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^{13}C NMR SPECTRAL ANALYSIS OF LIGNANS FROM *ARAUCARIA ANGUSTIFOLIA**

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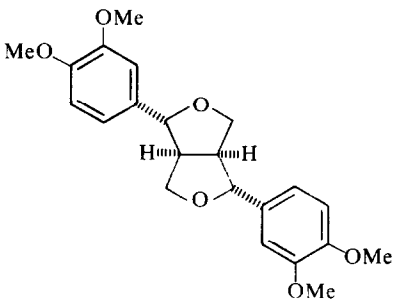
Key Word Index—*Araucaria angustifolia*; Araucariaceae; ^{13}C NMR; lignans; secoisolariciresinol; lariciresinol; isolariciresinol; isolariciresinol-4′-methyl ether; isolariciresinol-4-methyl ether; isolariciresinol dimethyl ether.

Abstract—Pinoresinol dimethyl ether, secoisolariciresinol, lariciresinol, isolariciresinol and isolariciresinol-4′-methyl ether were isolated from the knots of dead trees of *Araucaria angustifolia*. The ^{13}C NMR spectra of these compounds, their methyl and acetyl derivatives, and the corresponding one of matairesinol, have been recorded and the signals assigned. On the basis of these assignments, the structure of the new monomethyl ether of isolariciresinol has been established.

INTRODUCTION

In continuation with the work on the constituents of *Araucaria angustifolia* O. Ktze, [1] the components of the knot benzene extract were examined. By Si gel column chromatography the following lignans were isolated: pinoresinol diMe ether **1**, secoisolariciresinol **2a**, lariciresinol **4a**, isolariciresinol **6g**, and a monoMe ether of **6g**, not previously described in the literature, **6e**. The availability of representative members of the main classes of lignans [2] and the considerable interest focused on the ^{13}C NMR spectral studies of this and related families of natural products, [3–6] prompted us to undertake an analysis of these substances, as an aid in the structure elucidation of new compounds and also, as part of a project on the ^{13}C NMR spectroscopy of natural products [7].



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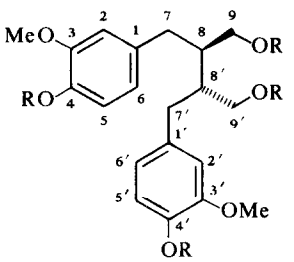
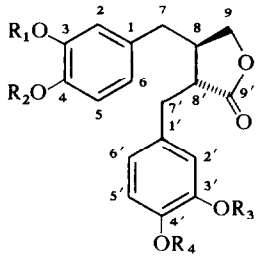
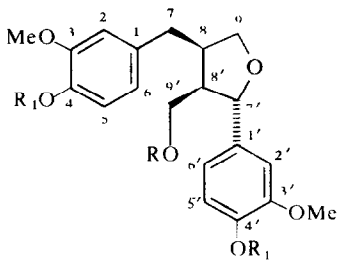


2a. R = H
2b: R = —C(=O)Me



3a: $R_1 = R_3 = Me$
3b: $R_1 = Me$, $R_2 = H$; $R_3 + R_4 = CH_2$
3c. $R_1 + R_2 = CH_2$; $R_3 + R_4 = CH_2$



4a: R = R_1 = H
4b: R = R_1 = —C(=O)Me
4c. R = H; R_1 = Me

* Dedicated to the memory of Professor Jayr de Paiva Campello.

Table 1. ^{13}C NMR data for secoisolariciresinol, matairesinol, lariciresinol and their derivatives*

