

A general CD-method for the configurational assignment of *erythro*-8.4'-oxyneolign-8'-enes

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Dedicated to Professor Karsten Krohn on the occasion of his 60th birthday
(received 11 May 04; accepted 23 Nov 04; published on the web 25 Nov 04)

Abstract

Resolution of *erythro*-7-hydroxy-3',5'-dimethoxy-3,4-methylenedioxy-8.4'-oxyneolign-8'-ene **1c** was achieved by preparing the diastereomeric esters with (–)-(R)- α -methoxyphenylacetic acid. The correlation of its CD properties with the absolute configuration established a general CD method for the configurational assignment of *erythro*-8.4'-oxyneolignenes: positive Cotton effects of the two low-energy CD transitions derive from 7*R*,8*S* configuration while the negative ones from 7*S*,8*R*. This rule allows the configurational assignment of *erythro*-8.4'-oxyneolignenes with different aromatic substitution pattern by a CD measurement.

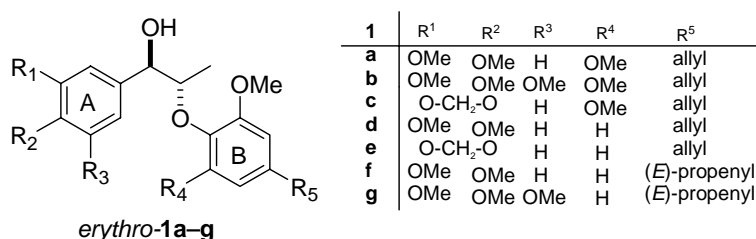
Keywords: 8.4'-Oxyneolign-8'-enes, absolute configuration, circular dichroism, Mosher's method

Introduction

Within the great structural variety of neolignans, the 8.O.4'-type represents a small subgroup whose members were isolated exclusively from the plants of *Myristicaceae* species and studied for a wide range of biological activities such as antifungal,^{1,2} antileukemic,³ anti-leishmanial,⁴ and antioxidant activity.^{5,6} While the *erythro* and *threo*-isomers of 8.4'-oxyneolign-8'-enes were found to exhibit significantly different activities in pharmacological assays,^{1,5,6} the differences in the activities of the enantiomers of the *erythro* or *threo* series have not been studied yet.

The absolute configurations of *erythro*-8.4'-oxyneolign-8'-enes were determined by Horeau's^{7,8} and Mosher's^{8,9} methods or on the basis of the transition state of the asymmetric

reduction if an enantioselective synthesis was achieved.^{7,8} Although the optical properties of these *erythro*-neolignenes were characterized by their optical rotation, these data can not be used for direct comparison if the substitution patterns of the aromatic rings are different or a glycoside residue is present. Thus, if the absolute configurations of *erythro*-8.4'-oxyneolign-8'-enes of natural origin are to be determined, a derivatization by Mosher's or Horeau's methods needs to be carried out in each case. This drawback can be the main reason that the absolute configurations of several recently isolated optically active *erythro*-8.4'-oxyneolign-8'-enes have not been determined.¹⁰⁻¹² For the configurational assignment of these compounds a circular dichroism (CD) spectroscopic method is required which can be generally applied regardless of the substitution pattern of the aromatic rings and the presence of a sugar moiety or acetate group. Recently, effective enantiomeric separations of *rac-erythro*-8.4'-oxyneolign-8'-enes **1a-g** with different aromatic substitution pattern were achieved on a Chiralcel OD column which allowed the LC/CD analysis of the enantiomers.¹³ These LC/CD data revealed that the different substitution patterns of the rings A and B induce only wavelength shifts in the CD spectra of the first eluted enantiomers while the two low-energy CD transitions were always negative. On the basis of Arnoldi and Merlini's CD results,¹⁴ the *7S,8R* absolute configuration was proposed for the first eluted enantiomers. This assignment suggested the revision of configuration of formerly isolated *erythro*-8.4'-oxyneolign-8'-enes¹⁵ and allowed the determination of configuration in others with CD data reported.^{16,17} The configurational assignments of the *erythro*-8.4'-oxyneolign-8'-enes **1a-g** rely on the comparison with Arnoldi and Merlini's CD results.¹⁴ In order to confirm our assignment independently and to establish a general CD method, the natural product *rac-1c* was chosen for resolution to determine the configuration of its enantiomers by Mosher's method. Then the comparison of the CD spectrum measured for the optically active **1c** with the on-line LC/CD spectra of **1a-g** would enable their independent configurational assignments, and the optical rotation of **1c** can be correlated with its CD data as well.

