



AkiNik

American Journal of Essential Oils and Natural Products

Available online at www.essencejournal.com

ISSN: 2321-9114
 AJEONP 2017; 5(1): 15-17
 © 2017 AkiNik Publications
 Received: 04-11-2016
 Accepted: 05-12-2016

William N Setzer
 Department of Chemistry,
 University of Alabama in
 Huntsville, Huntsville, USA

Prabodh Satyal
 Department of Chemistry,
 University of Alabama in
 Huntsville, Huntsville, USA

William N. Setzer
 Department of Chemistry,
 University of Alabama in
 Huntsville, Huntsville, USA

Chemical composition of the bark essential oil of *Cercis canadensis* L. (Fabaceae)

Kelly Marie Steinberg, Prabodh Satyal and William N Setzer

Abstract

The volatile components from the bark of *Cercis canadensis* L. (Fabaceae) were obtained by hydrodistillation and analyzed by gas chromatography–mass spectrometry as well as enantioselective gas chromatography. The bark volatiles were dominated by C₆ fatty-acid-derived compounds 1-hexanol (23.3%), hexanoic acid (18.2%), and (2*E*)-hexenoic acid (3.4%). The concentration of monoterpenoids in *C. canadensis* bark was low (4.1%), but did allow determination of the enantiomeric distribution of α -pinene (racemic), limonene (exclusively *d*-enantiomer), linalool and α -terpineol (predominantly *l*-stereoisomers).

Keywords: Essential oil composition, *Cercis canadensis*, enantiomeric distribution, Native American ethnopharmacology

1. Introduction

Cercis canadensis L. (Fabaceae), commonly known as “eastern redbud”, ranges throughout the southeastern United States. The tree was used by Native Americans for food as well as medicine. The bark of *C. canadensis* was used to make a tea as a remedy for whooping cough (pertussis), congestion, fever, and vomiting ^[1]. As part of our continuing interest in Cherokee traditional medicines ^[2-5], we have investigated the essential oil composition, including enantiomeric distribution of monoterpenoids, of the bark of *C. canadensis*. To our knowledge, this is the first examination of the essential oil of *C. canadensis* and the first report of enantiomeric distribution of monoterpenoids in the Fabaceae.

2. Materials and Methods

2.1 Plant Material

Branches of *C. canadensis* were collected from Huntsville, Alabama (34° 38' 46" N, 86° 33' 27" W, 191 m elevation) on November 5, 2016. The bark was stripped from the limbs and finely chopped. The chopped bark (87.78 g) was hydrodistilled using a Likens-Nickerson apparatus, with continuous extraction with dichloromethane, for 4 h. *C. canadensis* bark essential oil (1.6709 g, 1.904% yield) was obtained as a colorless liquid, which was stored at –20 °C until further analysis.

2.2 Gas Chromatography – Mass Spectrometry

GC-MS analysis was carried out using a Shimadzu GCMS-QP2010 Ultra. This instrument was operated in the electron impact (EI) mode set at electron energy 70eV with a scan range of 40-400 amu, a scan rate of 3.0 scans per second, and with GC-MS solution software. A ZB-5 fused silica capillary column with a (5% phenyl)-polymethylsiloxane stationary phase and a film thickness of 0.25 μ m was used as the GC column. Helium was used as the carrier gas and the pressure was set at 80 psi with a flow rate of 1.37 mL/min on the column head. The temperature of the injector was set at 250 °C and the temperature of the ion source was set at 200 °C. The temperature of the GC oven was programmed to be 50 °C initially and was programmed to increase at a rate of 2 °C/ min to a final temperature of 260 °C. The sample was prepared with CH₂Cl₂ in a 5% w/v solution. Then, 0.1 μ L of the solution was injected into the instrument with the splitting mode with a split ratio of 30:1. The retention indices were determined by reference to a homologous series of *n*-alkanes. The components of each essential oil sample were identified based on their retention indices and mass spectral fragmentation patterns compared to reference literature ^[6] and our in-house library.

Correspondence:
William N Setzer
 Department of Chemistry,
 University of Alabama in
 Huntsville, Huntsville, USA

2.3 Chiral Gas Chromatography – Mass Spectrometry

The essential oil from *C. canadensis* was also analyzed enantioselectively with a Shimadzu GCMS-QP2010S. The instrument was operated in the EI mode with electron energy of 70 eV, a scan range of 40–400 amu, and a scan rate of 3.0 scans/s. The capillary column used was a Restek B-Dex 325 with film dimensions of 30 m × 0.25 mm ID × 0.25 μm. The temperature of the oven was programmed to start at 50 °C and to rise at a rate of 1.5 °C/min to a final temperature of 120 °C. Then, the oven was raised to 200 °C at a faster rate of 2 °C/min and maintained for 5 min. The carrier gas, helium, was set at a constant flow rate of 1.8 mL/min. A solution (0.1 μL) of 3% w/v of the essential oil in CH₂Cl₂ was injected into the instrument in split mode with the split ratio of 1:45.

