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Diterpenoids from Cascarilla (*Croton eluteria* Bennet)

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Cascarilla is a commercially available and cheap source of polyfunctionalized diterpenoids belonging to the clerodane structural type. In addition to the bitter triol cascarillin, 10 additional new diterpenoids (eluterins A−J) have been isolated and characterized by spectroscopic means. Structural diversity within cascarilla clerodanes involves mainly the linkage between the carbocyclic and the heterocyclic moieties and the functionalization of C-3, C-4, and C-6 of the decalin core. Cascarillin was shown to be a mixture of interconverting γ-lactols and not a γ-hydroxyaldehyde as previously reported.

KEYWORDS: Cascarilla; *Croton eluteria*; eluterins; bitter compounds; diterpenes; clerodane; NMR spectroscopy

INTRODUCTION

Cascarilla, the bitter bark of the South American tree *Croton eluteria* Bennet (Euphorbiaceae), has long been used in medicine as a replacement for Cinchona bark and *Cascara sagrada* (*1*), but it is nowadays better known as a tobacco additive and as a flavoring agent for liqueurs (*2*). Cascarilla holds a venerable position in natural product chemistry as the source of the clerodane cascarillin (**1a**, **b**, **Figure 1**), the first diterpenoid obtained in pure form and one of the first nonalkaloidal bitter substances isolated from plants. The isolation of cascarillin was reported in 1845 (*3*), but its structure was not elucidated until 1966, when a derivative (7-deacetyl-3-iodoacetate) finally yielded to X-ray analysis (*4*). A related clerodane, named cascarillin A, was also obtained in these studies (*5*), and a recent report disclosed the presence of three additional analogues (*6*). Our interest in this drug was spurred by its current relevance as a flavoring agent, and by the discovery that clerodanes from certain *Croton* species show powerful anti-ulcer and gastroprotective activity (*7*). Substantiation of these properties also for the constituents from cascarilla, could provide a rationale for its use in bitter preparations aimed at improving digestion.

MATERIALS AND METHODS

General Experimental Procedures. Optical rotations were determined on a Perkin-Elmer 192 polarimeter equipped with a sodium lamp (589 nm) and 10-cm microcell. IR (KBr) spectra were obtained on a Perkin-Elmer 681 spectrophotometer. Mass spectra were recorded in the electrospray (ES) and EI (70 eV, direct inlet) modes on a Prospec Fisons mass spectrometer. ^{1}H and ^{13}C NMR spectra were recorded at 500 and 125 MHz, respectively, on a Bruker AMX-500 spectrometer. Chemical shifts were referenced to the residual solvent signal ($CDCl_3$: δ_H 7.26, δ_C 77.0). The multiplicities of ^{13}C resonances were determined by DEPT experiments. One-bond heteronuclear ^{1}H-^{13}C connectivities were determined with 2D HMQC experiments, with a BIRD pulse 0.5 s before each scan to suppress the signal from protons not directly bonded to ^{13}C. The interpulse delays were adjusted for an average $^1J_{CH}$ of 125 Hz. Two and three bond heteronuclear ^{1}H-^{13}C connectivities were determined with 2D HMBC experiments, optimized for $^{2-3}J_{CH}$ of 7 Hz. Nuclear Overhauser effect (nOe) measurements were performed by 2D ROESY experiments. MPLC was performed on a Büchi 861 apparatus using columns packed with silica gel (230−400 mesh) or with Sephadex LH-20. HPLC in isocratic mode was performed on a Beckmann apparatus equipped with a refractive index detector and using 250 × 4 mm or 250 × 10 mm LUNA SI60 columns.

Plant Material. Cascarilla, *Croton eluteria* Bennet, was purchased from Minardi, Bagnacavallo (RA). A voucher specimen (FRB103-4) is held at DISCAFF.

Extraction and Isolation. Powdered cascarilla (508 g) was exhaustively extracted by percolation with acetone at room temp (4 × 2 L). Evaporation of the pooled extracts left a brown gum (76 g). Part of this (38 g) was fractionated by column chromatography (300 g silica gel) with a petroleum ether/EtOAc gradient (from 9:1 to 1:9) to give four main fractions (A−D). Fraction A (petroleum ether/EtOAc, 9:1) was crystallized from petroleum ether to afford lupeol (0.25 g, 0.10%), and trituration of fraction D (petroleum ether/EtOAc, 4:6) with ether/acetone afforded cascarillin (1.55 g, 0.62%).

Fractions B (3.1 g) and C (2.9 g) (petroleum ether/EtOAc, 7:3 and 5:5, respectively) were complex mixtures, and were poorly resolved by column chromatography on silica gel with a variety of eluant mixtures (petroleum ether/EtOAc, chloroform/acetone, chloroform/ethanol). A better fractionation was achieved by chromatography on Sephadex LH-20 ($MeOH/CH_2Cl_2$, 1:1), which afforded six sub-

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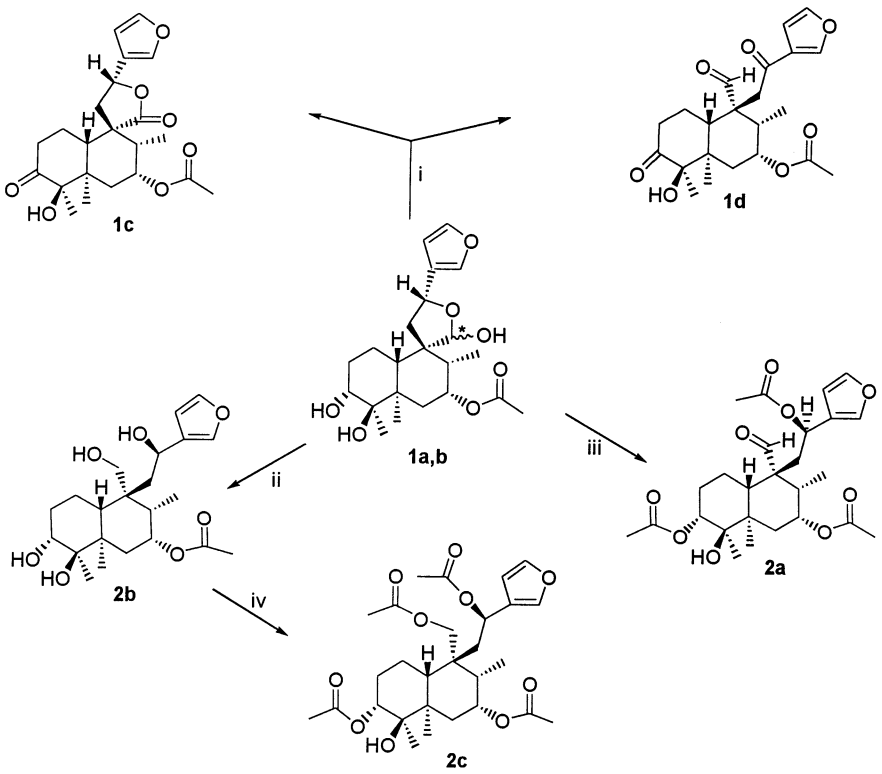

Figure 1. Chemical conversions of cascarillin: (i) PDC/Celite in CH_2Cl_2; (ii) $NaBH_4$ in methanol; (iii) Ac_2O in dry pyridine; (iv) Ac_2O/DMAP in dry pyridine.

fractions: A1 (99 mg), A2 (870 mg), A3 (450 mg), and B1 (760 mg), B2 (420 mg), and B3 (290 mg).