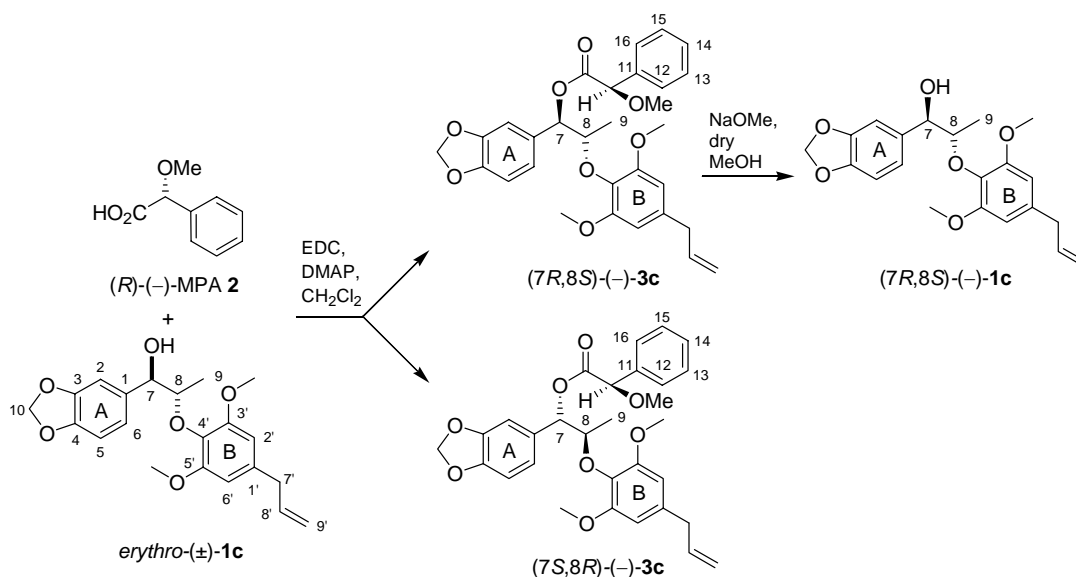


Results and Discussion

The *erythro*-8.4'-oxyneolign-8'-ene **1c**, isolated from the seeds of *Myristica fragrans* Houtt. (nutmeg) by Forrest *et al.*,¹⁸ was obtained in racemic form according to a known procedure.⁶ Racemic (\pm)-**1c** was converted with (*R*)-(-)- α -methoxyphenylacetic acid [(*R*)-(-)-MPA] into the diastereomeric esters (*7R,8S*)-(-)-**3c** and (*7S,8R*)-(-)-**3c** which could be separated by flash chromatography (Scheme 1). While the Mosher method¹⁹ is usually applied in a manner that the

(*R*)- and (*S*)-MPA esters of a given secondary alcohol are prepared for the determination of its configuration, in this case the (*R*)-MPA esters of two enantiomeric neolignenes were prepared to gain the same information.

In the low-energy conformer¹⁹ of (*7R,8S*)-(-)-**3c** the side with ring B is shielded by the phenyl group of (*R*)-MPA, while in (*7S,8R*)-(-)-**3c** ring A is shielded (Figure 1).



Scheme 1. Resolution of (±)-**1c** with (*R*)-(-)-MPA: methoxy(phenyl)acetic acid. EDC: *N*-[3-(dimethylamino)propyl]-*N'*-ethylcarbodiimide hydrochloride, DMAP: 4-dimethylaminopyridine.