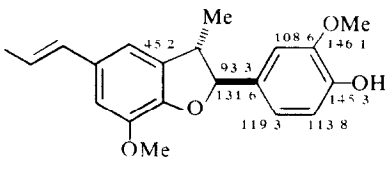
	2a‖	2b‡	3a	4a‖	4b§	4c‖
1	132.4	137.9	129.4	133.7	138.0	135.4
2	111.7	112.7	110.8	111.6	112.6	111.0†
3	146.6	150.8	146.4	146.9	150.9	148.2
4	143.7	138.4	144.2	143.9	138.7	147.3
5	114.3	122.4	113.9	114.6†	122.6†	111.9†
6	121.5	120.8	121.2	120.5	120.4	120.4
7	35.8	35.2	38.3	32.3	33.4	33.2
8	43.7	39.5	40.9	42.1	42.1	42.4
9	60.5	64.1	71.3	72.1	72.7	72.9
1′	132.4	137.9	129.5	131.6	138.7	132.9
2′	111.7	112.7	111.3	108.7	109.5	108.9
3′	146.6	150.8	146.5	146.9	150.9	148.9
4′	143.7	138.4	144.3	145.1	141.4	148.8
5′	114.3	122.4	114.3	114.4†	122.5†	111.3†
6′	121.5	120.8	121.9	118.1	117.6	117.9
7′	35.8	35.2	34.5	82.3	82.7	82.7
8′	43.7	39.5	46.5	52.2	49.0	52.5
9′	60.5	64.1	178.6	59.1	62.6	60.7
OMe	55.7	55.7	55.7	55.1	55.8	55.9

* The spectra were obtained at 25.2 MHz in the Fourier transform mode in $CDCl_3$ solns. The δ values are in ppm downfield from TMS. † Signals may be reversed. ‡ The acetyl C=O and Me shifts are 170.7; 168.8; 20.8 and 20.6 ppm, respectively. § The acetyl C=O and Me shifts are 170.7; 168.9 and 20.6 ppm, respectively. ‖ Some MeOH had been added for dissolution of the compound.

RESULTS AND DISCUSSION

Table 1 shows the δ values of compounds **2**–**4c**, since the shifts of compound **1**, has already been reported [3]. The spectra of secoisolariciresinol, **2a**, shows 10 signals which were assigned on the basis of chemical shift theory [8], comparison with shifts of related products [5] and analysis of the SFORD spectrum. The transformation of **2a** into its tetraacetyl derivative **2b**, causes the expected changes and confirms the above assignment.

The nonaromatic carbons of matairesinol **3a** [11] have their shift assigned by comparison with the recently reported δ values of like carbons of pluviatolide and hinokinin, **3b** and **3c**, respectively [5]. Although differences can be observed in the shifts of comparable sites of both benzene rings of **3a**, they are too small to be analyzed. In lariciresinol **4a**, however, the differences of the aromatic carbon shifts are larger and they were assigned by comparison with related products, compound **2a** for ring A and the neolignan licarin A, **5** [5] for ring B.



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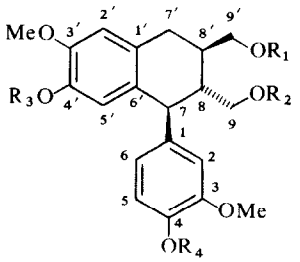
Carbon-1′, C-2′ and C-6′ are shielded, probably by steric compression, and C-4′ is deshielded, in relation to the same carbons of ring A. The nonaromatic carbons of **4a** were assigned by standard chemical shift theory, analysis of the SFORD spectrum and expected changes on acetylation, **4b**. Carbon-7 and C-9′ exhibit also the shielding effect observed on C-1′, C-2′ and C-6′, and resonate upfield in comparison with C-7 and C-9 of **2a**. The assignment of **4c** was based on the same arguments.

The nonaromatic carbons of isolariciresinol diMe ether **6a**, were split into two groups on the basis of their signal multiplicities from a SFORD spectrum, which shows triplets at 33.2, 62.6 and 66.2 ppm and doublets at 39.9, 48.0 and 48.2 ppm. The 33.2 ppm signal was assigned to C-7′ and the remaining ones to C-9′ and C-9, using *trans*-4-*t*-butylcyclohexylmethanol as reference [9]. The $\Delta\delta$

