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DRIMANE SESQUITERPENE LACTONES FROM *PENIOPHORA POLYGONIA*

WILLIAM A. AYER* and LATCHEZAR S. TRIFONOV

Department of Chemistry, University of Alberta, Edmonton, Alberta, Canada T6G 2G2

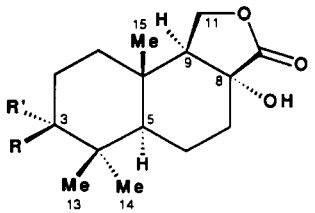
ABSTRACT.—Several new drimane sesquiterpenes, peniopholide [**1**], 3β-hydroxypeniopholide [**2**], 3α-hydroxypeniopholide [**3**], 3β-hydroxydihydroconfertifolin [**5**], 6β-hydroxycinnamolide [**8**], 6α-hydroxycinnamolide [**10**], and 7α-hydroxyconfertifolin [**11**], have been isolated from *Peniophora polygonia*, a fungus associated with aspen. The structures were established by spectroscopic methods. The known sesquiterpenes *cis*-dihydroconfertifolin [**4**], cinnamolide [**6**], and 3β-hydroxycinnamolide [**7**] were also isolated.

Peniophora polygonia (Pers.: Fr.) Bourd. & Galzin (=*Corticium polygonium*) (Peniophoraceae) is a wood-staining fungus often found on aspen. Recent experiments (1,2) have demonstrated a distinct antagonism between this fungus and *Phellinus tremulae*, one of the most common fungi causing trunk rot in aspen (3). In an attempt to determine the compounds responsible for this activity we have grown *Pe. polygonia* in liquid shake culture. We report herein the isolation and characterization of some sesquiterpenes produced when the fungus is cultured (23°, 49 days) on a medium of 10% V-8 juice containing 1% added glucose.

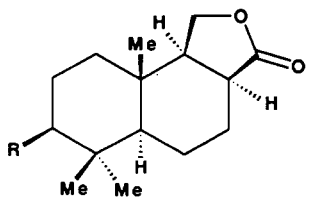
RESULTS AND DISCUSSION

The culture broth of *Pe. polygonia* was filtered, and the filtrate was concentrated and extracted with EtOAc. The crude extract was subjected to flash chromatography on Si gel. Pure components were obtained by crystallization and/or by further chromatography on preparative tlc plates.

Peniopholide [**1**], mp 119.5–120.5°, $[\alpha]^{21}D$ −5.2° (c=0.48, MeOH), has molecular formula $C_{15}H_{24}O_3$ as established by hreims. The presence of a saturated γ-lactone was indicated by strong absorption at 1767 cm^{-1} in the ir. The third oxygen is present as an OH group (broad band at 3100–3550 cm^{-1}). Peniopholide contains no olefinic carbons (^{13}C nmr) indicating that it is tricyclic. The ^1H-nmr spectrum showed three Me groups as singlets, which together with the characteristic high field signal at 0.98 ppm (H–5, dd, J=13.8, 2.0 Hz) suggeted a drimane sesquiterpenoid skeleton (4,5). The presence of the fragment CO-O-CH_2-CH was indicated by chemical shifts and the coupling pattern in the ^1H-nmr spectrum (see Table 1). The doublet in the ^1H-nmr spectrum for H-9 (coupling to only one of the CH_2O hydrogens, i.e., H-11α) places the lactone



1 R=R'=H
2 R=OH, R'=H
3 R=H, R'=OH



4 R=H
5 R=OH

TABLE 1. ^{1}H-nmr (500 MHz) Data of **1**, **2**, and **3**.