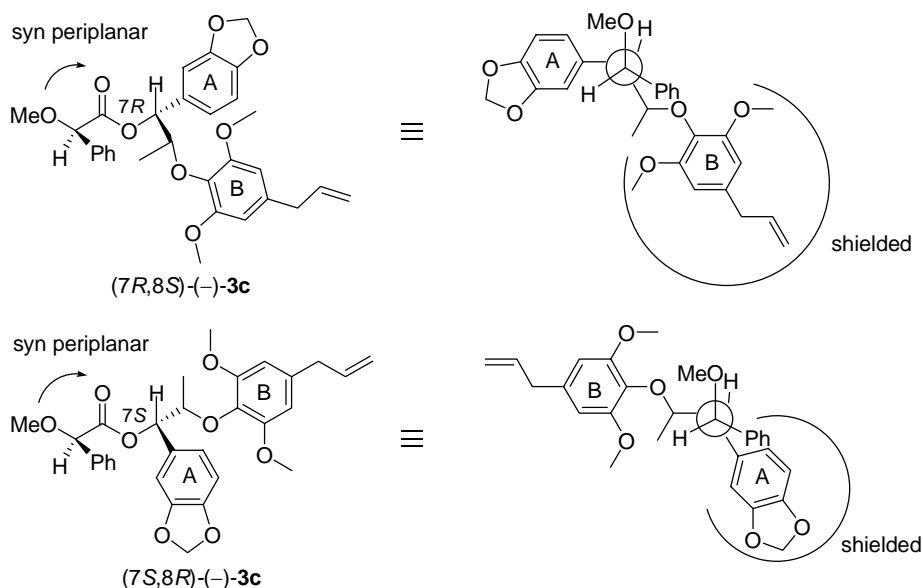


Figure 1. Low-energy conformers of esters (*7R,8S*)-(-)-**3c** and (*7S,8R*)-(-)-**3c** indicating the shielding effect of the MPA phenyl ring.

As a result, the subtraction of corresponding proton and carbon signals of (7*S*,8*R*)-(-)-**3c** from those of (7*R*,8*S*)-(-)-**3c** resulted in negative $\Delta\delta$ values for side B and positive ones for side A, which allowed the configurational assignment of (7*R*,8*S*)-(-)-**3c** and (7*S*,8*R*)-(-)-**3c** (Table 1). The proton signals of H-8 and the carbon signals of C-9, C-2' and C-6' showed discrepancies, which are likely due to the effect of the methoxy oxygens or experimental error of the NMR measurement. The ester (7*R*,8*S*)-(-)-**3c** was hydrolyzed with 1N NaOMe in anhydrous methanol to give (7*R*,8*S*)-(-)-**1c**; its CD spectrum has two positive maxima above 230 nm in acetonitrile [246 (2.22), 280 (0.46)]. These CD bands have opposite signs and much lower intensities compared to those obtained from the LC/CD measurement of the first eluted enantiomer of **1c** [n-hexane/2-propanol 9:1; 246 (-8.35), 281 (-1.35)];¹³ its absolute configuration 7*S*,8*R* was proposed on the basis of Arnoldi and Merlini's CD results.¹⁴

Table 1. Selected ¹H and ¹³C NMR (in *italics*) data of (7*R*,8*S*)-(-)-**3c**, (7*S*,8*R*)-(-)-**3c**, and chemical shift differences. Side A refers to ring A and side B refers to side chain atoms 8, 9 with ring B, both sides flanking the stereogenic center C-7

	Side A				Side B						
	2	5	6	10	OMe	8	9	7'	9'	2'	6'
(7 <i>R</i> ,8 <i>S</i>)-	6,81	6,70	6,68	5,90	3,67	4,30	1,12	3,29	5,05	6,33	6,33
(-)- 3c	<i>107,58</i>	<i>107,78</i>	<i>120,62</i>	<i>100,87</i>	<i>55,85</i>	<i>80,02</i>	<i>14,68</i>	<i>40,44</i>	<i>115,82</i>	<i>105,60</i>	<i>105,60</i>
(7 <i>S</i> ,8 <i>R</i>)-	6,16	6,47	6,15	5,38	3,79	4,25	1,21	3,33	5,08	6,40	6,40
(-)- 3c	<i>106,64</i>	<i>107,70</i>	<i>119,35</i>	<i>100,82</i>	<i>55,98</i>	<i>80,61</i>	<i>13,80</i>	<i>40,50</i>	<i>115,94</i>	<i>105,52</i>	<i>105,52</i>
$\Delta\delta$	0,65	0,23	0,53	0,52	-0,12	<u>0,05</u>	-0,09	-0,04	-0,03	-0,07	-0,07
	<i>0,94</i>	<i>0,09</i>	<i>1,27</i>	<i>0,05</i>	<i>-0,12</i>	<i>-0,59</i>	<i>0,88</i>	<i>-0,06</i>	<i>-0,12</i>	<i>0,08</i>	<i>0,08</i>

