Title
THE LIGNANS OF Myristica otoba

Author
Nemethy, E. K.

Publication Date
1985

Lawrence Berkeley Laboratory
UNIVERSITY OF CALIFORNIA

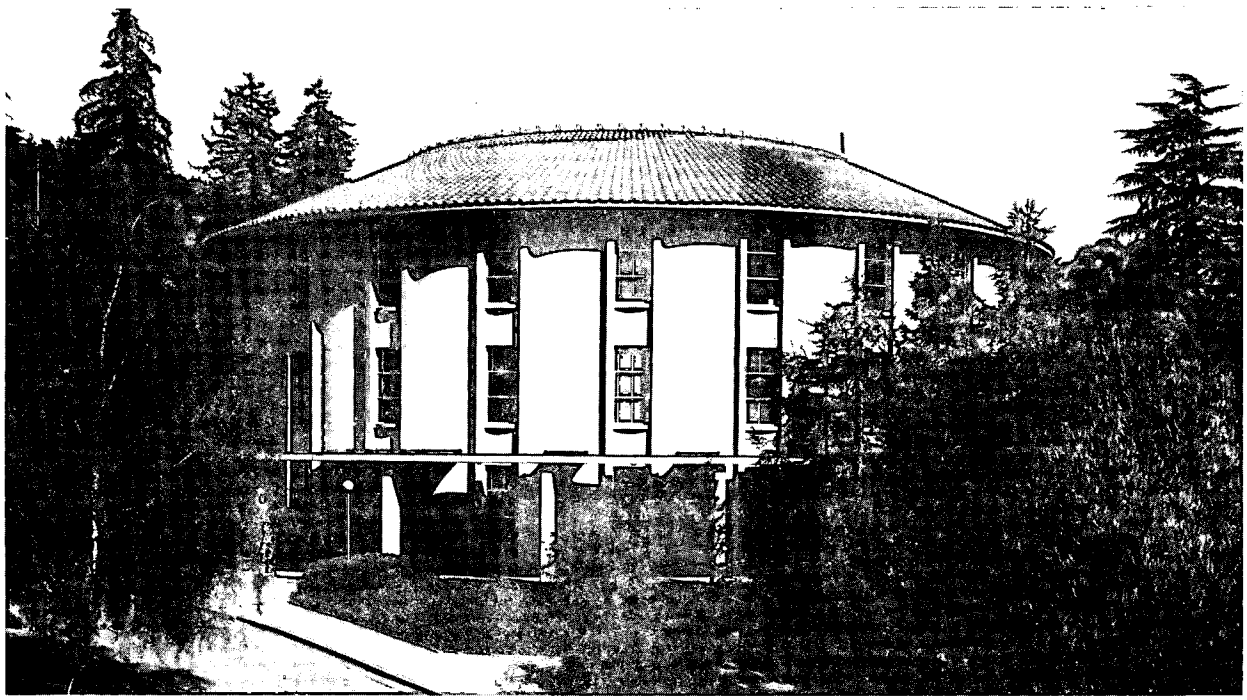
CHEMICAL BIODYNAMICS DIVISION

Submitted to Phytochemistry

THE LIGNANS OF *Myristica otoba*

E.K. Nemethy, R. Lago, D. Hawkins, and M. Calvin

January 1985



Prepared for the U.S. Department of Energy under Contract DE-AC03-76SF00098

DISCLAIMER

The Lignans of *Myristica otoba*

E. K. Nemethy, R. Lago*, D. Hawkins and M. Calvin

Lawrence Berkeley Laboratory and Department of Chemistry
University of California, Berkeley, California 94720

Key Word Index: *Myristica otoba*, Myrisiticaceae, aryltetralin lignans, galbulin, galcatin, isogalcatin.

Abstract: The neutral fraction of the oil of *Myristica otoba* fruits afforded a series of seven lignans of the aryltetralin type.

* Present address: CTAA-EMBRAPA, Av. das Americas 29501, Guaratiba-RJ-23.000, Rio de Janeiro, BRASIL

From the neutral fraction of the oil of Myristica otoba fruits we have isolated a series of seven aryltetralin lignans (I-VII, Fig. 1). The minor constituents galbulin (III) and galcatin (IV) have been isolated from other sources (1-3), but have not been identified as components of Myristica otoba oil. Isogalcatin (isootobain, V) has also been identified as a component of M. otoba by Wallace (7). The lignan otobain [7,8-methylenedioxy)-2,3 -dimethyl-1-(3'4'-methylenedioxyphenyl)-tetralin] has been isolated from this source previously, and its structure was elucidated (4-8). The absolute configuration of otobain has been suggested by Klyne et al. based on NMR and ORD spectroscopy (9). We have not been able to detect the presence of otobain at the 0.5% level in our sample of the oil; the two major aryltetralin lignans we have identified are I and II, comprising 37 and 12% of the neutral fraction, respectively.

