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LIGNANS OF *MYRISTICA OTOBA*

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Key Word Index—*Myristica otoba*; Myristicaceae; 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.

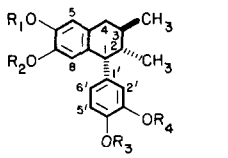
From the neutral fraction of the oil of *Myristica otoba* fruits we have isolated a series of seven aryltetralin lignans (**1**–**7**). The minor constituents galbulin (**3**) and galcatin (**4**) have been isolated from other sources [1–3], but have not been identified as components of *M. otoba* oil. Isogalcatin (isootobain, **5**) has also been identified as a component of *M. otoba* by Wallace [4]. 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 **1** and **2**, comprising 37 and 12% of the neutral fraction, respectively.

Compounds **1**–**7** 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. The absolute configurations shown are based on ^{1}H NMR and CD data. Hulbert and Klyne [9, 10] have carried out extensive studies on the circular dichroism of aryltetralin lignans, and have concluded that the sign of the first Cotton effect reflects the configuration of the pendant aryl group. All 4β compounds give a negative first Cotton effect and all 4α compounds a positive one.

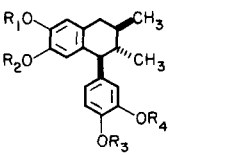
Compounds **3**–**5** had identical mass and ^{1}H NMR spectra to those reported in the literature [7, 8, 11, 12]. The substitution pattern on the phenyl rings was confirmed by the observed spectrum, showing two singlets for H-5 and H-8 on ring A and two doublets with coupling constants 8.2 (*ortho*) 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 **4** and **5** was based on the known chemical shift difference between ring A or ring C substitution [7]. The observed coupling constants, $J_{1,2}$ = 10 Hz and $J_{3,4}$ = 10 Hz for compounds **3**–**5** 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 **1** and **2** was identical to that of **3**–**5**, indicating the same substitution pattern. However, the smaller coupling constants for H-1-2 (5.5 Hz) as well as for H-3-4 and H-4 (7 and 5 Hz) indicated a different stereochemistry from that of compounds **3**–**5**. By decoupling experiments at 500 MHz (irradiation of the methyl), $J_{2,3}$ was determined to be 3 Hz, indicating that H-2 and H-3 are pseudo-equatorial. In the spectra of both **1** and **2** the two methyl resonances occurred at δ0.9. No deshielding of either methyl groups by phenyl ring C was observed as in the spectra of compounds **3**–**5**. Examination of all possible conformers indicated the *cis–trans* stereochemistry for **1** and **2** as shown in the formulae.

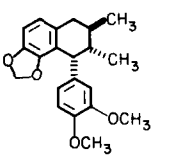
The ^{1}H NMR of compound **6** showed a different aromatic region for ring A protons. Instead of two singlets, two doublets with J = 8.2 Hz were observed, indicating the substitution pattern as shown. The methylenedioxy protons were nonequivalent (J = 1.4 Hz), due to the proximity of phenyl ring C. The observed coupling



	R_1	R_2	R_3	R_4
1	CH_3	CH_3	CH_3	CH_3
2	$-CH_2-$		CH_3	CH_3



	R_1	R_2	R_3	R_4
3	CH_3	CH_3	CH_3	CH_3
4	$-CH_2-$		CH_3	CH_3
5	CH_3	CH_3	$-CH_2-$	



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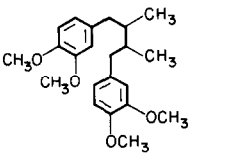

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constants for H-1-2 (3 Hz) and for H-3-4$_{\alpha,\beta}$ (12 and 6 Hz) indicated the relative stereochemistry.

The mass spectrum of compound 7 showed a $[M]^+$ at m/z 358, two mu higher than those in **1** and **3**, 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 **1**, the other with compound **3**.

Gilchrist [5] has reported the presence of 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 *M. 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.

EXPERIMENTAL

Myristica otoba fruits were collected in Panama. The shells were removed, the fruits cut into small pieces and extracted with CH_2Cl_2 for 4 hr at room temp. After removal of solvent, a viscous oil, 74% fr. wt, was obtained. Silica gel chromatography eluting with hexane, hexane–CH_2Cl_2, hexane–Et_2O, Et_2O, $CHCl_3$–MeOH (97:3) yielded a glyceride fraction (86% of the oil) and a neutral fraction (14%). The neutral fraction was separated into two components by silica gel chromatography, eluting with hexane, hexane–Et_2O (100:0 to 1:1), and $CHCl_3$–MeOH (1:1). Fraction I (20% of the neutral fraction) eluted with hexane–Et_2O (19:1), R_f = 0.41 on silica gel TLC, hexane–Et_2O (1:1); fraction II (40%) eluted from the column with hexane–Et_2O (4:1), R_f = 0.24. Infrared spectra of Fractions I and II showed no OH 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 MeOH H_2O (3:1). Compound **3** was further purified from a minor, isomeric component on a 5 μ Altex SiO_2 column, using hexane–Et_2O (87:13) as mobile phase.

^{1}H NMR spectra were recorded at 250 and 500 MHz in $CDCl_3$ with TMS as int. standard. MS were obtained at 70 eV; high resolution data were within 5 ppm of calculated values. CD spectra were taken in MeOH.