Proton	Compound					
	1	**2**			**3**	
	^{1}H δ	COSY	^{1}H δ	COSY	nOe	^{1}H δ
H-1ax	1.00 ddd, J=13.6, 3.8	1eq, 2ax, 2eq	1.16 ddd, J=12.5, 4.2	1eq, 2ax, 2eq, 15[a]	2eq, 3, 5, 9	1.51–1.6 m
H-1eq	ca. 1.65 m	1ax, 2ax, 2eq	ca. 1.70 m	1ax		1.41 ddd, J=13.0, 3.5
H-2ax	1.55 ddddd, J=13.5, 3.4	1ax, 1eq, 2eq 3ax, 3eq	ca. 1.65 m	1ax, 3		1.93 dddd, J=14.5, 3.5, 2.5
H-2eq	ca. 1.44 m	1ax, 1eq, 2ax 3ax, 3eq	ca. 1.70 m	1ax		1.5–1.6 m
H-3ax	1.18 ddd, J=14.0, 4.6	2ax, 2eq, 3eq	3.27 dd, J=11.0, 4.7	2ax, 2eq		—
H-3eq	ca. 1.44 m	2ax, 2eq, 3ax	—			3.48 dd, J=3.5, 2.5
H-5	0.98 dd, J=13.8, 2.0	6ax, 6eq	0.99 dd, J=12.0, 2.0	6ax, 6eq[a], 7eq	3, 7ax, 9	1.47 dd, J=12.5, 2.0
H-6ax	ca. 1.25 m	5, 6eq, 7ax, 7eq	ca. 1.35 m	5, 6eq, 7ax, 7eq	6eq, 7eq, 13, 15	1.29 dddd, J=13.0, 4.5
H-6eq	ca. 1.65 m	5, 6ax, 7eq	ca. 1.70 m	6ax, 7eq		1.5–1.6 m
H-7ax	ca. 1.65 m	6ax, 7eq	ca. 1.70 m	6ax, 7eq		1.68 ddd, J=13.0, 13.0, 6.5
H-7eq	ca. 2.50 m	6ax, 6eq, 7ax	ca. 2.50 m	5[a], 6ax, 6eq, 7ax, 9[a], 11β[a]	6ax, 6eq, 7ax	2.50 ddd, J=13.0, 5.0, 2.0
H-9	1.89 d, J=5.0	11α	1.88 d, J=5.2	5[a], 7eq[a], 11α, 11β[a], 15[a]	1ax, 5, 11α	2.01 d, J=5.2
H-11α	4.50 dd, J=9.5, 5.0	9, 11β	4.51 dd, J=9.3, 5.2	9, 11β		4.53 dd, J=9.5, 5.2
H-11β	4.17 d, J=9.5	11α	4.16 d, J=9.3	7eq[a], 9[a] 11α, 15[a]		4.18 d, J=9.5
H-13	0.82 s		0.79 s	14[a]	6ax[b]	0.85 s
H-14	0.92 s		1.05 s	13[a]	3, 5, 6eq	1.01 s
H-15	0.79 s		0.80 s	1ax[a], 9[a]	2ax, 6ax, 11β	0.82 s
OH	2.09 br s		ca. 2.0 br s			2.14 br s

[a]Correlation observed in long-range COSY only.
[b]The signal at 0.80 ppm is also irradiated.

carbonyl at C-12, not at C-11 as in some drimane lactones. The presence of six CH_2 groups, as indicated by the ^{13}C-nmr spectrum, left C-5 and C-8 as the only feasible positions for the OH group. Location of the OH group at C-8 is based on the unusually low-field chemical shift of H-11α (at 4.50 ppm) as well as on the multiplicity of H-9 (d, J=5.0) in the ^{1}H-nmr spectrum (5). In addition, this location accounts for the pronounced loss of $C_2H_3O_2$ in the eims spectrum of peniopholide. The 2D-COSY and especially the HMBC spectrum confirm the structure of peniopholide as that shown in **1**. Complete ^{1}H-nmr and ^{13}C-nmr assignments for **1** are shown in Tables 1 and 2.

3β-Hydroxypeniopholide [**2**], mp 204.5–206.0°, $[\alpha]^{21}D$ −3.1° (c=0.68, MeOH), has molecular formula $C_{15}H_{24}O_4$ (hreims). The absorption at 1765 cm^{-1} in the ir spectrum, the presence of a two-proton broad singlet at ca. 2.0 ppm in the ^{1}H-nmr spectrum (two OH groups), and the loss of $C_2H_3O_2$ observed in the eims spectrum strongly suggested that it has the same skeleton as **1** with one OH group at C-8 and one additional secondary OH group [the carbon bearing this OH appears at 78.7 ppm (d) in the ^{13}C-nmr spectrum]. The coupling constants of C*H*OH (dd, J=11.0, 4.7) as well as the nOe of the signal observed when the H-5 signal was irradiated revealed that this OH group is equatorial and is attached to C-1, C-3, or C-7. The coupling of this secondary carbon to the protons of both C-13 and C-14 revealed by the HMBC spectrum unequivocally located the second OH group at C-3. Full assignments of the ^{1}H-nmr and ^{13}C-nmr signals are listed in Tables 1 and 2.