All these fractions were repeatedly chromatographed by HPLC on silica gel by using the following eluents: fraction A1 (eluent *n*-hexanes/EtOAc, 85:15) afforded eluterin H (**10**, 12.5 mg) and J (**12**, 4.5 mg); fraction A2 (*n*-hexanes/EtOAc, 65:35) afforded eluterin A (**3**, 15.5 mg), B (**4**, 3.3 mg), and I (**11**, 25.7 mg); fraction A3 (*n*-hexanes/EtOAc, 8:2) afforded eluterin F (**8**, 3.0 mg); fraction B1 (*n*-hexanes/EtOAc, 75:25) afforded eluterin G (**9**, 2.0 mg); fraction B2 (*n*-hexanes/EtOAc, 55:45) afforded eluterin C (**5**, 7.2 mg) and D (**6**, 3.9 mg); and fraction B3 (*n*-hexanes/EtOAc, 1:1) afforded eluterin E (**7**, 6.9 mg).

Cascarillin (1a, b). White powder, mp 207 °C. $[\alpha]_D^{25}$ −4 (c = 1.5, acetone). IR (KBr): ν_{max} 3601, 3518, 1692, 1505, 1389, 1298, 1109, 1047, 833 cm^{-1}. HREIMS: m/z 390.2027 (calcd for $C_{22}H_{32}O_7 - H_2O^+$, 390.2042). ^1H NMR (500 MHz, $CDCl_3$): cascarillin appeared as a mixture (ca. 3:2) of isomers with duplication of all signals; however, we have assigned the two series of ^1H NMR signals. Anomer **1a**: δ 7.40 (s, H-15), 7.37 (s, H-16), 6.55 (s, H-13), 5.93 (s, H-20), 5.08 (overlapped, H-12), 5.07 (m, H-7), 3.58 (bs, H-3), 2.62 (dd, J = 11.5, 8.5 Hz, H-11a), 2.38 (m, H1-a), 2.10 (overlapped, H-6a), 1.97 (overlapped, H-2a), 1.92 (dd, J = 11.5, 7.5 Hz, H-11b), 1.90 (overlapped, H-1b), 1.75 (overlapped, H-10), 1.70 (dd, J = 10.5, 2.5 Hz, H-2b), 1.69 (overlapped, H-6b), 1.60 (dq, J = 7.3, 3.3 Hz, H-8), 1.30 (s, H_3-18), 1.27 (s, H_3-19), 1.10 (d, J = 7.3 Hz, H_3-17). Anomer **1b**: δ 7.42 (s, H-15), 7.40 (s, H-16), 6.42 (s, H-13), 5.75 (s, H-20), 5.18 (dd, J = 8.5, 7.5 Hz, H-12), 5.12 (m, H-7), 3.58 (bs, H-3), 2.21 (dd, J = 11.5, 8.5 Hz, H-11a), 2.11 (dd, J = 11.5, 7.5 Hz, H-11b), 2.10 (m, H1-a), 2.09 (overlapped, H-6a), 1.90 (overlapped, H-2a), 1.80 (overlapped, H-1b), 1.70 (overlapped, H-10), 1.68 (overlapped, H-6b), 1.60 (overlapped, H-2b), 1.60 (overlapped, H-8), 1.38 (d, J = 7.3 Hz, H_3-17), 1.32 (s, H_3-18), 1.26 (s, H_3-19). ^{13}C NMR (125 MHz, $CDCl_3$) (data for the most abundant anomer, **1a**): δ 168.3 (s, OAc), 141.4 (d, C-15), 140.2 (d, C-16), 127.1 (s, C-13), 110.6 (d, C-20), 107.2 (d, C-14), 75.3 (d, C-3), 74.0 (s, C-4), 73.9 (d, C-7), 68.3 (d, C-12), 53.5 (d, C-10), 52.2 (s, C-9), 44.3 (d, C-8), 41.2 (t, C-6), 40.7 (t, C-11), 39.3 (s, C-5), 30.5 (t, C-2), 21.3 (q, C-18), 21.1 (q, OAc), 20.0 (q, C-19), 18.1 (t, C-1), 17.8 (q, C-17).

Acetylation of Cascarillin. Cascarillin (200 mg) was dissolved in dry pyridine (2 mL) and treated with Ac_2O (2 mL). After being kept overnight, the reaction was worked up by addition of a few drops of methanol to destroy the excess Ac_2O, water (ca. 10 mL), and EtOAc (ca. 30 mL). The organic phase was washed sequentially with 2N H_2SO_4, sat. $NaHCO_3$, and brine. After drying (Na_2SO_4) and removal of the solvent, the residue was purified by column chromatography (petroleum ether/EtOAc, 7:3) to afford 186 mg of **2a**.

Diacetylcascarillin (2a). Amorphous solid; $[\alpha]_D^{25}$ −2.5 (c = 1.2, MeOH). IR (KBr): ν_{max} 3459, 1744, 1739, 1573, 1246, 1239, 1172, 1024 cm^{-1}. HREIMS m/z 492.2368 (calcd for $C_{26}H_{36}O_9^+$, 492.2359). ^1H NMR (500 MHz, $CDCl_3$) δ 10.31 (s, H-20), 7.43 (br s, H-16), 7.35 (br s, H-15), 6.41 (br s, H-14), 5.88 (dd, J = 7.3, 4.9 Hz, H-12), 5.29 (br t, J = 3.1 Hz, H-7), 4.64 (br t, J = 1.9 Hz, H-3), 2.46 (dd, J = 16.2, 4.8 Hz, H-11a), 2.16 (dd, J = 11.5, 3.5, H-6a), 2.08 (overlapped, H_2-2, H-11b), 2.04 (s, OAc), 1.99 (s, 6H, 2 × OAc), 1.67 (overlapped, H-1a), 1.62 (H-1b, overlapped), 1.56 (dd, J = 11.5, 2.1, H-6b), 1.32 (m, H-10), 1.19 (s, H-18), 1.11 (d, J = 6.7 Hz, H-17), 1.07 (s, H-19). ^{13}C NMR (125 MHz, $CDCl_3$) δ 207.5 (s, C-20), 170.3 (s, OAc), 170.1 (s, OAc), 170.0 (s, OAc), 143.4 (d, C-16), 140.1 (d, C-15), 125.5 (s, C-13), 108.6 (d, C-14), 76.5 (d, C-3), 74.8 (s, C-4), 74.7 (d, C-7), 64.6 (d, C-12), 54.7 (s, C-9), 42.4 (d, C-10), 40.5 (s, C-5), 37.2 (d, C-8), 34.9 (t, C-6), 34.4 (t, C-2), 26.5 (t, C-11), 21.4 (q, OAc), 21.3 (q, OAc), 21.1 (q, OAc), 20.9 (q, C-18), 19.7 (q, C-19), 17.6 (t, C-1), 13.5 (q, C-17).

Reduction of Cascarillin. To a stirred solution of cascarillin (400 mg) in methanol (8 mL), excess $NaBH_4$ (85 mg) was added portionwise during ca. 5 min. The reaction was worked up by addition of saturated aq NH_4Cl (20 mL) and EtOAc (60 mL). The organic phase was washed with brine, dried (Na_2SO_4), and evaporated, to give a semisolid residue. Trituration with ether afforded 310 mg of tetrol **2b**.