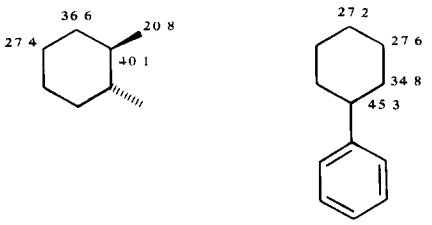


6a $R_1 = R_2 = H$; $R_3 = R_4 = Me$
6b: $R_1 = R_2 = -OC-Me$; $R_3 = R_4 = Me$
6c $R_1 = R_2 = R_3 = H$; $R_4 = Me$
6d: $R_1 = R_2 = R_3 = -OC-Me$; $R_4 = Me$
6e. $R_1 = R_2 = R_4 = H$; $R_3 = Me$
6f. $R_1 = R_2 = R_4 = -OC-Me$; $R_3 = Me$
6g $R_1 = R_2 = R_3 = R_4 = H$
6h $R_1 = R_2 = R_3 = R_4 = -OC-Me$

value of the carbinyl carbon shift of the model compound (68.9 ppm) and that of C-9′ (66.2 ppm), which is similar in magnitude to that observed in the methylcyclohexane to *trans*-dimethylcyclohexane change ($\Delta\delta$ = 2.5 ppm), can be explained by the reciprocal γ-effect of both carbinyl carbons. On C-9 this effect is enhanced by the vicinal aryl group.

The methines at 39.9, 48 and 48.2 ppm were distinguished by analysis of the corresponding diacetyl derivative, **6b**, carbon shifts, and comparison with the shifts of related carbons of *trans*-dimethylcyclohexane, **7**, and phenylcyclohexane, **8** [8]. As expected, acetylation induces deshielding of the carbinyl carbons [10] and shielding of their neighbours by magnitudes normal for saturated alcohols; the signal at 48 ppm, which is practically unaffected in the transformation can be therefore, assigned to C-7. The distinction of remaining methines is founded on the fact that C-8, which suffers an α-effect by C-9, and two β-effects by C-9′ and the dimethoxybenzene ring, should be deshielded in comparison to C-8′.

For the assignment of the aromatic carbons of **6a**, an exhaustive analysis of the spectra of the monomethyl ether **6c**, obtained by careful methylation of isolariciresinol, the monomethyl ether isolated from *A. angustifolia*, **6e**, and their corresponding di- and triacetyl derivatives **6b**, **6d** and **6f**, was undertaken. The aromatic methines were assigned by their multiplicities in the SFORD spectrum, standard chemical shift theory [8] and comparison of the shifts of the 6 compounds with each other. Comparison of the quaternary carbon shift data of **6a**, **6c** and **6d**, permits the assignment of the signals at 128.1 and 137.6 ppm of **6a**, to C-1′ and C-6′



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respectively, and the location of the OH group of **6c** at C-4′, on the basis of the known deshielding effect induced to the *para* and *ortho* positions and shielding effect to the ipso carbon on acetylation, [12] while the *meta* position is practically unaffected. The same type of analysis applied to compounds **6a**, **6e** and **6f**, shows the expected changes, deshielding effects on C-1, C-3 and C-5, and shielding on C-4 [13]. Carbon-6′ suffers also a strong shielding effect, probably due to the transmission of the substituent effect through the methylene group [14, 15]. The shifts of isolariciresinol **6g** and its tetraacetyl derivative are in good agreement with the above results, **6h** showing the sum of the effects observed on **6d** and **6f**.

The shifts of compounds **6a**–**6h**, which were used not only for the assignment of all carbons of isolariciresinol and its derivatives, but also to establish the structure of its new 4′-methyl ether, are listed in Table 2 [16].