The opposite CD signs unambiguously confirm the previously proposed configurational assignments of the *erythro*-8.4'-oxyneolign-8'-enes **1a-g** based on Arnoldi and Merlini's CD result¹⁴ as correct, and hence they can be used as reference. Moreover, the negative specific rotation of (7*R*,8*S*)-**1c**, [α]_D²⁰ -16.1, (CHCl₃; c = 0.26), is in agreement with Zacchino and Badano's result,⁸ who have reported the 7*R*,8*S* configuration for (-)-**1c**, [α]_D²⁰ -27, ee = 80% (CHCl₃; c = 1), but did not disclose any CD data. Both the lower CD intensities and the lower specific rotation of (-)-(7*R*,8*S*)-**1c** obtained by hydrolysis suggest that racemization may have occurred in the course of the hydrolysis of ester (7*R*,8*S*)-(-)-**3c**. Thus the product of the hydrolysis was studied by HPLC (Chiralcel OD, *n*-hexane/2-propanol 9:1) which proved that (7*R*,8*S*)-(-)-**1c** was partially racemized to (7*S*,8*R*)-(+)-**1c** during the hydrolysis which reduced the enantiomeric excess to 50% according to HPLC evidence. The inversion must have taken place on both stereogenic centers because the presence of *threo*-8.4'-oxyneolign-8'-ene was not detected.

The correlation of the absolute configuration of (7*R*,8*S*)-(-)-**1c** with its positive CD transitions at 246 and 280 nm unambiguously proved that the first eluted isomers of the *erythro*-8.4'-oxyneolign-8'-enes **1a-g** (Chiralcel OD column), showing negative Cotton effects (CEs) for

the two low-energy CD transitions (second and third columns in Table 2), have the *7S,8R* absolute configuration. In contrast to the optical rotation, the signs of these characteristic CD transitions are not sensitive to the substitution pattern of the aromatic rings which is exemplified by the LC/CD data of **1a–g** showing only wavelength shifts depending on the nature of the substituents.

Thus the absolute configurations of the isolated *erythro*-8.4'-oxyneolign-8'-enes can be deduced from a CD measurement; positive CEs of the two low-energy CD transitions derive from *7R,8S* configuration while the negative ones from *7S,8R*.

Table 2. CD data of first eluted *erythro*-(*7S,8R*)- 8.4'-oxyneolign-8'-enes **1a–g** obtained by LC/CD measurement (Chiralcel OD column)

Compound	CD Data in <i>n</i> -hexane/propan-2-ol (9:1) [nm ($\Delta\epsilon$)]		
1a	280 (–1.08)sh, 275 (–1.13), 265 (–0.88)	244 (–10.43)	229 (1.95)
1b	278 (–1.27), 270 (–1.30)	243 (–10.58)	230 (2.21)
1c	296 (–0.59)sh, 290 (–0.87)sh, 281 (–1.35)	246 (–8.35)	217 (–1.55)
1d	278 (–1.78)	239 (–9.52)	226 (0.39), 216 (–1.63)sh, 209 (–7.19)
1e	294 (–0.60)sh, 283 (–1.71)	239 (–6.79)	224 (–1.95)sh
1f	310 (–1.07)sh, 299 (2.15)sh, 289 (–2.38)	255 (–6.03)	234 (6.20)
1g	312 (–0.67)sh, 294 (–1.78), 277 (–1.93)sh	263 (5.00)sh	235 (5.21) 256 (–6.35)