3. Results and Discussion

The composition of *C. canadensis* bark essential oil is compiled in Table 1. A total of 57 compounds were identified in *C. canadensis* bark oil accounting for 97.9% of the composition. The essential oil was dominated by fatty acid-derived compounds (76.0%), including 1-hexanol (23.3%), hexanoic acid (18.2%), (2*E*)-hexenoic acid (3.4%), oleamide (3.2%), and 1-docosanol (3.0%). *n*-Alkanes (10.2%), and aromatics (5.5%), were also present. Fatty acids and fatty acid-derived alcohols and aldehydes have sometimes been shown to be a feature of essential oils of the Fabaceae. For example, the bark essential oil of *Cassia bakeriana* from Brazil revealed 51.3% fatty acids, 23.2% aldehydes, and 11.1% alcohols [7]; although it did not contain any C₆ compounds, the bark essential oil of *Inga laurina* from Brazil was composed of 46.8% fatty acids [8]; and the leaf essential

oil of *Robinia pseudoacacia* growing in Poland was composed of 65.1% aliphatic alcohols [9].

Nakamura and Hatanaka have shown that C₆ alcohols and aldehydes are bacteriostatic to several different strains of bacteria [10], but, in general, longer chain alcohols are more active [11, 12]. Huang and co-workers have demonstrated that medium-chain fatty acids as well as long-chain fatty acids exhibit antimicrobial activity; hexanoic acid was particularly active against *Candida albicans*, *Fusobacterium nucleatum*, and *Streptococcus mutans* [13].

Although the concentration of monoterpenoids was somewhat low, only 4.1%, it was possible to determine their enantiomeric distribution using chiral gas chromatography – mass spectrometry. α -Pinene was present as a racemic mixture, but limonene was present as the pure (+)-enantiomer. The (–)-enantiomers were the major stereoisomers for linalool (65%) and α -terpineol (70%). This, we believe, represents the first examination of the enantiomeric distribution of monoterpenoids in the Fabaceae.

4. Conclusions

The bark essential oil of *Cercis canadensis* was found to be rich in medium-chain and long-chain alcohols, aldehydes, and carboxylic acids, in particular C₆ compounds. The presence of these compounds may account for the traditional use of *C. canadensis* bark by the Cherokee and other Native Americans. Although monoterpenoid concentrations were low, the chiral gas chromatographic analysis was able to discern the relative enantiomeric concentrations of α -pinene, limonene, linalool, and α -terpineol.

Table 1: Volatile components of *Cercis canadensis* bark.

| RI ^a | Components | % | RI ^a | Components | % |
|-----------------|-----------------------------|------------------|-----------------|------------------------------|------|
| 799 | Hexanal | 0.9 | 1349 | Eugenol | 0.7 |
| 832 | 2-Methylbutanoic acid | 1.0 | 1397 | Methyleugenol | 0.6 |
| 844 | (3 <i>Z</i>)-Hexenol | 0.3 | 1418 | β -Caryophyllene | 0.7 |
| 849 | (3 <i>E</i>)-Hexenol | 2.2 | 1446 | Geranyl acetone | 0.5 |
| 859 | (2 <i>Z</i>)-Hexenol | 0.7 | 1508 | Dicyclohexyl ketone | 0.5 |
| 862 | 1-Hexanol | 23.3 | 1580 | Caryophyllene oxide | 1.1 |
| 885 | (4 <i>Z</i>)-Hepten-2-ol | 0.8 | 1600 | <i>n</i> -Hexadecane | 0.6 |
| 900 | 2-Heptanol | 1.7 | 1607 | 1,10-di- <i>epi</i> -Cubenol | 0.7 |
| 931 | α -Pinene | 0.2 ^b | 1654 | α -Cadinol | 0.9 |
| 967 | 1-Heptanol | 0.6 | 1700 | <i>n</i> -Heptadecane | 0.8 |
| 975 | Hexanoic acid | 18.2 | 1793 | 1-Octadecene | 0.5 |
| 977 | 1-Octen-3-ol | 1.5 | 1800 | <i>n</i> -Octadecane | 0.6 |
| 1003 | Octanal | 0.4 | 1894 | 1-Nonadecene | 0.7 |
| 1008 | (2 <i>E</i>)-Hexenoic acid | 3.4 | 1900 | <i>n</i> -Nonadecane | 0.9 |
| 1028 | Limonene | 2.0 ^c | 1956 | Palmitic acid | 2.5 |
| 1032 | Benzyl alcohol | 1.3 | 1986 | 1-Eicosene | 0.7 |
| 1042 | Benzene acetaldehyde | 1.0 | 2000 | <i>n</i> -Eicosane | 0.8 |
| 1069 | 1-Octanol | 1.2 | 2100 | <i>n</i> -Heneicosane | 2.7 |
| 1083 | <i>o</i> -Guaiacol | 0.3 | 2110 | Methyl linoleate | 1.2 |
| 1092 | Unidentified ^d | 2.1 | 2200 | <i>n</i> -Docosane | 0.7 |
| 1099 | Linalool | 0.8 ^e | 2300 | <i>n</i> -Tricosane | 0.9 |
| 1104 | Nonanal | 1.8 | 2371 | Oleamide | 3.2 |
| 1111 | 2-Phenylethanol | 0.6 | 2454 | Docosanal | 0.7 |
| 1159 | (2 <i>E</i>)-Nonenal | 0.6 | 2517 | 1-Docosanol | 3.0 |
| 1164 | Octanoic acid | 1.1 | 2600 | <i>n</i> -Hexacosane | 0.4 |
| 1194 | α -Terpineol | 0.7 ^f | 2700 | <i>n</i> -Heptacosane | 0.6 |
| 1205 | Decanal | 1.2 | 2806 | (<i>E,E,E</i>)-Squalene | 0.5 |
| 1230 | 2-Coumaranone | 0.5 | 2900 | <i>n</i> -Nonacosane | 1.2 |
| 1248 | Chavicol + Geraniol | 0.8 | | Total Identified | 97.8 |