3α-Hydroxypeniopholide [**3**], mp 178.0–179.5°, $[\alpha]^{21}D$ −34.8° (c=0.21, MeOH), is an isomer of **2**, as indicated by the hreims spectrum. The presence of a hydroxyl group at C-8 resulted in the loss of $C_2H_3O_2$ in the eims spectrum as in **1** and **2**. The coupling constants of C*H*OH (dd, J=3.5, 2.5) clearly show that the second hydroxyl group is axial. Its location at C-3 is based on the correlation between C-3 and H-13 and H-14 observed in the HMBC spectrum.

The cis configuration of the lactone ring in **1**, **2**, and **3** is demonstrated by the coupling of H-9 to only one of the hydrogen atoms at C-11, i.e., H-11α. Indeed, models show that a 90° dihedral angle H-9–C-9–C-11–H-11β is possible only when the lactone ring is cis-fused. This is in accordance with the observed coupling pattern of similar cis,

TABLE 2. ^{13}C-nmr Data (125 MHz) of **1–3**, **5–8**, **10**, and **11**.

Carbon	Compound								
	1	**2**	**3**	**5**	**6**	**7**	**8**	**10**	**11**
C-1	40.7	38.5	32.9	38.4	39.6[b]	37.3	41.7[a]	39.7[a]	35.6[a]
C-2	18.0	26.7	24.8	26.8	18.3	27.0	18.4	18.6	18.4
C-3	41.7	78.7	75.5	78.7	42.2[a]	78.7	44.8[a]	43.3[a]	41.5[a]
C-4	32.9	38.6	37.2	38.8	32.9	38.8	34.4[b]	40.1[b]	32.9[b]
C-5	51.4	50.7	44.6	51.0[a]	49.8	49.2	54.9[c]	58.1[c]	46.0
C-6	19.0	18.7	18.7	18.3	25.0	24.9	65.9	69.0	27.2
C-7	32.1	31.9	32.1	22.4	136.2	136.2	135.6	136.0	60.3
C-8	73.3	73.1	73.4	37.4	127.3	127.2	128.1	129.0	125.0
C-9	56.4	56.3	56.2	49.9[a]	50.9	50.8	52.0[c]	50.2[c]	173.9[c]
C-10	35.7	35.5	35.5	35.3	34.3	34.2	34.2[b]	33.4[b]	37.2[b]
C-11	67.4	67.3	67.4	67.6	67.3	67.0	67.5	67.3	68.5
C-12	177.6	177.5	177.6	178.8	169.8	170.0	170.0	169.9	174.2[c]
C-13	22.0	15.7	22.4	15.6	21.4	14.9	24.9	22.2	21.5
C-14	33.6	28.5	28.5	28.4	33.2	27.9	32.7	36.1	33.0
C-15	14.5	14.6	14.5	14.5	13.5	13.5	15.7	14.6	19.5

[a–c]Signals with the same superscript in the same column are interchangeable.

but not trans, lactones, in drimane sesquiterpenes (5). The nOe observed at H-1 ax and H-5 on irradiation of H-9 shows the β configuration of C-11, and thus the β configuration of C-12.