Cascarillin Tetrol (2b). White powder, mp 96 °C. $[\alpha]_D^{25}$ −13.3 (c = 1.0, MeOH). IR (KBr): ν_{max} 3534, 3380, 1713, 1501, 1399, 1263, 1044, 934, 793 cm^{-1}. HREIMS m/z 410.2301 (calcd for $C_{22}H_{34}O_7^+$, 410.2305). ^1H NMR (500 MHz, $CDCl_3$): δ 7.41 (s, H-15), 7.40 (s, H-16), 6.45 (s, H-14), 5.12 (m, H-7), 4.95 (dd, J = 10.2, 2.5 Hz, H-12), 4.16 (d, J = 11.8 Hz, H-20a), 3.97 (d, J = 11.8 Hz, H-20b), 3.56 (s, H-3), 2.20 (overlapped, H-2a), 2.18 (overlapped, H-8), 2.08 (s, OAc), 2.04 (overlapped, H_2-11), 1.99 (dd, J = 14.7, 2.0 Hz, H-6a), 1.61 (overlapped, H-6b), 1.60 (overlapped, H-2b), 1.58 (overlapped, H-1a), 1.35 (s, H_3-18), 1.26 (s, H_3-19), 1.25 (overlapped, H-10), 1.23 (overlapped, H-1b), 1.08 (d, J = 7.3 Hz, H_3-17). ^{13}C NMR (125 MHz, $CDCl_3$): δ 172.7 (s, OAc), 144.3 (d, C-15), 139.7 (d, C-16),

Table 1. ^{1}H (500 MHz) NMR Data of Eluterins A–E (**3**–**7**) in $CDCl_3$ (δ Values in ppm, Multiplicity, *J* in Hz)

pos.	3	4	5	6	7
1a	2.15[a]	2.10,m	2.05[a]	1.60[a]	1.48,m
b	1.98[a]	1.69,m	1.46,dt,12.5,3.7	1.50[a]	1.23,m
2a	2.46,dd,12.5,2.9	2.08[a]	2.15[a]	2.15[a]	2.14,dd,9.6,2.2
b	2.27,m	1.54,m	1.68[a]	1.65[a]	1.61,m
3		4.35,bs	2.97,bs	2.97,bs	2.97,bs
4	2.22,q,6.6				
6a	2.11,dd,14.7,2.2	2.25[a]	2.09[a]	2.08, dd,14.0,2.2	2.09[a]
b	1.51,dd,14.7,3.7	1.71[a]	1.64,dd,14.7,4.4	1.55[a]	1.70,dd,14.7,3.7
7	5.15,dt,3.7,2.2	5.18,dt,3.7,2.2	5.18,dt, 4.4,2.2	4.08,dt,3.5,2.2	5.13,dt,3.7,2.2
8	1.95[a]	1.84,m	1.82,m	1.74,dq,7.3,3.5	2.34,dq,7.3,3.7
10	1.97[a]	1.40,d,11.8	1.15[a]	1.20,dd,10.3,1.5	1.36,d,10.3
11a	1.98[a]	2.20[a]	2.08[a]	1.98,dt,14.0,5.1	2.08[a]
b	1.85,m	1.80[a]	1.74,dt,13.2,3.7	1.62[a]	2.08[a]
12a	2.40,m	2.28[a]	2.29,dt,13.2,4.4	2.30,dt,14.0,3.7	4.81,t,5.1
b	2.38,m	1.98,dt,12.5,5.9	2.14[a]	2.18[a]	
14	6.28,s	6.28,s	6.28,s	6.27,s	6.24,s
15	7.38,s	7.38,s	7.37,s	7.35,s	7.40,s
16	7.26,s	7.22,s	7.22,s	7.20,s	7.40,s
17	1.03,d,7.3	1.00,d,7.3	0.93,d,7.3	1.08,d,7.3	1.02,d,7.3
18a	0.90,d,6.6	4.91,bs	1.18,s	1.22,s	1.20,s
b		4.82,bs			
19	0.90,s	1.45,s	1.30,s	1.35,s	1.30,s
20a	4.47,s	4.51,d,15.1	4.10,dd,11.8,2.0	4.65,d,11.8	4.16,d,11.8
b		4.49,d,15.1	3.72,dd,11.8,2.0	4.12,d,11.8	3.73,d,11.8
Ac	2.03,s	2.10,s	2.10,s	2.02,s	2.09,s
	2.08,s	2.03,s			

[a] Overlapped with other signals.

132.8 (s, C-13), 109.9 (d, C-14), 77.9 (d, C-7), 77.0 (d, C-3), 76.8 (s, C-4), 66.0 (t, C-20), 64.1 (d, C-12), 49.9 (d, C-10), 44.9 (s, C-9), 42.1 (d, C-6), 40.9 (s, C-5), 40.1 (d, C-8), 37.0 (t, C-11), 31.2 (t, C-2), 21.3 (q, OAc), 19.7 (t, C-1), 19.0 (q, C-18), 18.3 (q, C-19), 14.5 (q, C-17).

Acetylation of Tetrol 2b. A 150-mg portion of **2b** was dissolved in dry pyridine (2 mL) and treated with Ac_2O (2 mL) and DMAP (10 mg). After being kept overnight, the reaction was worked up by addition of a few drops of methanol to destroy the excess Ac_2O, water (ca. 10 mL), and EtOAc (ca. 30 mL). The organic phase was washed sequentially with 2 N H_2SO_4, saturated $NaHCO_3$, and brine. After drying (Na_2SO_4) and removal of the solvent, the residue was washed with ether to afford 141 mg of **2c** as a white powder.

Compound 2c. White powder, mp 48 °C. $[\alpha]_D^{25}$ −11.2 (*c* = 0.5, $CHCl_3$). IR (KBr): ν_{max} 3517, 3146, 1732, 1717, 1373, 1242, 1159, 1024, 982 cm^{-1}. HREIMS *m*/*z* 536.2619 (calcd for $C_{28}H_{40}O_{10}^+$, 536.2622).^{1}H NMR (500 MHz, $CDCl_3$): δ 7.42 (s, H-15), 7.37 (s, H-16), 6.41 (s, H-14), 5.92 (dd, *J* = 11.5, 10.5 Hz, H-12), 5.13 (m, H-7), 4.65 (bs, H-3), 4.58 (d, *J* = 11.8 Hz, H-20a), 4.30 (d, *J* = 11.8 Hz, H-20b), 2.31 (dd, *J* = 10.8, 7.5 Hz, H-2a), 2.07 (overlapped, H-6a), 2.06 (s, OAc), 2.05 (s, OAc), 2.05 (overlapped, H_2-11), 2.02 (s, OAc), 2.00 (s, OAc), 1.95 (dq, *J* = 7.3, 3.3 Hz, H-8), 1.78 (dd, *J* = 10.8, 2.5 Hz, H-2b), 1.60 (overlapped, H-1a), 1.59 (overlapped, H-6b), 1.58 (overlapped, (H-1b), 1.30 (dd, *J* = 10.8, 2.5 Hz, H-10), 1.21 (s, H_3-18), 1.07 (s, H_3-19), 1.06 (d, *J* = 7.3 Hz, H-17). ^{13}C NMR (125 MHz, $CDCl_3$): δ 171.0 (s, OAc), 170.8 (s, OAc), 170.1 (s, OAc), 170.0 (s, OAc), 143.0 (d, C-15), 140.0 (d, C-16), 126.5 (s, C-13), 108.5 (d, C-14), 77.6 (d, C-3), 77.5 (d, C-7), 77.5 (s, C-4), 65.0 (t, C-20), 62.5 (d, C-12), 42.4 (d, C-10), 41.0 (s, C-9), 40.3 (t, C-6), 38.3 (d, C-8), 37.8 (d, C-5), 35.5 (t, C-11), 27.4 (t, C-2), 21.5 (q, OAc), 21.3 (q, OAc), 21.2 (q, OAc), 21.0 (q, OAc), 20.5 (t, C-1), 18.3 (t, C-18), 18 (t, C-19), 14 (t, C-17).

Oxidation of Cascarillin. To a solution of cascarillin (400 mg, 0.98 mmol) in dry CH_2Cl_2 (10 mL), pyridinium dichromate (PDC, 900 mg, 2.44 mmol, 2.5 mol equiv) and Celite (200 mg) were added. The suspension was stirred at room temp for 24 h, and then worked up by the addition of ether (ca. 50 mL), filtration, and evaporation. The black gummy residue was purified by column chromatography (10 g silica gel, elution with petroleum ether/EtOAc, 7:3) followed by HPLC (LUNA, SI60, *n*-hexane/EtOAc, 7:3) to afford cascarillin ketolactone **1c** (144 mg, 36%) and diketo aldehyde **1d** (72 mg, 18%).

