Chemical Composition of the Essential Oils From Twigs, Leaves, and Cones of *Thuja plicata* and Its Cultivar Varieties “Fastigiata”, “Kornik,” and “Zebrina”

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Abstract

The essential oils from different parts of *Thuja plicata* and its cultivar varieties “Fastigiata”, “Kornik,” and “Zebrina” were analyzed by gas chromatography (GC) and GC/mass spectrometry. More than 80 compounds were identified. The oils from leaves, twigs with leaves, and twigs without leaves contained mainly α-thujone (52.1%-59.2%), fenchone (10.0%-11.3%), and beyerene (3.7%-9.5%), whereas in the cone oil there were α-thujone (35.6%), sabinene (24.0%), and α-pinene (8.3%). The main constituents of the oils from twigs with leaves of “Fastigiata,” “Zebrina,” and “Kornik” cultivars were α-thujone (76.2%, 72.5%, and 67.4%, respectively) and β-thujone (7.6%, 6.2%, and 4.9%, respectively). The oils from cultivars contained more thujones and less fenchone and diterpenes in comparison with *T. plicata* oil.

Keywords

*Thuja plicata*, essential oil composition, α-thujone

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Table 1. Chemical Composition of the Essential Oils From *Thuja Plicata* Twigs With Leaves (A), Leaves (B), Twigs Without Leaves (C), Fruits (D), *T. Plicata* Cv. “Fastigiata” (E), *T. Plicata* Cv. “Kornik” (F), and *T. Plicata* Cv. “Zebrina” (G).

| Compound | RI | LRI | A | B | C | D | E | F | G |
|----------|----|-----|---|---|---|---|---|---|---|
| (Z)-Salvene | 843 | 849 | 0.1 | 0.1 | tr | tr | 0.3 | tr | 0.2 |
| Santolatriene | 897 | 909 | 0.4 | 0.4 | 0.2 | 0.8 | 0.3 | 0.2 | 0.4 |
| Tricyclene | 922 | 927 | 1.1 | 1.0 | 0.6 | 8.3 | 1.1 | 1.0 | 1.4 |
| α-Thuene | 925 | 932 | 0.8 | 0.7 | 0.5 | 0.4 | 0.4 | 0.4 | tr |
| α-Pinene | 928 | 936 | 0.7 | 0.7 | 0.5 | 0.5 | tr | 0.4 | tr |
| Myrcene | 939 | 941 | 5.3 | 5.6 | 1.9 | 24.0 | 4.8 | 3.0 | 7.1 |
| α-Phellandrene | 944 | 950 | 1.2 | 1.1 | 0.1 | 3.2 | 1.4 | 1.1 | 1.8 |
| α-Terpinene | 968 | 973 | 5.3 | 5.6 | 1.9 | 24.0 | 4.8 | 3.0 | 7.1 |
| p-Cymene | 970 | 978 | 0.1 | 0.1 | 0.1 | 0.3 | 0.1 | 0.1 | 0.1 |
| β-Phellandrene | 982 | 987 | 0.8 | 0.7 | 0.5 | 0.4 | 0.4 | 0.4 | tr |
| Limonene | 984 | 990 | 0.7 | 0.7 | 0.5 | 0.5 | tr | 0.3 | tr |
| γ-Terpinene | 997 | 1002 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| trans-Sabinene hydrate | 1006 | 1013 | 0.4 | 0.3 | 0.2 | 0.6 | 0.4 | 0.3 | 0.4 |
| Camphor | 1009 | 1015 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| β-Phellandrene | 1018 | 1023 | 1.2 | 1.1 | 0.7 | 1.5 | 0.8 | 1.0 | 0.9 |
| β-Pinene | 1020 | 1025 | 0.5 | 0.4 | 0.5 | 0.9 | 0.6 | 0.5 | 0.6 |
| trans-Sabinene hydrate | 1021 | 1026 | 0.4 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| Fenchone | 1028 | 1034 | 11.3 | 10.6 | 10.0 | 3.8 | 5.1 | 1.1 | 1.2 |
| Terpinolene | 1035 | 1041 | 0.0 | 0.2 | 0.1 | 0.2 | 0.2 | 0.2 | 0.2 |
| α-Thujone | 1036 | 1042 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| β-Thujone | 1038 | 1044 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| α-Campholenal | 1039 | 1045 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| cis-p-Menth-2-en-1-ol | 1040 | 1046 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| trans-p-Menth-2-en-1-ol | 1041 | 1047 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| Camphor | 1042 | 1048 | 2.1 | 2.0 | 2.1 | 0.8 | 0.1 | 0.3 | 0.1 |
| Sabina ketone | 1043 | 1049 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Camphene hydrate | 1044 | 1050 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| Thujol | 1045 | 1051 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Bornol | 1046 | 1052 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| Terpinen-4-ol | 1047 | 1053 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| α-Terpineol | 1048 | 1054 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| Myrtenol | 1049 | 1055 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Verbenone | 1050 | 1056 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| trans-Piperitol | 1051 | 1057 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| trans-Verbenol | 1052 | 1058 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Camphene hydrate | 1053 | 1059 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| Fenchyl acetate | 1054 | 1060 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| Thymol methyl ether | 1055 | 1061 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Carvacrol methyl ether | 1056 | 1062 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| cis-Sabinene hydrate | 1057 | 1063 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| trans-Sabinene hydrate | 1058 | 1064 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Bornyl acetate | 1059 | 1065 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 | 0.3 |
| trans-Sabinyl acetate | 1060 | 1066 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Neothujyl acetate | 1061 | 1067 | 0.4 | 0.4 | 0.4 | 0.4 | 0.4 | 0.4 | 0.4 |
| Menthol acetate | 1062 | 1068 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Terpinen-4-yl acetate | 1063 | 1069 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| α-Cubebene | 1064 | 1070 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| Geranyl acetate | 1065 | 1071 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| α-Copaene | 1066 | 1072 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |

(Continued)
| Compound                        | RI     | LRI     | A   | B   | C   | D   | E   | F   | G   |
|--------------------------------|--------|---------|-----|-----|-----|-----|-----|-----|-----|
| β-Elemene                      | 1387   | 1389    |     |     |     |     |     |     |     |
| (E)-β-Caryophyllene            | 1417   | 1421    |     |     |     |     |     |     |     |
| Guai-6,9-diene                 | 1438   | 1443    | tr  | tr  |     |     |     |     |     |
| (E)-β-Farnesene                | 1446   | 1446    |     |     |     |     |     |     |     |
| α-Humulene                     | 1452   | 1455    |     |     |     |     |     | tr  | tr  |
| Germacrene D                   | 1475   | 1479    |     |     |     |     |     |     | tr  |
| α-Muurolene                    | 1492   | 1496    |     |     |     |     |     |     |     |
| γ-Cadinene                     | 1505   | 1507    | tr  | tr  |     |     |     | tr  | tr  |
| δ-Cadinene                     | 1515   | 1520    | tr  | 0.1 | tr  | tr  | 0.1 | tr  | 0.1 |
| Elemol                         | 1538   | 1541    |     |     |     |     |     |     |     |
| Furoelepargone B               | 1561   | 1567   | 0.3 | 0.3 | 0.4 | 0.2 | tr  | 0.2 | 0.1 |
| Germacrene D-4-ol              | 1566   | 1571    | 0.1 | 0.1 |     | 0.5 | tr  | 0.1 |     |
| Caryophyllene oxide            | 1571   | 1578    | 0.1 | 0.1 | 0.1 | 0.1 | tr  | 0.4 | tr  |
| β-Olopone                      | 1589   | 1595    | tr  | tr  | tr  | tr  |     |     |     |
| Humulene epoxide II            | 1594   | 1602    | tr  | tr  | 0.1 | 0.1 | tr  | 0.1 | tr  |
| ω-α-Muurolol                   | 1626   | 1633    | tr  | tr  | 0.1 | tr  | 0.1 |     |     |
| α-Cadinol                      | 1638   | 1643    | 0.1 | 0.1 | 0.3 | tr  | 0.1 |     |     |
| 14-Hydroxy-β-caryophyllene     | 1652   | 1656    |     |     |     |     |     |     |     |
| Olopone                        | 1706   | 1703   | tr  | tr  | tr  | tr  | 0.1 | tr  |     |
| Rimuene                        | 1896   | 1907    | 1.3 | 1.7 | 0.6 | 0.1 | 0.1 | 0.9 | 0.2 |
| Beyereone                      | 1944   | 1951    | 6.7 | 9.5 | 3.7 | 0.3 | 0.3 | 2.2 | 0.4 |
| Pimara-8(14),15-diene          | 1950   | 1955    | tr  | tr  | tr  | tr  |     |     |     |
| Sandaracopimara-8(14),15-diene | 1964   | 1967   | 0.1 | 0.1 | 0.1 |     |     |     |     |
| Manool oxide                   | 1999   | 2007    | tr  | tr  | tr  | tr  |     |     |     |
| (E)-Biformene                  | 2002   | 2008    |     |     |     |     |     |     |     |
| Kaur-15-ene                    | 2006   | 2011    | tr  | tr  | tr  | tr  |     |     |     |
| Abieta-8,12-diene              | 2017   | 2022   |     |     |     |     |     |     |     |
| Abiatriene                     | 2039   | 2046    | 0.1 | 0.1 | 0.1 |     | tr  | tr  |     |
| Abieta-7,13-diene              | 2079   | 2084    | tr  | tr  | 0.1 | 0.7 | tr  |     |     |
| Abieta-8(14),13(15)-diene      | 2147   | 2152    |     |     |     |     |     |     | 0.1 |
| Abieta-6(8),14-dien-18-al      | 2213   | 2215   |     |     |     |     |     |     |     |
| Dehydroabietal                 | 2234   | 2241   |     |     |     |     |     |     | 0.1 |
| α-α-Totarol                    | 2249   | 2252    | tr  | tr  | 0.7 |     |     |     |     |
| Abietol                        | 2256   | 2261    |     |     | 0.1 | 1.3 |     |     | 0.1 |
| Beyeren-19-yl acetate          | 2308   | 2316    |     |     |     |     | 0.1 | 0.6 | 1.2 |
| 4-α-Abietol                    | 2329   | 2341   |     |     |     |     |     | 0.4 |     |
| Abieta-8,13(15)-dien-18-al     | 2340   | 2347   |     |     |     |     |     | 0.1 |     |
| Abietol                        | 2378   | 2389   |     |     |     |     |     |     | 0.3 |
| Total                          |         |         | 99.5| 99.6| 96.2| 98.2| 99.2| 99.7| 99.3|
| Monoterpene hydrocarbons       |         |         | 12.2| 12.0| 6.2 | 41.3| 10.3| 8.5 | 13.5|
| Oxygenated monoterpenes        |         |         | 78.5| 75.5| 84.0| 49.1| 88.4| 86.8| 83.4|
| Sesquiterpene hydrocarbons     |         |         |     |     |     | 0.1 |     |     |     |
| Oxygenated sesquiterpenes      |         |         | 0.6 | 0.6 | 0.6 | 1.7 | tr  | 0.7 | 0.4 |
| Diterpene hydrocarbons         |         |         | 8.2 | 11.4| 4.6 | 1.6 | 0.4 | 3.1 | 0.6 |
| Oxygenated diterpenes          |         |         |     |     |     | 0.8 | 2.5 | 0.1 | 0.6 | 1.3 |
| Oil yield                      |         |         | 0.7 | 0.8 | 0.2 | 1.2 | 1.6 | 0.8 | 1.2 |