Compounds I-VII have been separated and purified by HPLC. Structure assignments were based on mass spectra and ^{1}H NMR at 250 and 500 MHz. Coupling constants, where appropriate, were determined by decoupling experiments.

Compounds III, IV and V had identical MS and ^{1}H NMR spectra to those reported in the literature (7,8,10,11). The substitution pattern on the phenyl rings was confirmed by the observed spectrum, showing two singlets for H-5 and H-8 on ring A and two doublets with coupling constants 8.2 (ortho) and 2.2 (meta) for H-5' and H-2', respectively, as well as an AB pattern for H-6' with J = 8.2 and 2.2 Hz. the Assignment of the methylenedioxy groups in IV and V was based on the known chemical

shift difference between ring A or ring C substitution (6). The observed coupling constants, $J_{1,2}$ = 10 Hz and $J_{3,4}$ = 10 Hz for compounds III, IV and V confirm the all trans stereochemistry with the methyl groups and the pendant phenyl substituent all pseudo-equatorial.

The aromatic region of the NMR spectrum of I and II was identical to that of III, IV and V, indicating the same substitution pattern. However, the smaller coupling constants for H-1-2 (5.5 Hz) as well as for H-3-4_β and H-4_α (7 and 5 Hz) indicated a different stereochemistry from that of compounds III, IV and V. By decoupling experiments at 500 MHz (irradiation of the CH_3), $J_{2,3}$ was determined to be $\sim$3 Hz, indicating that H-2 and H-3 are pseudo-equatorial. In the spectra of both I and II the two methyl resonances occurred at δ= 0.9. No deshielding of either methyl groups by phenyl ring C was observed as in the spectra of compounds III, IV and V. Examination of all possible conformers indicated the cis-trans stereochemistry for I and II as shown in Fig. 1.

The ^{1}H NMR of compound VI showed a different aromatic region for ring A protons. Instead of two singlets, two doublets with J= 8.2 Hz were observed, indicating the substitution pattern as shown. The methylenedioxy protons were nonequivalent (J= 1.4 Hz), due to the proximity of phenyl ring C. The observed coupling constants for H-1-2 (3 Hz) and for H-3-$4_{\alpha,\beta}$(12 and 6 Hz) indicated the relative stereochemistry.

The MS of compound VII showed a molecular ion at m/e 358, two mass units higher than I and III, but the same number of aromatic protons in

the NMR. Examination of the aromatic region indicated the substitution pattern as shown.

Two additional minor components of the aryltetralin lignan type were detected by HPLC, but these were present in insufficient quantities for NMR analysis. Mass spectra indicated that one is isomeric with compound I and the other with compound III.

Gilchrist (4) has reported the presence of a phenolic mixture in otoba fat, from which he isolated one compound, otobain. By HPLC we have successfully resolved this mixture and identified new lignans of Myristica otoba. We could not detect otobain in this mixture, which may be due to a different state of ripeness of the fruit, or to the fact that we have used a fresh fruit extract, whereas other workers have used commercial otoba fat.

Acknowledgements: We would like to thank Dr. J. D. Bultman, Environmental Sciences Division, Naval Research Laboratory, Washington, D. C. for the supply of Myristica otoba fruits. This work was supported by the Assistant Secretary for Conservation and Renewable Energy, Office of Renewable Energy, Biomass Energy Technologies Division of the U.S. Department of Energy under Contract No. DE-AC03-76SF00098.

Experimental

Myristica otoba fruits (without shells)were cut into small pieces and extracted with CH_2Cl_2 for 4 hrs at ambient temperature. After removal of the solvent, a viscous oil, 74% of the fresh weight, was obtained. SiO_2 chromatography eluting with hexane, hexane:CH_2Cl_2, hexane:diethyl ether, ethyl ether, $CHCl_3$, CH_3OH, yielded a glyceride fraction (86% of the oil) and a neutral fraction (14%). The neutral fraction was separated into two components by SiO_2 chromatography, eluting with hexane, hexane:ether (100:0 to 1:1), and $CHCl_3$:MeOH (1:1). Fraction I (20% of the neutral fraction) eluted with 95:5 hexane:ether, R_F = 0.41 on SiO_2 TLC, hexane:ether 1:1; fraction II (40%) eluted from the column with 80:20 hexane:ether, R_F = 0.24. Infrared spectra of Fractions I and II showed no hydroxyl or carbonyl absorption, but a C-O-C stretch at 1270 cm^{-1}. Fractions I and II were resolved into individual components by HPLC using an Altex 5 μ ODS column with a mobile phase of 75:25 MeOH:H_2O. Compound III was further purified from a minor, isomeric component on a 5 μ Altex SiO_2 column, using hexane:ether 87:13 as a mobile phase.