3-Dehydrocascarillinlactone (1c). Amorphous solid. $[\alpha]_D^{25}$ −8 (*c* = 0.4, MeOH). IR (KBr): ν_{max} 3495, 3154, 1764, 1747, 1730, 1599, 1502, 1461, 1387, 1254, 1186, 1015 cm^{-1}. HREIMS: *m*/*z* 404.1835 (calcd for $C_{22}H_{28}O_7^+$, 404.1835). ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.45 (br s, H-15), 7.45 (br s, H-16), 6.39 (br s, H-14), 5.40 (t, *J* = 8.5 Hz, H-12), 5.10 (m, H-7), 2.85, (ddd, *J* = 11.5, 7.5, 2.5 Hz, H-2a), 2.52 (d, *J* = 8.5 Hz, H_2-11), 2.32 (dd, *J* = 11.5, 3.5 Hz, H-2b), 2.07 (overlapped, H-6a), 2.06 (s, OAc), 1.97 (dq, *J* = 7.3, 3.5 Hz, H-8), 1.82 (overlapped, H-1a), 1.82 (overlapped, H-6b), 1.78 (overlapped, H-1b), 1.30 (overlapped, H-10), 1.26 (s, H_3-18), 1.20 (d, *J* = 7.3 Hz, H_3-17), 1.19 (s, H_3-19). ^{13}C NMR (125 MHz, $CDCl_3$): δ 210.8 (s, C-3), 175.2 (s, C-20), 171.1 (s, OAc), 144.1 (d, C-15), 139.5 (d, C-16), 126.0 (s, C-13), 107.9 (d, C-14), 80.8 (d, C-11), 72.4 (s, C-4), 71.8 (d, C-7), 52.0 (d, C-10), 47.0 (s, C-9), 43.1 (t, C-6), 41.9 (d, C-8), 41.8 (d, C-5), 37.0 (t, C-11), 34.3 (t, C-2), 22.5 (t, C-1), 21.2 (q, OAc), 17.9 (q, C-18), 17.0 (q, C-19), 16.8 (q, C-17).

3,12,20-Tridehydrocascarillin (1d). Amorphous solid. $[\alpha]_D^{25}$ −12 (*c* = 0.4, MeOH). IR (KBr): ν_{max} 3560, 1743, 1730, 1595, 1506, 1351, 1265, 1114, 938, 795 cm^{-1}. HREIMS *m*/*z* 404.1830 (calcd for $C_{22}H_{28}O_7$, 404.1835). ^{1}H NMR (500 MHz, $CDCl_3$): δ 10.33 (s, H-20), 8.09 (s, H-15), 7.46 (s, H-16), 7.25 (s, H-14), 5.37 (m, H-7), 3.41 (d, *J* = 13.5 Hz, H-11a), 2.96 (d, *J* = 13.5 Hz, H-11b), 2.83 (ddd, *J* = 11.5, 7.5, 2.5 Hz, H-2a,), 2.76 (dq, *J* = 7.3, 3.3 Hz, H-8), 2.30 (dd, *J* = 11.5, 3.5 Hz, H-2b), 2.21 (dd, *J* = 12.5, 3.5 Hz, H-6a), 2.07 (s, OAc), 1.89 (dd, *J* = 12.5, 2.1 Hz, H-6b), 1.60 (overlapped, H-10), 1.55 (ddd, *J* = 11.8, 7.5, 3.5 Hz, H-1a,) 1.19 (s, H_3-18), 1.03 (d, *J* = 7.3 Hz, H_3-17), 0.90 (s, H_3-19). ^{13}C NMR (125 MHz, $CDCl_3$): δ 211.2 (s, C-3), 207 (s, C-12), 193.4 (s, C-20), 170.0 (s, OAc), 147.5 (d, C-15), 144.7 (d, C-16), 128.5 (s, C-13), 108.2 (d, C-14), 80.0 (s, C-4), 74.8 (d, C-7), 56.3 (d, C-10), 42.5 (s, C-9), 40.0 (t, C-6), 38.0 (d, C-8), 36.7 (s, C-5), 36.4 (t, C-11), 34.3 (t, C-2), 22.8 (t, C-1), 21.7 (q, OAc), 20.0(q, C-18), 20.0 (q, C-19), 17.1 (q, C-17).

Eluterin A (3). White powder. $[\alpha]_D^{25}$ −5 (*c* = 0.1, $CHCl_3$). IR (KBr): ν_{max} 1722, 1705, 1240, 875 cm^{-1}. ESIMS (positive mode): *m*/*z* 441 $[M + Na]^+$. HREIMS: found, *m*/*z* 418.2368; calcd for $C_{24}H_{34}O_6$, *m*/*z* 418.2355. ^{1}H and ^{13}C NMR data are reported in **Tables 1** and **2**, respectively.

Eluterin B (4). White powder. $[\alpha]_D^{25}$ −13 (*c* = 0.02, $CHCl_3$). IR (KBr): ν_{max} 3640, 1722, 1240, 910, 872 cm^{-1}. ESIMS (positive ion mode): *m*/*z* 441 $[M + Na]^+$. HREIMS: found, *m*/*z* 418.2390; calcd

Table 2. ^{13}C NMR Data of Eluterins A–E (**3**–**7**) in $CDCl_3$ (δ in ppm, Multiplicity)

pos.	3	4	5	6	7
1	23.1, CH_2	19.5, CH_2	19.3, CH_2	20.2, CH_2	19.5, CH_2
2	40.8, CH_2	37.2, CH_2	28.2, CH_2	28.1, CH_2	30.2, CH_2
3	211.3, C	76.5, CH	62.3, CH	62.5, CH	62.4, CH
4	59.2, CH	160.2, C	65.9, C	66.0, C	65.4, C
5	39.9, C	41.3, C	36.4, C	37.7, C	37.3, C
6	43.3, CH_2	43.0, CH_2	40.5, CH_2	42.5, CH_2	41.2, CH_2
7	77.4, CH	77.7, CH	75.8, CH	70.9, CH	75.4, CH
8	38.3, CH	40.1, CH	44.3, CH	45.1, CH	40.2, CH
9	41.9, C	42.2, C	39.3, C	40.2, C	45.3, C
10	48.0, CH	51.0, CH	47.5, CH	49.1, CH	49.1, CH
11	33.3, CH_2	36.8, CH_2	37.0, CH_2	40.3, CH_2	33.2, CH_2
12	21.7, CH_2	23.1, CH_2	24.4, CH_2	24.3, CH_2	64.2, CH
13	124.2, C	126.2, C	125.6, C	125.5, C	131.0, C
14	110.5, CH	113.2, CH	111.1, CH	110.3, CH	109.6, CH
15	142.2, CH	144.0, CH	141.2, CH	142.0, CH	142.1, CH
16	138.1, CH	139.3, CH	137.3, CH	138.2, CH	138.2, CH
17	14.5, CH_3	15.1, CH_3	18.1, CH_3	18.1, CH_3	18.2, CH_3
18	8.5, CH_3	112.5, CH_2	18.3, CH_3	17.8, CH_3	20.3, CH_3
19	17.3, CH_3	25.5, CH_3	19.2, CH_3	20.0, CH_3	17.3, CH_3
20	69.3, CH_2	69.5, CH_2	66.3, CH_2	69.4, CH_2	64.8, CH_2
Ac	171.2, C	171.2, C	171.2, C	170.4, C	171.0, C
	22.1, CH_3	23.0, CH_3	22.0, CH_3	21.2, CH_3	21.3, CH_3
	170.5, C	170, C			
	23.1, CH_3	22.7, CH_3			

for $C_{24}H_{34}O_6$, *m/z* 418.2355. ^1H and ^{13}C NMR data are reported in **Tables 1** and **2**, respectively.

