                           |                           |
| β-Sabinene                | 970    | 2.0         | 3.9                         | 1.2                       | 4.2                       | 7.0                       | 4.2                       | 0.7                       | 7.1                       |
| β-Pinene                  | 978    | 1.6         |                             |                           |                           |                           |                           |                           |                           |
| Myrcene                   | 990    | 18.6        | 17.8                        | 13.3                      | 11.8                      | 17.9                      | 18.6                      | 5.1                       | 6.0                       |
| α-Terpine                   | 1015  | 0.1         |                             |                           |                           |                           |                           |                           |                           |
| Limonene                   | 1024  | 0.5         |                             |                           |                           |                           |                           |                           |                           |
| trans-β-Ocimene           | 1041  | 1.5         | 13.6                        | 0.1                       | 3.7                       | 6.3                       | 2.4                       |                           |                           |
| 1-Nonanal                 | 1103  | 1.6         |                             |                           |                           |                           |                           |                           |                           |
| 1-Nonanol                 | 1171  | 3.7         |                             |                           |                           |                           |                           |                           |                           |
| 1-Decanal                 | 1204  | 0.8         | 40.1                        | 18.2                      | 27.0                      | 11.9                      | 0.8                       | 0.2                       |                           |
| α-Cyclogeranyl acetate    | 1241  | 0.5         |                             |                           |                           |                           |                           |                           |                           |
| Bornyl acetate            | 1270  | 0.9         | 0.5                         |                           |                           |                           |                           |                           |                           |
| 2-Undecanone              | 1294  | 0.7         |                             |                           |                           |                           |                           |                           |                           |
| 1-Undecanal               | 1305  | 0.1         |                             |                           |                           |                           |                           |                           |                           |
| Geranyl acetate           | 1385  | 0.6         |                             |                           |                           |                           |                           |                           |                           |
| β-Elemene                 | 1389  |             |                             |                           |                           |                           |                           |                           |                           |
| 1-Dodecanal               | 1409  | 3.8         |                             |                           |                           |                           |                           |                           |                           |
| β-Caryophyllene           | 1421  | 1.5         |                             |                           |                           |                           |                           |                           |                           |
| α-Humulene                | 1455  | 0.3         |                             |                           |                           |                           |                           |                           |                           |
| 4-Tridecanone             | 1474  | 58.9        |                             |                           |                           |                           |                           |                           |                           |
| β-Caryophyllene oxide     | 1578  |             |                             |                           |                           |                           |                           |                           |                           |
| Spathulenol               | 1580  |             |                             |                           |                           |                           |                           |                           |                           |
| (E)-Phytol                | 2116  |             |                             |                           |                           |                           |                           |                           |                           |
| Unknowns                   |       | 11.3        |                             |                           | 8.0                       | 0.8                       | 23.5                      | 20.3                      | 5.0                       | 2.2                       |
| Total(%)                  |       | 100         |                             |                           | 100                       | 100                       | 100                       | 100                       | 100                       | 100                       |

\textsuperscript{a}: Collected in April 2013; \textsuperscript{b}: in May; \textsuperscript{c}: in August; \textsuperscript{d}: dried for 2 months
Table 2  GC/MS analysis of volatile components of different organs of *Litsea cubeba* (*L. citriodora*).