Table 2. ^{13}C NMR data for isolariciresinol and its derivatives*

	6a¶	6b†	6c¶	6d‡	6e¶	6f§	6g¶	6h‖
1	131.7	131.0	132.8	131.7	131.9	138.4	132.6	138.4
2	112.8	112.5	112.5	111.9	112.6	113.1	112.0	113.1
3	148.9	148.9	149.1	149.1	146.4	150.9	145.2	151.0
4	146.9	147.1	145.8	147.8	144.1	143.4	143.5	142.7
5	110.8	111.0	111.5	111.1	114.3	122.7	114.5	122.7
6	121.7	121.6	122.1	121.7	121.6	121.5	121.9	121.5
7	48.0	47.3	47.7	47.0	47.0	47.6	47.4	47.2
8	48.2	43.7	48.0	43.4	47.0	43.8	47.5	43.5
9	62.6	63.4	62.4	63.1	61.3	63.4	62.1	63.0
1′	128.1	127.5	127.7	133.8	128.3	127.7	127.2	134.0
2′	110.7	110.7	111.0	111.7	110.6	110.8	110.6	111.7
3′	147.3	147.6	147.6	149.1	147.0	147.4	147.1	149.2
4′	147.0	147.1	144.0	137.8	146.5	147.3	144.1	137.9
5′	111.9	111.9	116.3	123.5	111.8	112.6	115.8	123.6
6′	137.6	136.6	138.4	135.9	136.5	130.4	136.8	131.0
7′	33.2	32.7	33.2	33.1	32.4	32.7	32.8	33.1
8′	39.9	35.4	39.9	35.3	38.9	35.5	39.5	35.2
9′	66.2	66.4	66.0	66.3	65.0	66.4	65.7	66.2
OMe	55.7	55.8	56.0	56.4	55.1	55.9	55.6	55.9

* The spectra were obtained at 25.2 MHz in the Fourier transform mode in $CDCl_3$ solns. The δ values are in ppm downfield from TMS. † The acetyl C=O and Me shifts are 170.8; 170.7 and 20.9 ppm, respectively. ‡ The acetyl C=O and Me shifts are 171.4; 171.2; 169.5; 21.4 and 21.1, respectively. § The acetyl C=O and Me shifts are 170.9; 170.7; 168.4 and 20.9 ppm, respectively. ‖ The acetyl C=O and Me shifts are 170.8; 170.6; 169.0; 168.8 and 20.8 ppm, respectively. ¶ Some MeOH had been added for the dissolution of the compound.

EXPERIMENTAL

Knots of *A. angustifolia* (from dead trees exposed to the weather for a long time) were collected at Araucaria (Estado do Paraná, Brasil). Powdered knots (2.7 kg) were continuously extracted with C_6H_6 for 24 hr. The C_6H_6 extract, was concd *in vacuo* and extracted with 3 × 1l. portions of hexane, the insoluble residue, chromatographed on a Si gel column and eluted with C_6H_6, C_6H_6–Et_2O, Et_2O, EtOAc and Me_2CO, yielding the following compounds in order of elution. pinoresinol diMe ether **1**, lariciresinol **4a**, secoisolariciresinol **2a**, isolariciresinol-4′-Me ether **6e** and isolariciresinol **6g**. The acetates and Me ethers were all prepared by standard methods, Ac_2O and C_5H_5N and ethereal CH_2N_2 respectively, except for compound **6e**, and the physical data were compared with those from the literature [17, 18].

Pinoresinol diMe ether, **1**. (3 g), mp 107–109° (from $CHCl_3$–MeOH); $[\alpha]_D^{25°}$ +46° (*c* 1.0, $CHCl_3$). Identical with authentic sample.

Lariciresinol, **4a**. (0.55 g), mp 162–164° (from $CHCl_3$–MeOH); $[\alpha]_D^{25°}$ +18° (*c* 1.0, Me_2CO), M^+ 360. Identical with authentic sample. *Lariciresinol triacetate*, **4b**, viscous oil, $[\alpha]_D^{25°}$ +8° (*c* 1.0, $CHCl_3$), M^+ 486. *Lariciresinol diMe ether*, **4c**, mp 78–80°, $[\alpha]_D^{25°}$ +12° (*c* 1.0, Me_2CO), M^+ 388.