Eluterin C (5). White powder. $[\alpha]_D^{25}$ −40 (c = 0.04, $CHCl_3$). IR (KBr): ν_{max} 3655, 1722, 1240, 890 cm^{-1}. ESIMS (positive ion mode): *m/z* 399 $[M + Na]^+$. HREIMS: found, *m/z* 376.2222; calcd for $C_{22}H_{32}O_5$, *m/z* 376.2250. ^1H and ^{13}C NMR data are reported in **Tables 1** and **2**, respectively.

Eluterin D (6). White powder. $[\alpha]_D^{25}$ −28 (c = 0.02, $CHCl_3$). IR (KBr): ν_{max} 3620, 1720, 1228, 888 cm^{-1}. ESIMS (positive ion mode): *m/z* 399 $[M + Na]^+$. HREIMS: found, *m/z* 376.2240; calculated for $C_{22}H_{32}O_5$ *m/z* 376.2250. ^1H and ^{13}C NMR data are reported in Tables 1 and 2, respectively.

Eluterin E (7). White powder. $[\alpha]_D^{25}$ −21 (c = 0.02, $CHCl_3$). IR (KBr): ν_{max} 3650, 1721, 1240, 890 cm^{-1}. ESIMS (positive ion mode): *m/z* 415 $[M + Na]^+$. HREIMS: found *m/z* 392.2210; calcd for $C_{22}H_{32}O_6$, *m/z* 392.2199. ^1H and ^{13}C NMR data are reported in **Tables 1** and **2**, respectively.

Preparation of the MTPA diesters of eluterin E. Eluterin E (**7**, 1.5 mg) was dissolved in 1.5 mL of dry pyridine, treated with (−)-MTPA chloride (90 μL) and then maintained at room temperature, with stirring, overnight. Solvent was removed from the reaction mixture, and the mixture was purified by HPLC on SI60 column (eluent *n*-hexane/EtOAc, 85:15), affording (*S*)-MTPA ester **7a** in a pure state (1.6 mg). Using (+)-MTPA chloride, the same procedure afforded the (*R*)-MTPA ester **7b** in the same yield. $\Delta\delta$ (**7a**−**7b**) values are reported in **Figure 3**.

Compound 7a (*S* MTPA diester). Amorphous solid. FABMS (glycerol matrix, positive ions): *m/z* 825 $[M + H]^+$. ^1H NMR (500 MHz, $CDCl_3$): δ 7.47 and 7.37 (MTPA phenyl protons), 7.43 (s, H-15), 7.41 (s, H-16), 6.27 (s, H-14), 5.63 (t, J = 5.1 Hz, H-12), 5.14 (m, H-7), 4.55 (d, J = 11.8 Hz, H-20a), 4.21 (d, J = 11.8 Hz, H-20b), 3.59 (MTPA-OCH_3), 2.98 (bs, H-3), 2.55 (m, H_2-11), 2.36 (dq, J = 7.3, 3.7 Hz, H-8), 2.14 (dd, J = 9.6, 2.2 Hz, H-2a), 2.10 (s, OAc), 2.09 (overlapped, H-6a), 1.70 (dd, J = 14.8, 3.7 Hz, H-6b), 1.60 (m, H-2b), 1.48 (m, H-1a), 1.34 (d, J = 10.3 Hz, H-10), 1.30 (s, H_3-19), 1.26 (m, H-1b), 1.21 (s, H_3-18), 1.02 (d, J = 7.3 Hz, H_3-17).

Compound 7b (*R* MTPA diester). Amorphous solid. FABMS (glycerol matrix, positive ions): *m/z* 825 $[M + H]^+$. ^1H NMR (500 MHz, $CDCl_3$): δ 7.55 and 7.33 (MTPA phenyl protons), 7.45 (s, H-15), 7.43 (s, H-16), 6.38 (s, H-14), 5.63 (t, J = 5.1 Hz, H-12), 5.13 (m, H-7), 4.44 (d, J = 11.8 Hz, H-20a), 4.10 (d, J = 11.8 Hz, H-20b), 3.64 (MTPA-OCH_3), 2.98 (bs, H-3), 2.50 (m, H_2-11), 2.37 (dq, J = 7.3, 3.7 Hz, H-8), 2.14 (dd, J = 9.6, 2.2 Hz, H-2a), 2.10 (s, OAc), 2.09 (overlapped, H-6a), 1.71 (dd, J = 14.8, 3.7 Hz, H-6b), 1.60 (m, H-2b), 1.48 (m, H-1a), 1.32 (d, J = 10.3 Hz, H-10), 1.30 (s, H_3-19), 1.26 (m, H-1b), 1.21 (s, H_3-18), 1.02 (d, J = 7.3 Hz, H_3-17).

Eluterin F (8). White powder. $[\alpha]_D^{25}$ −4 (c = 0.01, $CHCl_3$). IR (KBr): ν_{max} 1724, 1722, 1233, 885 cm^{-1}. ESIMS (positive ion mode): *m/z* 499 $[M + Na]^+$. HREIMS: found, *m/z* 476.2433; calcd for $C_{26}H_{36}O_8$, *m/z* 476.2410. ^1H and ^{13}C NMR data are reported in **Tables 3** and **4**, respectively.

Eluterin G (9). White powder. $[\alpha]_D^{25}$ −25 (c = 0.01, $CHCl_3$). IR (KBr): ν_{max} 1746, 1734, 1233, 891 cm^{-1}. ESIMS (positive ion mode): *m/z* 455 $[M + Na]^+$. HREIMS: found, *m/z* 432.2153; calcd for $C_{24}H_{32}O_7$, *m/z* 432.2148. ^1H and ^{13}C NMR data are reported in **Tables 3** and **4**, respectively.

Table 3. ^1H (500 MHz) NMR Data of Eluterins F–J (**8**–**12**) in $CDCl_3$ (δ Values in ppm, Multiplicity, J in Hz)

pos.	8	9	10	11	12
1a	1.71[a]	2.00,m	1.76,m	1.43[a]	1.55[a]
b	1.53[a]	1.53,m	1.63,m	1.41[a]	1.31[a]
2a	2.18,dd,13.2,2.9	2.18,m	2.05[a]	2.11[a]	2.15,dd,11.8,2.5
b	1.68[a]	1.63[a]	1.95[a]	1.58[a]	1.72,ddd,11.8,7.5,3.5
3	2.90,bs	2.90,bs	5.17,bs	2.92,bs	2.98,bs
6a	4.90,d,3.7	4.95,d,3.5	2.17,dd,13.5,2.5	2.12[a]	2.07[a]
b			1.42[a]	1.60[a]	1.62,dd,11.5,3.7
7	5.43,t,3.7	5.54,t,3.5	5.05,dt,3.7,2.5	5.07[a]	5.16,dt,3.7,2.5
8	2.02[a]	2.09[a]	2.03[a]	2.03,dq,7.3,3.5	1.53[a]
10	1.25,dd,10.5,2.3	1.34,dd,11.1,2.3	1.40[a]	1.01,dd,11.5,2.2	1.30[a]
11a	1.97[a]	2.01[a]	2.43,dd,13.5,6.2	2.37,dd,11.5,7.5	
b	1.57[a]	1.68[a]	1.93[a]	1.86,dd,11.5,9.5	
12a	2.29,dt,14.0,3.7	2.28,dd,12.9,3.4	5.11,t,6.2	5.08[a]	4.52,s
b	2.13[a]	2.18[a]			
14	6.25,s	6.26,s	6.42,s	6.38,s	6.45,s
15	7.38,s	7.37,s	7.44,s	7.40,s	7.52,s
16	7.23,s	7.24,s	7.41,s	7.39,s	7.39,s
17	0.95,d,7.3	1.11,d,6.9	1.35,d,6.7	1.33,d,7.3	1.18,d,7.3
18	1.16,s	1.18,s	1.57,s	1.15,s	1.04,s
19	1.35,s	1.25,s	1.26,s	1.26,s	1.00, s
20a	4.44,d,11.8	10.13,s	6.77,s	6.58,s	4.58,d,11.9
b	4.25,d,11.8				4.10,d,11.9
Ac	2.12,s	2.10,s	2.01,s	2.02,s	2.08,s
	2.03,s	2.03,s	2.10,s	2.08,s	
	2.02,s				

[a] Overlapped with other signals.