^a RI = "Retention Index", determined with respect to a series of *n*-alkanes on a ZB-5 column. ^b 50% (+)- α -pinene / 50% (–)- α -pinene. ^c 100% (+)-limonene. ^d Unidentified: MS, m/e 196(2%), 128(2%), 101(23%), 99(44%), 83(52%), 71(48%), 55(100%), 45(38%), 43(74%), 41(34%). ^e 35% (+)-linalool / 65% (–)-linalool. ^f 30% (+)- α -terpineol / 70% (–)- α -terpineol.

5. References

- 1 Moerman DE. Native American Ethnobotany. Timber Press, Portland, Oregon, 1998.
- 2 Woods KE, Chhetri BK, Jones CD, Goel N, Setzer WN. Bioactivities and compositions of *Betula nigra* essential oils. *Journal of Medicinally Active Plants*, 2013; 2(1):1-9.
- 3 Stewart CD, Jones CD, Setzer WN. Leaf essential oil compositions of *Rudbeckia fulgida* Aiton, *Rudbeckia hirta* L., and *Symphytotrichum novae-angliae* (L.) G.L. Nesom (Asteraceae). *American Journal of Essential Oils and Natural Products*, 2014; 2(1):36-38.
- 4 Stewart CD, Jones CD, Setzer WN. Essential oil compositions of *Juniperus virginiana* and *Pinus virginiana*, two important trees in Cherokee traditional medicine. *American Journal of Essential Oils and Natural Products*. 2014; 2(2):17-24.
- 5 Setzer WN. Chemical composition of the leaf essential oil of *Lindera benzoin* growing in north Alabama. *American Journal of Essential Oils and Natural Products*, 2016; 4(3):1-3.
- 6 Adams RP. Identification of Essential Oil Components by Gas Chromatography / Mass Spectrometry, 4th Ed., Allured Publishing Corporation, Carol Stream, Illinois.
- 7 Cunha LCS, de Moraes SAL, Martins CHG, Martins MM, Chang R, de Aquino FJT *et al.* Chemical composition, cytotoxic and antimicrobial activity of essential oils from *Cassia bakeriana* Craib. against aerobic and anaerobic oral pathogens. *Molecules*, 2013; 18:4588-4598.
- 8 Furtado FB, de Aquino FJT, Nascimento EA, Martins CM, de Moraes SAL, Chang R *et al.* Seasonal variation of the chemical composition and antimicrobial and cytotoxic activities of the essential oils from *Inga laurina* (Sw.) Willd. *Molecules*, 2014; 19:4560-4577.
- 9 Kicel A, Olszewska MA, Owczarek A, Wolbiś M. Preliminary study on the composition of volatile fraction of fresh flowers and leaves of *Robinia pseudoacacia* L. growing in Poland. *Acta Poloniae Pharmaceutica – Drug Research*, 2015; 72:1217-1222.
- 10 Nakamura S, Hatanaka A. Green-leaf-derived C6-aroma compounds with potent antibacterial action that act on both Gram-negative and Gram-positive bacteria. *Journal of Agricultural and Food Chemistry*, 2002; 50:7639-7644.
- 11 Kubo I, Muroi H, Kubo A. Antibacterial activity of long-chain alcohols against *Streptococcus mutans*. *Journal of Agricultural and Food Chemistry*, 1993; 41:2447-2450.
- 12 Mukherjee K, Tribedi P, Mukhopadhyay B, Sil AK. Antibacterial activity of long-chain fatty alcohols against mycobacteria. *FEMS Microbiology Letters*, 2012; 338:177-183.
- 13 Huang CB, Alimova Y, Myers TM, Ebersole JL. Short- and medium-chain fatty acids exhibit antimicrobial activity for oral microorganisms. *Archives of Oral Biology*, 2011; 56:650-654.