Experimental Section

General Procedures. Column chromatography was performed on silica gel (Merck 60, 70–230 mesh). The thin layer chromatography was performed on aluminum backed TLC plates of silica gel 60 F254 (Merck, 0.2 mm) with the indicated eluent. The HPLC was performed on Chiralcel OD column (250x4.6 mm, 10 μ m, analytical column, Daicel Chemical Industries, LTD.) with Jasco type HPLC system: Jasco PU-980 HPLC Pump, Jasco MD-910 Multiwavelength detector. NMR spectra were recorded on a Bruker AM 360 (360.13 MHz for ^1H , 90.03 MHz for ^{13}C) spectrometer. Chemical shifts (δ) are given from internal CHCl_3 (7.26) for ^1H NMR, $^{13}\text{CDCl}_3$ (77.0). Coupling constants (J in Hz) are accurate to ± 0.2 Hz for ^1H . CD spectra were obtained with a Jasco-810 spectropolarimeter in Sharlau Spectosolv grade solvents at room temperature. (\pm)-*erythro*-7-Hydroxy-3',5'-dimethoxy-3,4-methylenedioxy-8.4'-oxyneolign-8'-ene [(\pm)-**1c**] was prepared according to the literature.⁶

(R)-MPA Esters of (*7R,8S*)-(–)-*erythro*-7-hydroxy-3',5'-dimethoxy-3,4-methylenedioxy-8.4'-oxyneolign-8'-ene [(*7R,8S*)-(–)-3c**] and (*7S,8R*)-(–)-*erythro*-7-hydroxy-3',5'-dimethoxy-3,4-methylenedioxy-8.4'-oxyneolign-8'-ene [(*7S,8R*)-(–)-**3c**].** To a magnetically stirred solution of

R-(–)-MPA (64 mg, 0.39 mmol), *N*-[3-(dimethylamino)propyl]-*N'*-ethylcarbodiimide hydrochloride (EDC) (74 mg, 0.39 mmol), and 4-dimethylaminopyridine (DMAP) (8 mg, 0.07 mmol) in anhydrous dichloromethane (5 mL), a solution of (±)-**1c** (49 mg, 0.13 mmol) in anhydrous dichloromethane (2 mL) was slowly added. The reaction mixture was stirred at room temperature for 24 h, and was then evaporated to dryness under reduced pressure. The residue was purified by flash chromatography (hexane/EtOAc 6:1) to afford (7*R*,8*S*)-(–)-**3c** (30 mg, 45%) as an oil and (7*S*,8*R*)-(–)-**3c** (25 mg, 37%) as white crystals, mp 107–108 °C.

(*R*)-MPA Ester of (7*R*,8*S*)-(–)-3c**.** $[\alpha]_{\text{D}}^{20}$ –7.5 (CHCl₃; *c* = 0.60), CD (CH₃CN, 0.20 mM), λ ($\Delta\epsilon$): 282 (0.27), 246 (1.26), 226 (–2.81) nm. ¹H NMR (360.13 MHz, CDCl₃): δ 7.49 (2H, d, *J* = 7.2 Hz, H-12, H-16), 7.33 (3H, m, *J* = 7.2 Hz, H-13, H-14, H-15), 6.81 (1H, s, H-2), 6.70 (1H, d, *J* = 3.6 Hz, H-5), 6.68 (1H, d, *J* = 3.6 Hz, H-6), 6.33 (2H, s, H-2', H-6'), 5.97 (1H, m, H-8'), 5.90 (2H, s, H-10), 5.08 (2H, m, *J* = 9.7 Hz, H-9'), 4.84 (1H, s, α -H_(MPA)), 4.29 (1H, m, *J* = 4.6 Hz H-8), 3.67 (3H, s, OMe), 3.48 (3H, s, OMe), 3.31 (1H?, d, *J* = 6.5 Hz, H-7'), 1.14 (3H, d, *J* = 6.1 Hz, H-9). ¹³C NMR (90.03 MHz, CDCl₃): δ 169.7 (C=O), 153.3 (C-3', C-5'), 147.4 (C-3), 147.0 (C-4), 137.2 (C-8'), 136.3 (C-4'), 135.6 (C-1), 134.0 (C-1'), 131.6 (C-11), 128.4 (C-12, C-16), 127.1 (C-13, C-14, C-15), 120.6 (C-6), 115.8 (C-9'), 107.8 (C-5), 107.6 (C-2), 105.6 (C-2', C-6'), 100.9 (C-10), 82.9 (CH_{MPA}), 80.0 (C-8), 77.8 (C-7), 57.5 (OMe_{MPA}), 55.9 (OMe), 40.4 (C-7'), 14.7 (C-9).