Table 4. ^{13}C NMR Data of Eluterins F–J (**8**–**12**) in CDCl (δ in ppm, Multiplicity)

pos.	8	9	10	11	12
1	19.2, CH_2	18.7, CH_2	23.2, CH_2	16.3, CH_2	20.2, CH_2
2	30.5, CH_2	29.5, CH_2	29.1, CH_2	28.6, CH_2	28.6, CH_2
3	64.4, CH	65.1, CH	121.4, CH	61.2, CH	62.0, CH
4	66.0, C	67.0, C	142.2, C	65.6, C	64.4, C
5	37.4, C	37.3, C	38.4, C	37.5, C	37.5, C
6	77.2, CH	76.6, CH	39.2, CH_2	40.0, CH_2	40.5, CH_2
7	75.1, CH	75.3, CH	74.3, CH	74.5, CH	75.5, CH
8	39.6, CH	38.5, CH	43.2, CH	43.2, CH	42.1, CH
9	41.2, C	55.3, C	51.0, C	53.2, C	55.3, C
10	47.2, CH	49.5, CH	52.3, CH	52.4, CH	50.2, CH
11	34.3, CH_2	33.8, CH_2	45.4, CH_2	46.3, CH_2	213.1, C
12	21.8, CH_2	17.8, CH_2	71.2, CH	72.2, CH	105.5, CH
13	126.0, C	125.2, C	125.5, C	125.5, C	117.1, C
14	113.1, CH	112.3, CH	116.3, CH	109.1, CH	109.1, CH
15	142.3, CH	142.2, CH	144.3, CH	138.6, CH	142.2, CH
16	138.2, CH	138.4, CH	138.1, CH	144.2, CH	140.2, CH
17	14.2, CH_3	15.0, CH_3	14.2, CH_3	14.2, CH_3	20.3, CH_3
18	21.3, CH_3	22.1, CH_3	18.7, CH_3	17.7, CH_3	17.7, CH_3
19	19.1, CH_3	18.3, CH_3	19.5, CH_3	18.1, CH_3	14.8, CH_3
20	68.0, CH_2	207.3, CH	99.3, CH	98.9, CH	73.2, CH
Ac	171.4, C	171.4, C	170.1, C	171.3, C	170.3, C
	23.1, CH_3	23.1, CH_3	20.3, CH_3	21.2, CH_3	21.2, CH_3
	170.9, C	170.6, C	169.5, C	169.5, C	
	23.4, CH_3	23.4, CH_3	20.1, CH_3	20.2, CH_3	
	170.5, C				
	22.5, CH_3				

Eluterin H (10). White powder. $[\alpha]_D^{25}$ −57 (c = 0.07, $CHCl_3$). IR (KBr): ν_{max} 1739, 1502, 1461, 1387, 1233, 890 cm^{-1}. ESIMS (positive ion mode): m/z 439 $[M + Na]^+$. HREIMS: found, m/z 416.2220; calcd for $C_{24}H_{32}O_6$, m/z 416.2199. ^1H and ^{13}C NMR data are reported in **Tables 3** and **4**, respectively.

Eluterin I (11). White powder. $[\alpha]_D^{25}$ −48 (c = 0.02, $CHCl_3$). IR (KBr): ν_{max} 1738, 1461, 1390, 1233, 888 cm^{-1}. ESIMS (positive ion mode): m/z 455 $[M + Na]^+$. HREIMS: found, m/z 432.2150; calcd for $C_{24}H_{32}O_7$, m/z 432.2148. ^1H and ^{13}C NMR data are reported in **Tables 3** and **4**, respectively.

Eluterin J (12). White powder. $[\alpha]_D^{25}$ −37 (c = 0.02, $CHCl_3$). IR (KBr): ν_{max} 1766, 1747, 1730, 1599, 1461, 1387, 1233, 888 cm^{-1}. ESIMS (positive ion mode): m/z 411 $[M + Na]^+$. HREIMS: found, m/z 388.1893; calcd for $C_{22}H_{28}O_6$, m/z 388.1886. ^1H and ^{13}C NMR data are reported in **Tables 3** and **4**, respectively.

RESULTS AND DISCUSSION

Chromatographic purification of the acetone extract from a commercial sample of cascarilla bark from the Bahamas afforded two crystalline compounds. These were identified as the triterpene lupeol and the clerodane diterpenoid cascarillin (0.10% and 0.62% on the dried plant material, respectively). All the other constituents were isolated in much lower yield, and their purification required further, extensive chromatographic steps.

The NMR spectra of cascarillin (**1a, b**) showed duplication of all signals. The lack of an aldehyde resonance and its replacement by two semiacetal carbons at δ 110.6 and 109.2 in the ^{13}C NMR spectrum indicated the presence of a ca. 3:2 mixture of γ-lactols; the rapid mutual conversion of **1a** and **1b** did not allow the isolation of the two epimers as pure compounds. To obtain single derivatives amenable to full spectroscopic analysis, a few simple reactions were carried out on cascarilla in order to obtain a single derivative for the spectroscopic analysis. Thus, acetylation selectively trapped the fledging hydroxyaldehyde in equilibrium with the two semiacetal forms, affording the diacetate **2a** as the only reaction product (**Figure 1**), while hydride reduction gave the tetrol **2b**, further characterized as the peracetate **2c**. Attempts to methylate cascarillin gave a complex mixture under different conditions (MeI/Ag_2O; dimethyl sulfate/K_2CO_3), whereas oxidation with transition metal-based reagents (PDC) afforded a separable mixture of the ketolactone **1c** and the diketo aldehyde **1d**. These reactions afforded compounds easily amenable to spectroscopic analysis and further supported the existence of a lactol/hydroxyaldehyde equilibrium in the natural product.

The diterpenoid fraction of cascarilla afforded a series of novel furo-clerodanes differing from cascarillin in the modification of the 3,4-diol system, the functionalization of the linkage between the furan and decalin rings, and/or the presence of an additional oxygen function at C-6. Owing to substantial structural deviations from cascarillin, and the confusing situation regarding the trivial names of cascarilla diterpenoids (*5*, *6*), we have named these new compounds eluterins.

HRMS analysis of eluterin A (**3**, **Figure 2**) indicated the molecular formula $C_{24}H_{34}O_6$. The presence of IR (KBr) absorptions at ν_{max} 1705 and 1722 cm^{-1} suggested the presence of ketone and ester functionalities, respectively; a finding supported by ^{13}C NMR spectrum (**Table 2**), which contains one ketone (singlet at δ 211.3) and two ester signals (singlets at δ 170.5 and 171.2). In addition, carbon resonances ascribable to oxymethylene (δ 69.3, t) and oxymethine (δ 77.4, d) groups were present; the corresponding proton signals appeared at δ 5.15 (double triplet, oxymethine) and δ 4.47 (broad singlet, oxymethylene) in the ^1H NMR spectrum of **3** ($CDCl_3$, **Table 1**). This also showed three broad singlets at δ 6.28, 7.26, and 7.38, typical proton resonances of a β-alkyl substituted furan ring, and a series of resonances between δ 2.50 and 0.90. These last signals encompassed three methyl singlets (δ 0.90, 2.03, and 2.08), and a series of multiplets resolved by COSY experiment into four spin systems. The first one extended from H-10 to a deshielded methylene (δ 2.46 and 2.27) passing through H_2-1; the second one connected a methylene (H-6a,b) to a methyl doublet (H_3-17) via an oxymethine (H-7) followed by a tertiary methine (H-8). The remaining systems were an isolated CH_2CH_2 group (H-11a,b, H-12a,b) and a methyl-substituted methine (H-4, H_3-18).