^1H NMR spectra were recorded at 250 and 500MHz in $CDCl_3$ with TMS as an internal standard. Mass spectra were obtained on a Finnigan 4000 mass spectrometer at 70 ev; high resolution data were obtained on a Cratos MS 50 instrument. Measured masses were within 5 ppm of calculated values.

6,7-dimethyoxy-2$_{\beta}$ 3$_{\alpha}$ -dimethyl-1β-(3',4'-dimethoxyphenyl)-tetralin (I). (37% of neutral fraction).

^{1}H NMR: δ 6.75 (d; J = 8.2 Hz, H-5'), 6.6 (s;H-5), 6.57 (d; J = 2.2 Hz, H-2'), 6.52 (dd; J = 8.2, 2.2 Hz, H-6), 6.35 (s; H-8), 3.87 3.85, 3.80, 3.67 (s; -OCH_3), 3.6 (d; J = 5.5 Hz, H-1), 2.85 (dd; J = 16.5, 5.4 Hz, H-4_α), 2.45 (dd; J = 16.5, 7 Hz, H-4_β), 2.18 (m; H-3); 1.95 (m; J = 2.3, 3 Hz, H-2), 0.9 (d; J = 6 Hz,-CH_3), 0.9 (d; J = 6 Hz,-CH_3).
MS (m/z): 356 ($C_{22}H_{28}O_4$), 299, 285, 269 ($C_{17}H_{17}O_3$ b.p.), 254, 238, 203, 165, 151, 135, 91, 69.

6,7-methylenedioxy-$2_\beta 3_\alpha$-dimethyl-1β-(3',4'-dimethoxyphenyl)-tetralin (II). (12%)
^{1}H NMR: δ 6.75 (d; J = 8.2 Hz, H-5'), 6.6 (s; H-5), 6.57 (d; J = 2.2 Hz H-2'), 6.52 (dd; J = 8.2, 2.2 Hz, H-6'), 6.35 (s; -H-8) 5.85 (s; -OCH_2O-), 3.85 (s; OCH_3), 3.82 (s; OCH_3), 3.6 (d; J = 6 Hz,H-1), 2.9 (dd; J = 16.5, 7.3 Hz, H-4_α), 2.5 (dd; J = 16.5, 5.3 Hz, H-4_β), 1.9-2.1 (m; H-2, H-3), 0.9 (d; J - 6 Hz, -CH_3), 0.9 (d; J - 6 Hz, -CH_3).
MS (m/z): 340 ($C_{21}H_{24}O_4$) 309, 283, 269, 253 ($C_{16}H_{13}O_3$ b.p.), 238, 222, 209, 202, 187, 165, 152.

6,7-dimethoxy-$2_\alpha 3_\beta$-dimethyl-1 β(3',4'-dimethoxyphenyl)-tetralin (III). (5%).
^{1}H NMR: δ 6.81 (d; J = 8.2 Hz,H-5'), 6.67 (dd; J = 8.2, 2.2 Hz, H-6'), 6.57 (s; H-5), 6.55 (d; J = 2.2 Hz, H-2'), 6.16 (s; H-8), 3.89, 3.85, 3.8, 3.6 (s; OCH_3), 3.42 (d; J = 10 Hz, H-1), 2.76 (dd; J = 16.3, 4.2 Hz, H-4_α), 2.62 (dd; J = 16.3, 10.4 Hz, H-4_β), 1.45-1.65 (m; H-2, H-3), 1.1 (d; J = 6 Hz,-CH_3), 0.9 (d; J = 6 Hz,-CH_3).
MS (m/z): 356 ($C_{22}H_{28}O_4$), 299, 269 ($C_{17}H_{17}O_3$ b.p.) 238, 203, 165, 151, 135, 91, 69.

6,7-methylenedioxy-$2_\alpha 3_\beta$-dimethyl-1β-(3',4'-dimethoxyphenyl)-tetralin (IV). (4%).

^{1}H NMR: δ 6.75 (d; J = 8 Hz H-5'), 6.7 (d; J = 2 Hz, H-2'), 6.55 (dd; J =8.2, 2.2 Hz H-6'), 6.5 (s; H-5), 6.25 (s; H-8), 5.8 (s;-OCH_2O-), 3.87, 3.83 (s; OCH_3), 3.4 (d; J = 9.8 Hz, H-1), 2.75 (dd; J = 16.5, 4.1 Hz, H-4_α), 2.6 (dd; J = 16.5, 10 Hz, H-4_β), 1.7-1.5 (m; H-2, H-3), 1.08 (d; J = 6 Hz,-CH_3), 0.9 (d; J = 6 Hz,-CH_3).