Secoisolariciresinol, **2a**. (0.6 g), mp 112–114° (from C_6H_6–Me_2CO), $[\alpha]_D^{25°}$ −32° (*c* 1.0, Me_2CO), M^+ 362. *Secoisolariciresinol tetraacetate* **4b**, viscous oil, $[\alpha]_D^{25°}$ −8° (*c* 1.0 $CHCl_3$), M^+ 530.

Isolariciresinol-4′-Me ether, **6e**. (0.11 g), mp 188–190° (from MeOH–Me_2CO), $[\alpha]_D^{25°}$ +16° (*c* 1.0 MeOH), MS (high resolution), found 374.1643, calc. for $C_{21}H_{26}O_6$, 374.1729, M^+ *m/e* 374 (100), 325 (70), 255 (36), 201 (20), 194 (18), 189 (48), 151 (20), 137 (65). PMR ($CDCl_3$): δ 2.65–2.9 (2H, *m*), 3.6; 3.85; 3.9 (each 3H, *s*); 3.75 (*m*); 6.2 (1H, *s*); 6.6–6.85 (4H, *m*). *Isolariciresinol-4′-Me ether triacetate*, **6f**, viscous oil, $[\alpha]_D^{25°}$ +3° (*c* 1.0 $CHCl_3$), M^+ 500; PMR ($CDCl_3$): δ 2.1 (6H, *s*), 2.3 (3H, *s*), 2.75–2.95 (2H, *m*), 3.6, 3.75, 3.85 (each 3H, *s*), 4–4.25 (*m*), 6.6–6.8 (4H, *m*), 7 (1H, *d*, *J* = 8 Hz).

Isolariciresinol, **6g**. (0.75 g), mp 155–157° (from $CHCl_3$–MeOH), $[\alpha]_D^{25°}$ +68° (*c* 1.0 Me_2CO), M^+ 360. *Isolariciresinol tetraacetate*, **6h**, mp 163–164°, $[\alpha]_D^{25°}$ −3.5° (*c* 1.0 $CHCl_3$), M^+ 528. *Isolariciresinol diMe ether*, **6a**, mp 175–177°, $[\alpha]_D^{25°}$ +12° (*c* 1.0 $CHCl_3$), M^+ 388. *Isolariciresinol diMe ether diacetate* **6b**, viscous oil, $[\alpha]_D^{25°}$ −4° (*c* 1.0 $CHCl_3$), M^+ 472.

Isolariciresinol-4-Me ether, **6c**. Isolariciresinol, **6g**, (0.35 g) in MeOH was treated with CH_2N_2–Et_2O, at 10°. When the dimethylated product was detected on TLC, excess reagent was evapd and the residue chromatographed on a Si gel column. Elution with $CHCl_3$ containing 1% MeOH yielded the following compounds: **6a** (70 mg), **6c** (75 mg), a mixture of **6c** and **6e** (85 mg) and **6g** (60 mg). *Isolariciresinol-4-Me ether* showed mp 142–143°, $[\alpha]_D^{25°}$ +43° (*c* 1.0 MeOH), M^+ *m/e* 374 (80), 325 (100), 298 (22), 255 (40), 201 (18), 194 (32), 189 (43), 187 (52), 175 (70), 151 (65), 137 (27). PMR ($CDCl_3$): δ 2.65–2.9 (2H, *m*); 3.75 (*m*); 3.8 (3H, *s*), 2.9 (6H, *s*), 6.3 (1H, *s*), 6.6–6.85 (4H, *m*). The diMe ethers prepared from **6c** and **6e** were identical to each other and identical in all respects to the diMe ether, **6a** prepared from isolariciresinol. *Isolariciresinol-4-Me ether triacetate*, **6d**, mp 110–112° $[\alpha]_D^{25°}$ −3 (*c* 1.0 $CHCl_3$), M^+ 500, PMR ($CDCl_3$). δ 2.15 (6H, *s*), 2.2 (3H, *s*), 2.75–3 (2H, *m*), 3.8 (6H, *s*), 3.9 (3H, *s*), 3.95–4.3 (*m*), 6.4, 6.6 (each 1H, *s*), 6.7–6.9 (3H, *m*).