The individual spin systems could be combined by HMBC analysis into the clerodane structure **3**, having the following: (i) a ketone group at C-3 (HMBC cross-peaks H_3-18/C-3, H-4/C-3, and H_2-1/C-3); (ii) a furan ring connected to C-9 by a CH_2CH_2 moiety (HMBC cross-peaks of H_2-12 with C-13, C-14, and C-16); (iii) an acetoxymethylene at C-9 (HMBC correlations of H_2-20 with the ester carbonyl at δ 171.2, and with C-8, C-9, C-10, and C-11); and (iv) an acetoxyl at C-7 (HMBC correlation of H-7 with one of the acetoxy carbonyls).

The 2D ROESY spectrum of **3** was useful in assigning the relative configuration of this compound. Thus, H-10 showed correlation with H-8 and the methyl H_3-18, while the methylene H_2-20 correlated with the methyl H_3-19. Finally, the α-orientation for the acetoxyl at C-7 was deduced from the coupling constant pattern of H-7, which indicated the equatorial (α)-orientation of this proton. The relative configuration at C-5, C-7, C-8, C-9, and C-10 of **3**, thus deduced, is the same as that of cascarillin, and the two compounds most likely share also the absolute configuration; therefore, the stereostructure reported in formula **3** has been assigned to eluterin A.

High-resolution mass measurements of eluterin B (**4**) showed the same molecular formula as eluterin A (**3**), from which it differs only in the functionalization of C-3, C-4, and C-18. Thus, in the IR (KBr) spectrum of **4**, the ketone band was replaced by a hydroxyl absorption (ν_{max} 3640 cm^{-1}), while the ^1H NMR

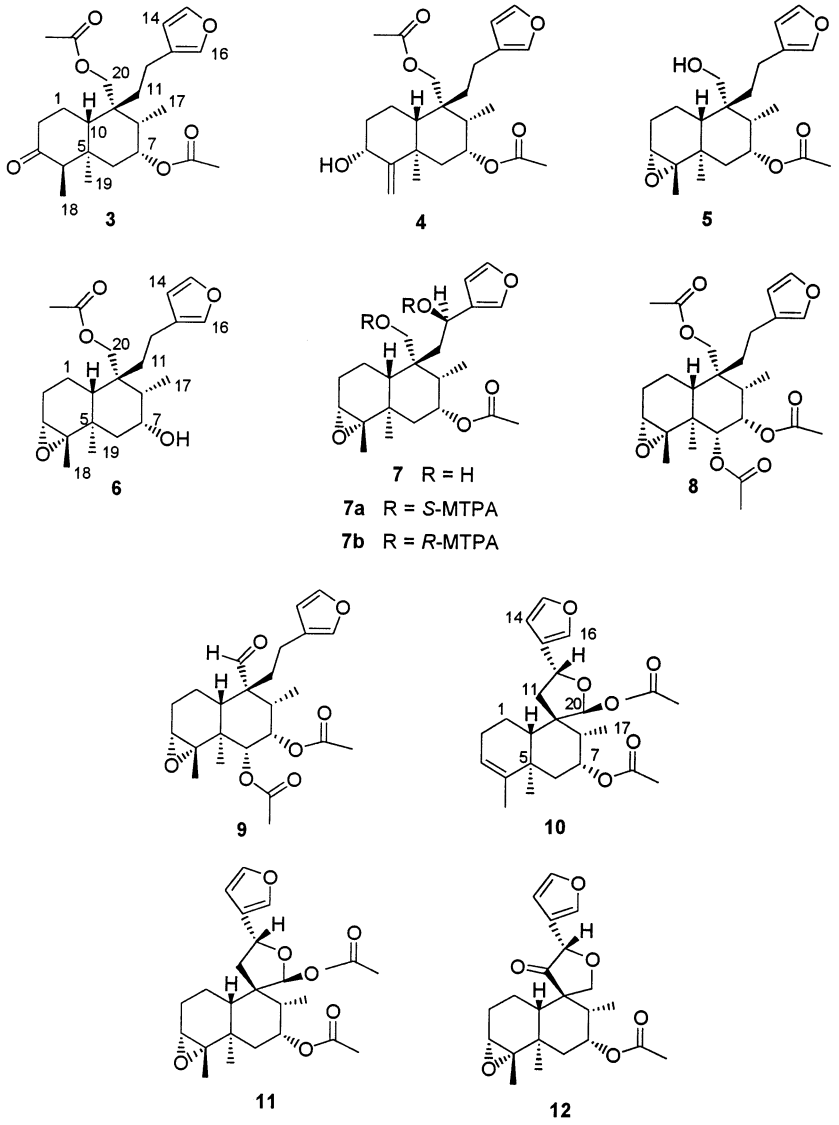

Figure 2. Eluterins A–J (**3**–**12**).

spectrum of **4** ($CDCl_3$, **Table 1**) was almost superimposable to that of **3,** apart from the resonances of ring A protons. An additional oxymethine signal at δ 4.35 and the replacement of the secondary methyl at C-4 with an *exo*-methylene (δ 4.91 and 4.82, broad singlets, associated through the HMQC spectrum with the carbon triplet at δ 112.5) were the only significant differences. The *exo*-methylene protons showed HMBC (^3J) coupling with C-3 (δ 76.5, coupled with signal at δ 4.35 in the HMQC spectrum), C-4 (δ 160.2), and C-5 (δ 41.3), unambiguously indicating the proposed structure **4**. A parallel pattern of ROESY correlations suggested that eluterin B possesses the same configuration as eluterin A for the stereogenic centers belonging to the decalin system. The α-orientation for both the acetoxy group at C-7 and the OH group at C-3 was deduced by the relatively small coupling constants observed for both H-7 (dt, J = 3.7, 2.2 Hz) and H-3 (broad singlet), indicative of the equatorial positions of these protons.

Compared to eluterins A and B, eluterin C (**5**, $C_{22}H_{32}O_5$, HRMS) lacked the 20-acetyl group and possessed, as additional functionality, a 3,4-epoxide ring, a structural motif common to most eluterins. The hallmark of these changes was the upfield resonance of the 20-methylene (δ 4.10 and 3.72), the appearance of a broad singlet at δ 2.97 in the ^{1}H NMR spectrum (**Table 1**), and ^1J coupled (HMQC correlation) with the carbon atom resonating at δ 62.3. The HMBC cross-peaks of the signal at δ 2.97 with C-2 (δ 28.2), C-1 (δ 19.3), C-4 (δ 65.9), C-5 (δ 36.4), and C-18, and of H_3-18 with C-3, C-4, and C-5 unambiguously located the epoxide ring at positions 3 and 4. In the same way, the acetoxy group was located at C-7 and the oxymethylene group was located at C-20. The α-orientation of the epoxy group was in accordance with the presence of a ROESY correlation between H_3-18 and H-10. The remaining ROESY cross-peaks indicated that the configurations of C-5, C-8, and C-9 in eluterin C and A are the same.

Four analogues of eluterin C (eluterins D−G, **6**−**9**, **Figure 2**) were also isolated, and these were structurally elucidated by comparison with the NMR spectra of **5**. On the basis of the spectroscopic analysis, compounds **5**−**9** differ from one another only in the functionalization at C-6, C-12, and C-20 already indicated that structures **5**−**9** have been deduced by the sum of all the spectral data. In particular, they showed identical scalar and dipolar couplings of the decalin protons, suggesting that these moieties have the same steric features. Eluterin D (**6**) has the same molecular formula as that of eluterin C, from which it differs only for the acylation pattern. The downfield shift of H-20 ($\Delta\delta$ = +0.50), and the upfield shift of H-7 ($\Delta\delta$ = −1.10) in **6** located the acetyl at the primary rather than the secondary hydroxyl.