| Compounds                  | Organs (BC-WAX) | Flowers\(^a\) | Flowers\(^b\) | Flowers\(^c\) | Leaves\(^d\) | Stems\(^e\) |
|----------------------------|-----------------|---------------|---------------|---------------|---------------|--------------|
| Ethyl acetate              | 914             | 0.8           |               |               |               |              |
| Ethanol                    | 944             | 0.6           | 3.5           |               |               |              |
| \(\alpha\)-Pinene          | 1043            | 0.3           | 0.6           |               |               |              |
| \(\beta\)-Pinene           | 1119            | 0.2           | 1.4           |               |               |              |
| Myrcene                    | 1158            | 2.0           |               |               |               |              |
| \(\alpha\)-Phellandrene    | 1174            | 5.4           |               |               |               |              |
| Heptanal                   | 1185            | 0.2           |               |               |               |              |
| Isoamyl alcohol            | 1199            | 1.7           |               |               |               |              |
| Limonene                   | 1207            | 0.3           | 6.3           | 6.0           |               |              |
| *cis*-\(\beta\)-Ocimene   | 1231            | 0.2           | 16.4          |               |               |              |
| *trans*-\(\beta\)-Ocimene | 1251            | 4.7           |               |               |               |              |
| 4-Cymene                   | 1264            | 1.0           |               |               |               |              |
| Acetoin                    | 1275            | 2.2           |               |               |               |              |
| \((E)-4,8\)-Dimethyl-1,3,7-nonatriene | 1298          |               |               |               |               |              |
| Hexanol                    | 1337            | 1.2           |               |               |               |              |
| Acetic acid                | 1431            | 0.4           | 0.7           |               |               |              |
| *trans*-Linalool-3,6-oxide | 1438            | 0.5           | 4.5           |               |               |              |
| Citronellal                | 1464            | 0.7           | 0.6           | 3.3           |               |              |
| Bicycloelemene             | 1484            | 0.5           | 5.4           |               |               |              |
| Pentadecane                | 1500            |               |               |               |               |              |
| Linalool                   | 1531            | 0.9           | 4.5           | 1.1           |               |              |
| \(\beta\)-Copaene         | 1581            |               |               | 0.6           |               |              |
| \(\beta\)-Elemene         | 1584            | 0.3           | 1.3           | 16.6          |               |              |
| \(\beta\)-Caryophyllene   | 1589            | 1.1           | 1.4           | 4.6           | 30.0          | 25.0         |
| Hotrienol                  | 1593            | 0.3           |               |               |               |              |
| \(\alpha\)-Humulene       | 1659            |               |               | 1.7           |               |              |
| *trans*-\(\beta\)-Farnesene | 1659        | 5.9           | 4.1           | 12.7          | 26.7          |              |
| Neral                      | 1663            | 6.5           | 3.3           | 17.5          |               |              |
| \(\gamma\)-Muurolene      | 1677            |               |               | 3.4           |               |              |
| Heptadecane                | 1700            | 9.0           | 4.9           |               |               |              |
| Germacrene D               | 1701            | 1.0           | 0.2           | 3.5           | 15.0          |              |
| Geranial                   | 1710            | 17.5          | 4.6           | 35.3          |               |              |
| \(\beta\)-Bisabolene      | 1718            |               |               | 2.6           |               |              |
| Bisabolene-type sesquiterpene |           |               |               | 1.1           |               |              |
| \((8Z)-Heptadecene         | 1716            | 19.1          | 13.0          | 0.8           |               |              |
| Bicyclogermacrene          | 1719            | 3.5           | 1.0           | 10.0          |               |              |
| \((3E,6E)-\alpha\)-Farnesene | 1737        | 0.3           |               |               |               |              |
| *trans*-Linalool-3,7-oxide | 1744            | 0.3           |               |               |               |              |
| \(\beta\)-Sesquiphellandrene | 1755        | 0.6           |               |               |               |              |
| Nerol                      | 1784            | 0.6           |               |               |               |              |
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In this case, the extracts of Houttuynia cordata were measured by BC-WAX column. On the other hand, the volatile compound -β-farnesene in the stems. They elaborated mainly ubiquitous plant sesquiterpene hydrocarbons, β-elemene, β-caryophyllene, germacrene D, and others. Wang and Liu reported that the major volatiles of the flower were β-pinene (33%), 1,8-cineole (14%), α-pinene (8%), and β-pinene (7%) and neither geranial, neral nor (8Z)-heptadecene which were the major constituents of the Japanese flowers was detected. On contrary, β-pinene and 1,8-cineole were not found in the Chinese flower. The Chinese leaves produce 1,8-cineole as the major component, with β-caryophyllene and β-elemene, as minor components. The Japanese ones did not contain 1,8-cineole, instead, the latter two sesquiterpene hydrocarbons were predominant components. β-Phellandrene (19%) and terpinen-4-ol were the major volatiles in the Chinese stem, however, these monoterpenes were not detected in the Japanese one, otherwise trans-β-farnesene and β-caryophyllene were the predominant. It is noteworthy that the chemical constituents of the root oils of the Chinese Litsea cubeba were chemically very similar to those of the flowers of Japanese Litsea cubeba.

The odor-contributing compounds by GC/O analysis of the fresh flowers of Litsea cubeba are shown in Table 3. A predominant content of neral and geranial played an important role in sweet-lemon fragrance of the flowers. The role of a high content of (8Z)-heptadecene was still unknown but it might play a significant role in the dispersion of the volatile monoterpenoid hydrocarbons and aldehydes.

In conclusion, the volatile compounds obtained from the different organs of Houttuynia cordata (Saururaceae) and Litsea cubeba (Lauraceae) were analyzed by GC/MS, HS-SPME-GC/MS and GC/O. 4-Tridecanone was the major volatile in all parts of different organs of H. cordata. Myrcene was the major monoterpenoids in all organs. The major monoterpenoids in the rhizomes and roots was β-pinene.