(*R*)-MPA Ester of (7*S*,8*R*)-(–)-3c**.** $[\alpha]_{\text{D}}^{20}$ –67.40 (CHCl₃; *c* = 0.50), CD (CH₃CN, 0.35 mM), λ ($\Delta\epsilon$): 281 (–0.51), 246 (–2.36), 225 (–7.19) nm. ¹H NMR (360.13 MHz, CDCl₃): δ 7.46 (2H, d, *J* = 7.2 Hz, H-12, H-16), 7.39 (3H, *J* = 7.2 Hz, H-13, H-14, H-15), 6.48 (1H, d, *J* = 3.6 Hz, H-5), 6.40 (2H, s, H-2', H-6), 6.16 (1H, s, H-2), 6.14 (1H, d, *J* = 3.6 Hz, H-6), 5.98 (1H, m, H-8'), 5.83 (2H, s, H-10), 5.08 (2H, m, *J* = 9.7 Hz, H-9'), 5.01 (1H, s, α -H_(MPA)), 4.25 (1H, m, *J* = 4.6 Hz, H-8), 3.79 (3H, s, OMe), 3.41 (3H, s, OMe), 3.34 (2H, d, *J* = 6.5 Hz, H-7'), 1.22 (3H, d, *J* = 6.1 Hz, H-9). ¹³C NMR (90.03 MHz, CDCl₃): δ 169.2 (C=O), 153.3 (C-3', C-5'), 146.7 (C-3, C-4), 137.2 (C-8'), 136.4 (C-4'), 136.0 (C-1), 133.7 (C-1'), 131.3 (C-11), 128.7 (C-12, C-16), 128.6 (C-13, C-14, C-15), 119.4 (C-6), 115.9 (C-9'), 107.7 (C-5), 106.6 (C-2), 105.5 (C-2', C-6'), 100.8 (C-10), 82.2 (CH-MPA), 80.6 (C-8), 76.7 (C-7), 56.9 (OMe-MPA), 55.9 (OMe), 40.5 (C-7'), 13.8 (C-9).

(7*R*,8*S*)-(–)-7-Hydroxy-3',5'-dimethoxy-3,4-methylenedioxy-8,4'-oxyneolign-8'-ene [(7*R*,8*S*)-(–)-1c**].** To a magnetically stirred solution of (7*R*,8*S*)-(–)-**3c** (30 mg, 0.06 mmol) in anhydrous MeOH (5 mL) was added 1*N* NaOMe solution (5 drops) at room temperature. After 24 h, the reaction mixture was evaporated to dryness under reduced pressure, and the residue was purified by preparative TLC (hexane/EtOAc 4:1) to give (7*R*,8*S*)-(–)-**1c** as a colorless oil (13 mg, 59%). $[\alpha]_{\text{D}}^{20}$ = –16.1 (CHCl₃; *c* 0.26). CD (CH₃CN, 0.35 mM): λ ($\Delta\epsilon$) 246 (2.22), 280 (0.46), 292sh (0.25), 296sh (0.18) nm. ¹H NMR (CDCl₃): δ 6.86 (1H, s, H-2), 6.74 (2H, s, H-5, H-6), 6.45 (2H, s, H-2', H-6'), 6.02 (1H, m, H-8'), 5.92 (2H, s, H-10), 5.08 (2H, m, *J* = 9.7 Hz, H-9), 4.76 (1H, s, H-7), 4.31 (1H, m, *J* = 4.6 Hz, H-8), 3.86 (6H, s, 2xOMe), 3.35 (2H, d, *J* = 6.7 Hz, H-7'), 1.13 (3H, d, *J* = 4.1 Hz, H-9).

Acknowledgements

The authors thank the National Science Foundation (OTKA T-034250, F-043536 and F-047025) for financial support. Tibor Kurtán is indebted to the Hungarian Academy of Sciences for the Bolyai Postdoctoral Fellowship and László Juhász for the Békésy György Postdoctoral Fellowship.

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