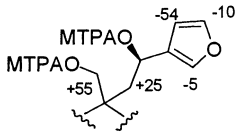

Figure 3. Application of the modified Mosher's method for secondary alcohols to eluterin E. $\Delta\delta$ ($\delta_S - \delta_R$) values are given in Hz.

Compared to eluterin D, eluterins E−G showed further oxygenation of the clerodane framework. Eluterin E (**7**, $C_{22}H_{32}O_6$, HREIMS) is structurally characterized by hydroxylation as the CH_2CH_2 moiety connecting the furan and decalin rings. Location of the hydroxyl at C-12 (δ 64.2) was secured by inspection of the HMBC spectrum, which evidenced a correlation of the oxymethine proton (δ 4.81) and the furan carbons C-13, C-14, and C-16. To assign the configuration at C-12, **7** was independently treated with (−)- and (+)-MTPA chloride in dry pyridine, providing the corresponding *S*-(**7a**) and *R*-(**7b**) MTPA diester, respectively. The distribution of $\Delta\delta$ ($S-R$) values (**Figure 3**), reflecting the anisotropic effect of MTPA according to the Mosher's model (*8*), indicated a 12*R* configuration, the same as that observed in cascarillin.

In eluterins F (**8**) and G (**9**), the site of further oxygenation is the decalin core. In both compounds, analysis of COSY spectral data readily indicated that an additional acetoxyl group is present at C-6, and its α-orientation was established by inspection of the ROESY correlations. Eluterins F (**8**) and G (**9**) differed only for the functionalization of C-20, which was an acetoxymethyl in **8** (AB system at δ 4.25 and 4.44; δ_C 68.0) and a formyl in **9** (δ_H 10.13, singlet; δ_C 207.3).

In the remaining three eluterins, an oxygen bridge was present between C-12 and C-20, resulting in a spirane ring centered on C-9. Compared to eluterin C (**3**), eluterin H (**10**, **Figure 2**) ($C_{24}H_{32}O_6$, from HREIMS and ^{13}C NMR data) also showed changes in the decalin ring, as the 3,4-epoxide was replaced by a double bond (δ 5.17, HMQC correlated to the carbon at δ 121.4). Apart from the signals of a β-substituted furan ring (δ 6.42, H-14; δ 7.44, H-15; δ 7.41, H-16), the olefinic proton H-3 (δ 5.17), and the oxymethine at C-7 (δ 5.05), two further signals (δ 6.77 and 5.11) were present in the low-field part of the ^1H NMR spectrum of **10**; the remaining proton signals were confined between δ 1.25 and 2.45. All the ^1H NMR signals (**Table 3**) were associated with the relevant carbon signals of the ^{13}C NMR spectrum (**Table 4**) through the HMQC experiment and, in particular, a hemiacetal carbon resonance (δ 99.3) was associated with the signal at δ 6.77.

The COSY spectrum of **10** indicated the presence of three partial structures, which were interconnected with the aid of the HMBC experiment. The first spin system extends from H-10 (δ 1.40) to H-3, which exhibited also an allylic long-range coupling with the methyl at δ 1.57. In turn, this latter signal showed HMBC cross-peaks with both the sp^2 signals at δ 121.4 (C-3) and δ 142.2 (C-4), and with C-5 (δ 38.4). The second spin system connects H_2-6 to H_3-17. The junction between the decalin system and the furan moiety encompassed the isolated hemiacetal proton (δ 6.77) and a CH_2CH moiety. The HMBC cross-peaks of the proton at δ 6.77 with C-8, C-9, C-10, C-11, and C-12, as well as with the acetyl carbonyl at δ 169.5, were crucial to build up the tetrahydrofuran ring. This is an acetylated version of the spirosemiacetal system of cascarillin, which, as reported above, was not obtained by standard acetylation of cascarillin. The relative configuration of the stereogenic carbons in the spirane moiety was assessed by an analysis of the ROESY correlations (H-20/H_3-19, H-12/H_3-17). It should be noted that the 20-deacetyl derivative of **10** was reported under the name of cascarillin C from a sample of cascarilla collected in Equador (*6*).

Eluterin I (**11**) ($C_{24}H_{32}O_7$, HRMS) is a close analogue of eluterin H (**10**) from which it differed only in the epoxidation of the decalin 3,4-double bond, as evidenced by the replacement in the ^1H NMR spectrum (**Table 3**) of the olefin signal at δ 5.17 with a 1H broad singlet at δ 2.92, and by the lack of olefin resonances in the ^{13}C NMR spectrum (**Table 4**), where two additional resonances were present in the midfield region (δ 61.2, d and 65.6, s). Both scalar and dipolar couplings of eluterin I (**11**) were identical to those above-described for eluterin H, whereas the ROESY cross-peak between the H_3-18 and H-10 indicated α-orientation for the epoxide oxygen (*9*).

Eluterin J (**12**) turned out to be characterized by the oxygenation of C-11, a very unusual feature in furoclerodanes (*10*, *11*). The NMR spectroscopic data of decalin and furan moieties were quite similar to those of eluterin I (**11**); but in contrast the partial structure connecting them displayed ^1H and ^{13}C NMR resonances (**Tables 3** and **4**, respectively) indicative of an oxymethylene group (AB system at δ_H 4.58 and 4.10, δ_C 73.2, t), a keto group (δ_C 213.1, s), and an oxymethine group (δ_H 4.52, δ_C 105.5, d). All proton resonances were spectroscopically isolated in terms of scalar coupling from the furan and the decalin domain. Taking into account that the molecular formula required the accommodation of a further unsaturation and the presence of only two oxygens on the chain, the keto group was located at C-11 and an ether bridge was positioned to connect the oxymethine and the oxymethylene group. This assumption was supported by the detection of a HMBC correlation between the oxymethine proton (H-12) and the oxymethylene carbon (C-20). The configuration of C-12 was established by the detection of a ROESY correlation between H-12 and the H_3-17. In a similar way, the configuration at C-9 could be established from the ROESY correlation pattern of H_2-20 (cross-peaks with H_3-19, H-14, and H-16). The α-carbon of tetrahydrofurans are reported to be remarkably sensitive to the deshielding effect of an adjacent oxygenated function (*12*), and this is presumably responsible for the unusually downfield chemical shift of the oxymethine carbon C-12 (δ 105.5).

C. eluteria is a prolific producer of diterpenoids. In addition to sizable amounts of cascarillin (ca. 0.62%), a series of clerodane diterpenoids structurally differing mostly in the functionalization of the C-3/C-4 fragment and the decalin−furan tether were also obtained. An interesting finding is the occurrence of compounds with an acetylated semiacetal moiety (**10**, **11**). Indeed, we were unable to obtain compounds of this type by standard acetylation of cascarillin, a reaction which selectively trapped the hydroxyaldehyde form at the expense of the semiacetalic ones.

Clerodanes are versatile natural products with regard to bioactivity. Early studies centered mainly on their insect antifeedant activity (*10*), but recent investigations have disclosed powerful anticancer (*13*) and antiinflammatory (*14*) properties. In this context, cascarilla can provide sizable amounts of the densely functionalized and stereochemically rich clerodane diterpene **1**, a compound which can serve as starting material for the synthesis of libraries of derivatives to test against useful biological targets. Furthermore, the easy availability of cascarillin will also pave the way to the elucidation of the structure−activity relationship within bitter diterpenoids, one of the first classes of bitter nonalkaloidal compounds to be discovered.

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Supporting Information Available: Key ROESY correlations of eluterin A and key ROESY correlations of eluterin F. This material is available free of charge via the Internet at http://pubs.acs.org.

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