### Table 2 GC/MS analysis of volatile components of different organs of Litsea cubeba (L. citriodora).

| Compounds            | Organs (RI BC-WAX) | Flowers<sup>a</sup> | Flowers<sup>b</sup> | Flowers<sup>c</sup> | Leaves<sup>d</sup> | Stems<sup>e</sup> |
|----------------------|--------------------|----------------------|----------------------|----------------------|-------------------|-------------------|
| Geraniol             | 1828               | 0.5                  | 0.8                  |                      |                   |                   |
| Nonadecane           | 1900               | 0.8                  |                      |                      |                   |                   |
| 3,7-Dimethyl-1,5-octadiene-3,7-diol | 1921 | 8.5                  | 0.5                  |                      |                   |                   |
| trans-β-Caryophyllene oxide | 1971 |                      |                      | 1.1                  | 12.0              |                   |
| Unknown + Eicosane   | 2000               | 1.4                  |                      |                      |                   |                   |
| Heneicosane          | 2100               | 0.8                  |                      |                      |                   |                   |
| Spathulenol          | 2103               | 0.3                  |                      | 1.1                  |                   |                   |
| Docosane             | 2200               | 0.3                  |                      |                      |                   |                   |
| (7Z)-8-Hydroxylinalool | 2253 | 0.3                  |                      |                      |                   |                   |
| Tricosane            | 2300               | 2.7                  |                      |                      |                   |                   |
| Tetracosane          | 2400               | 0.8                  |                      |                      |                   |                   |
| Pentacosane          | 2500               | 0.8                  |                      | 0.7                  |                   |                   |
| unknowns             | 12.4               | 11.4                 | 4.3                  | 21.3                 | 32.9              |                   |
| Total (%)            | 100                | 100                  | 100                  | 100                  | 100               | 100               |

<sup>a</sup>: Ether extract (BC-WAX)<sup>b</sup>: SPME (BC-WAX)<sup>c</sup>: Ether extract (HP-5MS)

3.2 Litsea cubeba (L. citriodora)

Table 2 shows the distributions of volatile terpenoids and n-alkanes and n-alkenes detected in different organs of Litsea cubeba. The major components of the ether extract of L. cubeba were measured by BC-WAX column. In this case, (8Z)-heptadecene and geraniol were detected as the major compounds. When the same extract was measured by HP-5MS column, geraniol and neral were identified as the major monoterpenoids, together with trans-β-farnesene. On the other hand, the volatile compound profile which was analyzed by SPME method was totally different from the above methods, except for the presence of a large amount of (8Z)-heptadecene.

The chemical constituents of the leaves and stems of Litsea cubeba were totally different from the dried flowers except for the presence of relatively large amount of trans-β-farnesene in the stems. They elaborated mainly ubiquitous plant sesquiterpene hydrocarbons, β-elemene, β-caryophyllene, germacrene D, and others. Wang and Liu reported that the major volatiles of the flower were β-pinene (33%), 1,8-cineole (14%), α-pinene (8%), and β-pinene (7%) and neither geranial, neral nor (8Z)-heptadecene which were the major constituents of the Japanese flowers was detected. On contrary, β-pinene and 1,8-cineole were not found in the Chinese flower. The Chinese leaves produce 1,8-cineole as the major component, with β-caryophyllene and β-elemene, as minor components. The Japanese ones did not contain 1,8-cineole, instead, the latter two sesquiterpene hydrocarbons were predominant components. β-Phellandrene (19%) and terpinen-4-ol were the major volatiles in the Chinese stem, however, these monoterpenes were not detected in the Japanese one, otherwise trans-β-farnesene and β-caryophyllene were the predominant. It is noteworthy that the chemical constituents of the root oils of the Chinese Litsea cubeba were chemically very similar to those of the flowers of Japanese Litsea cubeba.

The odor-contributing compounds by GC/O analysis of the fresh flowers of Litsea cubeba are shown in Table 3. A predominant content of neral and geranial played an important role in sweet-lemon fragrance of the flowers. The role of a high content of (8Z)-heptadecene was still unknown but it might play a significant role in the dispersion of the volatile monoterpenoid hydrocarbons and aldehydes.

In conclusion, the volatile compounds obtained from the different organs of Houttuynia cordata (Saururaceae) and Litsea cubeba (Lauraceae) were analyzed by GC/MS, HS-SPME-GC/MS and GC/O. 4-Tridecanone was the major volatile in all parts of different organs of H. cordata. Myrcene was the major monoterpenoids in all organs. The major monoterpenoid in the rhizomes and roots was β-pinene.
instead of myrcene. 1-Decanal which was responsible for the potent odor of this plant, was the predominant polyketide in both leaves and stems. The presence of 1-decanal was very poor in flowers, and stems, rhizomes, and roots. The content of sesquiterpenoids was extremely poor. (8Z)-Heptadecene, geranial, and neral were detected as the major components in *Litsea cubeba*. Odor-contributing components by GC/O analysis of the ether extract of the fresh flowers of *L. cubeba* are neral and geranial which played an important role in sweet-lemon fragrance of the flowers. The role of a high content of (8Z)-heptadecene was still unknown but it might play a significant role in the dispersion of the volatile monoterpene hydrocarbons and aldehydes. The flower volatiles of the Japanese *L. cubeba* were chemically quite different from those of the Chinese same species.

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