MS (m/z): 340 ($C_{21}H_{24}O_4$), 309, 283, 253 ($C_{16}H_{13}O_3$ b.p.) 239, 223, 187, 151.

6,7-dimethyoxy-2_α,3_β-dimethyl-1β (3',4'-methylenedioxyphenyl)-tetralin. (V). (2%).

^{1}H NMR δ 6.8 (d; J = 8.2 Hz, H-5'), 6.72 (dd; J = 8.2, 2.2 Hz, H-6'), 6.6 (d; J = 2.2 Hz, H-2'), 6.57 (s; H-5), 6.3 (s; H-8), 5.95 (s;-OCH_2O-), 3.87, 3.8 (s;-OCH_3), 3.4 (d; J = 9.8 Hz, H-1), 2.75 (dd; J = 16.5, 4.1 Hz, H-4_α), 2.6 (dd; J = 16.5, 10 Hz H-4_β), 1.7-1.5 (m; H-2, H-3), 1.08 (d; J = 6 H-2, CH_3), 0.9 (d; J = 6 Hz, CH_3).

MS (m/z): 340 ($C_{21}H_{24}O_4$), 309, 283, 253 ($C_{16}H_{13}O_3$ b.p.) 239, 223, 187, 151.

7,8-methylenedioxy-$2_\alpha 3_\beta$-dimethyl-1β-(3',4'-dimethoxyphenyl)-tetralin. (VI) (1.2%).

^{1}H NMR: δ 6.72 (d; J = 8.3 Hz, H-5') 6.70 (d, J = 8.0 H_2, H-6),, 6.67 (d; J = 8.0 Hz, H-5), 6.52 (d; J = 2.2 Hz, H-2'), 6.43 (dd; J = 8.2, 2.2 Hz, H-6'), 5.77 (d; J = 1.4 Hz,-O-CH_2-O-), 3.9 (d; J = 3 Hz, H-1), 3.84, 3.82 (s;-OCH_3), 2.77 (dd; J = 16.5, 12 Hz, H-4_α), 2.5 (dd; J = 16.5, 5.8 Hz, H-4_β) 1.7-1.5 (m; H-2, H-3), 1.0 (d; J = 6 Hz, CH_3), 0.9 (d; J = 6 Hz, CH_3).

MS (m/z): 340 ($C_{21}H_{24}O_4$ b.p.), 309, 283, 269, 253 ($C_{16}H_{13}O_3$), 239, 223, 202, 187, 151.

2,3-dimethyl-1,4- bis-(3,4-dimethyoxyphenyl)-butane (VII). (0.2%). ^1H NMR: δ 6.82-6.6 (m; ArH); 3.9, 3.85 (s; OCH_3) 2.76 (dd; J = 16.3, 4.1 Hz, H-1, H-4) 2.3 (dd; J = 16.3, 7.8 Hz, H-1, H-4), 1.8-1.6 (m; H-2, H-3), 0.9 (d;-CH_3).

MS (m/z): 358 ($C_{22}H_{30}O_4$), 269, 206, 179, 165, 151 (b.p).

References

1. Hughes, G. K. and Ritchie, E. (1954). Aust. J. Chem. 7, 104.
2. Gottlieb, O. R., Maia, J.G.S. and De S. Ribeiro, N. M. (1976). Phytochem. 15, 773.
3. Hobbs, T. T. and King, F. E. (1960). J. Chem. Soc. 4732.
4. Gilchrist, T., Hodges, R. and Porte, A. L. (1962). J. Chem. Soc. 1780.
5. Adjangba, S. M. and Billet, D. (1962). Bull. Soc. Chim. France, 1970.
6. Bhacca, N. S. And Stevenson, R. (1963). J. Org. Chem. 28, 1638.
7. Wallace, R., Porte, A. L. ahd Hodges, R. (1962). J. Chem. Soc. 1445.
8. Kohen, F., Maclean, I. and Stevenson, R. (1966). J. Chem. Soc. (C), 1775.
9. Klyne, W., Stevenson, R. and Swan, R. J. (1966). J. Chem. Soc. (C) 893.
10. Adjangba, M. S. (1963). Bull. Soc. Chim. France, 1007.
11. Pelter, A. (1968). J. Chem. Soc. (C) 74.

This report was done with support from the Department of Energy. Any conclusions or opinions expressed in this report represent solely those of the author(s) and not necessarily those of The Regents of the University of California, the Lawrence Berkeley Laboratory or the Department of Energy.

Reference to a company or product name does not imply approval or recommendation of the product by the University of California or the U.S. Department of Energy to the exclusion of others that may be suitable.

TECHNICAL INFORMATION DEPARTMENT
LAWRENCE BERKELEY LABORATORY
UNIVERSITY OF CALIFORNIA
BERKELEY, CALIFORNIA 94720