LRI, literature retention indices on DB-1 column according to MassFinder 3.1; RI, retention indices on Rtx-1 column; tr, trace (<0.05%).

- Literature retention indices on DB-5 column according to Adams.\(^1\)
- Literature retention indices according to NIST (\(^2\)BP-1 column, \(^3\)HP-1 column, \(^4\)CP Sil 5 column, \(^5\)HP-5 column, \(^6\)DB-1 column).
- Literature retention indices on DB-5 column according to Adams.\(^1\)
- Literature retention indices according to NIST (\(^2\)BP-1 column, \(^3\)HP-1 column, \(^4\)CP Sil 5 column, \(^5\)HP-5 column, \(^6\)DB-1 column).
The essential oil from *T. plicata* contained several times more oil than twigs without leaves (0.8% and 0.2%, respectively). Cones contained 1.2% of the oil. “Fastigiata” and “Zebrina” cultivars were about 2 times richer in oil than *T. plicata* (1.6%, 1.2%, and 0.7%, respectively). More than 80 compounds representing 96% to 99% of the oils were identified, with about 40 for the first time in this species. The composition of the oils from leaves, twigs with leaves, and twigs without leaves of *T. plicata* were similar. The main constituents of the oils were α-thujone (52.1%-59.2%), fenchone (10.0%-11.3%), and beyerene (3.7-9.5). A few components were present in concentrations of 2%-5%: sabine, β-thujone, camphor, terpinen-4-ol, and bornyl acetate, while others constituted less than 1%. The oil from cones differed in composition. In comparison with those oils, it contained lower amounts of α-thujone (35.6%), fenchone (3.8%), and beyerene (0.3%), but the amounts of the monoterpene hydrocarbons sabine (24.0%) and α-pinene (8.3%) were much higher. Additionally, it contained many sesquiterpenes and diterpenes, which were not found in the oil from leaves and twigs. Thus, both leaves and twigs with leaves give oils rich in thujones. The presence of cones in the raw material may reduce the content of thujones in the oil.