6,7-Dimethoxy-2,3-dimethyl-1α-(3′,4′-dimethoxyphenyl)-tetralin (**1**, 37% of neutral fraction). ^{1}H NMR: δ6.75 (*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*, OMe), 3.6 (*d*, J = 5.5 Hz, H-1), 2.85 (*dd*, J = 16.5, 5.4 Hz, H-4), 2.45 (*dd*, J = 16.5, 7 Hz, H-4), 2.18 (*m*, H-3), 1.95 (*m*, J = 2.3, 3 Hz, H-2), 0.9 (*d*, J = 6 Hz, Me), 0.9 (*d*, J = 6 Hz, Me). MS (*m/z*): 356 ($C_{22}H_{28}O_4$), 299, 285, 269 ($C_{17}H_{17}O_3$ base), 254, 238, 203, 165, 151, 135, 91, 69. CD: $[\theta]_{285}$ + 5520, $[\theta]_{263}$ − 6380.

6,7-Methylenedioxy-2α,3β-dimethyl-1α-(3′,4′-dimethoxyphenyl)-tetralin, (**2**, 12%). ^{1}H NMR: δ6.75 (*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*, OMe), 3.82 (*s*, OMe), 3.6 (*d*, J = 6 Hz, H-1), 2.9 (*dd*, J = 16.5, 7.3 Hz, H-4), 2.5 (*dd*, J = 16.5, 5.3 Hz, H-4), 1.9–2.1 (*m*, H-2, H-3), 0.9 (*d*, J = 6 Hz, Me), 0.9 (*d*, J = 6 Hz, Me). MS (*m/z*): 340 ($C_{21}H_{24}O_4$) 309, 283, 269, 253 ($C_{16}H_{13}O_3$ base), 238, 222, 209, 202, 187, 165, 152. CD: $[\theta]_{288}$ + 8600; $[\theta]_{264}$ − 5700.

6,7-Dimethoxy-2α,3β-dimethyl-1α-(3′,4′-dimethoxyphenyl)-tetralin (**3**, 5%). ^{1}H NMR: δ6.81 (*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*, OMe), 3.42 (*d*, J = 10 Hz, H-1), 2.76 (*dd*, J = 16.3, 4.2 Hz, H-4), 2.62 (*dd*, J = 16.3, 10.4 Hz, H-4), 1.45 1.65 (*m*, H-2, H-3), 1.1 (*d*, J = 6 Hz, Me), 0.9 (*d*, J = 6 Hz, Me). MS (*m/z*): 356 ($C_{22}H_{28}O_4$), 299, 269 ($C_{17}H_{17}O_3$ base), 238, 203, 165, 151, 135, 91, 69. CD: $[\theta]_{284}$ − 8040, $[\theta]_{263}$ + 4780.

6,7-Methylenedioxy-2α,3β-dimethyl-1β-(3′,4′-dimethoxyphenyl)-tetralin (**4**, 4%). ^{1}H NMR: δ6.75 (*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*, OMe), 3.4 (*d*, J = 9.8 Hz, H-1), 2.75 (*dd*, J = 16.5, 4.1 Hz, H-4), 2.6 (*dd*, J = 16.5, 10 Hz, H-4), 1.7–1.5 (*m*, H-2, H-3), 1.08 (*d*, J = 6 Hz, Me), 0.9 (*d*, J = 6 Hz, Me). MS (*m/z*): 340 ($C_{21}H_{24}O_4$), 309, 283, 253 ($C_{16}H_{13}O_3$ base), 239, 223, 187, 151. CD: $[\theta]_{288}$ − 15 100; $[\theta]_{265}$ + 5920.

6,7-Dimethoxy-2α,3β-dimethyl-1β-(3′,4′-methylenedioxy-phenyl)-tetralin (**5**, 2%). ^{1}H NMR: δ6.8 (*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*, OMe), 3.4 (*d*, J = 9.8 Hz, H-1), 2.75 (*dd*, J = 16.5, 4.1 Hz, H-4), 2.6 (*dd*, J = 16.5, 10 Hz, H-4), 1.7 1.5 (*m*, H-2, H-3), 1.08 (*d*, J = 6 H-2, Me), 0.9 (*d*, J = 6 Hz, Me). MS (*m/z*): 340 ($C_{21}H_{24}O_4$), 309, 283, 253 ($C_{16}H_{13}O_3$ base), 239, 223, 187, 151. CD: $[\theta]_{288}$ − 11 600; $[\theta]_{267}$ + 4930.

7,8-Methylenedioxy-2α,3β-dimethyl-1α-(3′,4′-dimethoxyphenyl)-tetralin (**6**, 1.2%). ^{1}H NMR: δ6.72 (*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, –OCH_2O–), 3.9 (*d*, J = 3 Hz, H-1), 3.84, 3.82 (*s*, OMe), 2.77 (*dd*, J = 16.5, 12 Hz, H-4), 2.5 (*dd*, J = 16.5, 5.8 Hz, H-4), 1.7–1.5 (*m*, H-2, H-3), 1.0 (*d*, J = 6 Hz, Me), 0.9 (*d*, J = 6 Hz, Me). CD: $[\theta]_{285}$ + 12 250; $[\theta]_{262}$ + 3500. MS (*m/z*): 340 ($C_{21}H_{24}O_4$ base), 309, 283, 269, 253 ($C_{16}H_{13}O_3$), 239, 223, 202, 187, 151.

2,3-Dimethyl-1,4-bis-(3,4-dimethoxyphenyl)-butane (**7**, 0.2%). ^{1}H NMR: δ6.82–6.6 (*m*, ArH); 3.9, 3.85 (*s*, OMe), 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*, Me). MS (*m/z*): 358 ($C_{22}H_{30}O_4$), 269, 206, 179, 165, 151 (base).

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