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REFERENCES

1. de Paiva Campello, J. and Ferreira Fonseca, S. (1975) *Phytochemistry* **14**, 2299.
2. The nomenclature and the numbering used in this paper are the ones reported by Weinges, K. and Spänig, R. (1967) in

Oxidative Coupling of Phenols (Taylor, W. I. and Battersby, A. R. eds). Marcel Dekker, New York; and Klyne, W. and Buckingham, J (1974) *Atlas of Stereochemistry, Absolute Configurations of Organic Molecules* p. 185. Chapman & Hall London
3. Pelter, A., Ward, R. S., Venkata Rao, E. and Sastry, K V. (1976) *Tetrahedron* **32**, 2873
4. Anjaneyulu, A. S. R., Madhusudhana Rao, A., Kameswara Rao, V., Ramachandra Row, L., Pelter, A. and Ward, R. S. (1977) *Tetrahedron* **33**, 133.
5. Wenkert, E., Gottlieb, H. E., Gottlieb, O. R., Pereira, M. O. da S. and Formiga, M. D. (1976) *Phytochemistry* **15**, 1574, and references cited therein.
6 Kelley, C. J., Harruff, R C. and Carmack, M. (1976) *J. Org. Chem.* **41**, 449; and Castelão Jr J. F., Gottlieb, O. R., de Lima, R. A., Mesquita, A. A. L., Gottlieb, H. E. and Wenkert, E. (1977) *Phytochemistry* **16**, 735.
7 Imamura, P. M., Marsaioli, A. J., Barata, L. E S. and Ruveda, E. A. (1977) *Phytochemistry* **16**, 1842.
8. Stothers, J. B (1972) *Carbon-13 NMR Spectroscopy* Academic Press, New York; Levy, G. C. and Nelson, G. L. (1972) *Carbon-13 Nuclear Magnetic Resonance for Organic Chemists.* Wiley-Interscience, New York.
9. Buchanan, G. W., Stothers, J. B. and Wu, S. T. (1969) *Can. J. Chem.* **47**, 3113.
10. The deshielding effects observed on C-9′ and C-9 by acetylation, although small (C-9′, $\Delta\delta$ = 66.4–66.2 = 0.2 ppm, C-9, $\Delta\delta$ = 63.4–6.26 = 0.8 ppm) are in agreement with the reported values for *trans-4-t*-butylcyclohexylmethanol acetate and *trans*-4-*t*-butylcyclohexylmethanol (carbinyl carbon $\Delta\delta$ = 69.3–68.9 = 0.4 ppm) [9].
11. We thank Professor H. Erdtman (Royal Institute of Technology, Stockholm) and Professor R. C Cambie (University of Auckland) for generous gift of lariciresinol and matairesinol respectively.
12. Pelter, A., Ward, R. S. and Gray, T. I. (1976) *J. Chem. Soc. Perkin I* 2475.
13. Acetylation of **6c** to **6d**, the same as **6e** to **6f**, shields C-4′ ($\Delta\delta$ = 6.2 ppm) and C-4 ($\Delta\delta$ = 0.7 ppm), but this effect is lower in the latter. Differences in shielding effects on acetylation were also observed in the transformation of **4a** to **4b**, ($\Delta\delta$ for C-4 = 5.2 ppm. $\Delta\delta$ for C-4′ = 3.7 ppm).
14. Shapiro, M. J. (1977) *Tetrahedron* **33**, 1091.
15. Carbon-1′ of *p*-benzylphenol suffers a shielding effect ($\Delta\delta$ = 0.7 ppm) on acetylation.
16. The PMR spectrum of **6f** is in agreement with the proposed structure, since it shows a clear doublet at δ 7 (J = 8 Hz), produced by the deshielding effect of acetylation on the *ortho* positions, in comparison with the corresponding ones of **6e**.
17. Erdtman, H. and Tsuno, K. (1969) *Acta Chem. Scand.* **23**, 2021.
18. Popoff, T. and Theander, O. (1977) *Acta Chem. Scand.* **B31**, 329.