The oils of “Fastigiata” and “Zebrina” varieties had similar composition with α-thujone (76.2% and 72.5%, respectively), β-thujone (7.6% and 6.2%, respectively), and sabine (4.8% and 7.1%, respectively) as the main constituents. “Kornik” oil contained α-thujone (67.4%), fenchone (7.5%), and β-thujone (4.9%). The chemical profiles of the oils from twigs with leaves of *T. plicata* and its cultivars “Fastigiata,” “Kornik,” and “Zebrina” were comparable having α-thujone as the dominant compound. On the other hand, significant differences between the oils were observed. The oils of “Fastigiata” and “Zebrina” in comparison with *T. plicata* oil contained more thujones (78%-83% and 57%, respectively) and less fenchone (trace and 11%, respectively) and diterpenes (1% and 8%, respectively).

With regard to the previously reported *T. plicata* oils, the composition of “Fastigiata” and “Zebrina” oils were similar to that of laboratory distilled Canadian oils and commercial oil from the United States, with high level of α-thujone (72%-82%), no fenchone, and no diterpenes. In turn, the *T. plicata* oil investigated by us resembled commercial Canadian oil with similar contents of α-thujone (54%), fenchone (15%), and β-thujone (6%-8%), but differed in content of diterpenes. Such high levels of diterpenes were previously found only in the oil from *T. plicata* var. “Gracilis.”

Our investigation showed that the yield and quantitative composition of *T. plicata* oil depended on the plant organ and variety. Twigs with leaves of *T. plicata*, particularly from “Fastigiata” and “Zebrina” cultivars, due to their high amount of essential oil and high content of thujones can be a good source of natural thujones for the pharmaceutical and fragrance industries.

### Experimental

#### Plant Material

*Thuja plicata* Donn ex D. Don and its cultivars “Fastigiata,” “Kornik,” and “Zebrina” were collected in the Lodz Botanical Garden, Poland, in October 2016. The voucher specimens (Tpl/2016, Tplf/2016, Tplk/2016 and Tplz/2016) have been deposited in the Herbarium of the Institute of General Food Chemistry, Lodz University of Technology.

The fresh branches of *T. plicata* were divided into twigs with leaves, leaves, twigs without leaves, and cones. These samples of fresh plant material (100.0 g each), as well as twigs with leaves of “Fastigiata,” “Kornik,” and “Zebrina” cultivars were cut into small pieces (0.5 cm long) and separately hydrodistilled in a Clevenger-type apparatus for 3 hours to obtain essential oils. The procedure was replicated 3 times. After decanting and drying over anhydrous MgSO4, the oils were stored at low temperature (5°C) before analysis. The oils had pale yellow color and an intensive herbal-camphoraceous aroma.

#### Analysis of the Essential Oils

The chemical composition of the oils was determined by simultaneous gas chromatography (GC)/flame detection (FID) and GC/mass spectrometry (MS) analyses using an MS-FID splitter (SGE Analytical Science) and a Trace GC ultra gas chromatograph coupled with a DSQ II mass spectrometer (Thermo Electron Corporation). The apparatus was equipped with an apolar capillary column Rtx-1 (dimethylpolysiloxane), 60 m × 0.25 mm × 0.25 µm film thickness (Restek, Bellafonte, PA, USA). The oven temperature was programmed from 50 to 300°C with rises of 4°C/min. Injector temperature was 280°C, detector temperature 300°C, ion source temperature 200°C, carrier gas helium with constant pressure 300 kPa, ionization voltage 70 eV, mass range 33-420 amu.

Identification of components was based on comparison of their retention indices relative to *n*-alkanes (C8-C26) and their mass spectra with those of commercial libraries (MassFinder 3.1, NIST 98.2, Wiley Registry of Mass Spectral Data 8th ed.) and literature.

A quantitative analysis (expressed as percentage of each component) was carried out by peak normalization measurement without correction factors.

#### Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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