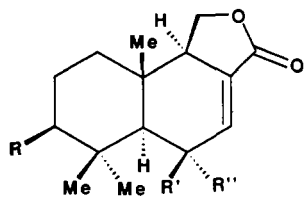
The absolute configuration of **1**, **2**, and **3** was determined by using the CD empirical rule for α-hydroxyl-γ-lactones (6). The negative Cotton effect shown by **1** ($\Delta\epsilon$ −1.4, c=0.48, MeOH, at 224 nm), **2** ($\Delta\epsilon$ −1.2, c=0.68, MeOH, at 226 nm), and **3** ($\Delta\epsilon$ −1.4, c=0.21, MeOH, at 224 nm) is indicative of the C-8 *R* and C-9 *S* absolute stereochemistry shown. The absolute configuration at C-3 of **2** was shown by the Horeau method (7) to be *S*, confirming the absolute configuration of **2**, which is identical with that of the bicyclic sesquiterpene drimane-8,11-diol (8,9); i.e., **1–3** possess "normal" (steroidal) absolute stereochemistry (10).

cis-Dihydroconfertifolin [**4**] was not isolated in pure form but as a mixture with cinnamolide (vide infra). Its identity as **4** is based on ir, ms, ^{1}H-nmr (500 MHz), and 2D-COSY spectra of this mixture. Compound **4** was previously isolated from a liverwort (11).

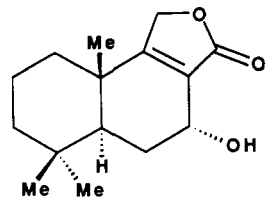
3β-Hydroxydihydroconfertifolin [**5**], mp 176.0–178.0°, $[\alpha]^{21}D$ −4.3° (c=0.37, MeOH), exhibited spectral properties similar to those of **4**, except that it also shows characteristics of a secondary alcohol. Assignment of the hydroxyl group as equatorial at C-3 is based on the coupling constants of C*H*OH (dd, J=11.5, 4.5) and on the similarity to compound **2**, the latter differing only by the presence of an additional hydroxyl group at C-8. The possibility that this OH group is located at C-1 is ruled out since such a group would cause a significant downfield shift of the signal of H-11β (5). Indeed, the ^{13}C-nmr signals of **5** are almost identical with those of **2**, except for C-7, C-8, and C-9 (see Table 2). The absolute configuration of **5** ($\Delta\epsilon$ +1.5, c=0.37, MeOH, at 216 nm) corresponds to that of the parent compound *cis*-dihydroconfertifolin [**4**] ($\Delta\epsilon$ +0.6, MeOH) (12).

Cinnamolide [**6**] shows spectral characteristics, including optical rotation, identical with those reported. This sesquiterpene has been isolated previously from *Cinnamosma fragrans* Baillon growing in Madagascar (4) and from *Warburgia ugandensis* and *Warburgia stuhlmanni* (13).

In addition to cinnamolide [**6**], three hydroxy derivatives of this sesquiterpene were isolated, namely 3β- [**7**], 6β- [**8**], and 6α-hydroxycinnamolide [**10**]. Compound **7**, mp 164.5–167.0°, $[\alpha]^{21}D$ −22.6° (c=0.42, MeOH), shows spectral data (^{1}H-nmr, ir, ms) identical with those of the recently reported "cinnamolide-3β-ol", except for the optical rotation, which has been reported to be $[\alpha]D$ +37° (MeOH) (14). The cd spectrum of **7** [$\Delta\epsilon$ +3.4 (250), −5.4 (ca. 210), c=0.042, MeOH] is very similar to that of the parent compound cinnamolide [**6**] [$\Delta\epsilon$ +4.0 (249), −6.3 (ca. 210), c=0.028, MeOH], which confirms its absolute stereochemistry as that of a "normal" drimane sesquiterpene (10).



6 R=R′=R″=H
7 R=OH, R′=R″=H
8 R=R″=H, R′=OH
9 R=R″=H, R′=OAc
10 R=R′=H, R″=OH



11

6β-Hydroxycinnamolide [**8**], mp 190.0–191.0°, $[\alpha]^{21}D$ −162.7° (c=0.22, MeOH), exhibits a ^{1}H-nmr spectrum similar to that of **6**, except that one of the hydrogen atoms at C-6 (as revealed by decoupling experiments) is replaced by a hydroxyl group. The β orientation of this hydroxyl group is apparent from the lack of a large coupling of C*H*OH to H-5. Final proof of the structure and absolute configuration of **8** was accomplished by acetylation (Ac_2O/pyridine), leading to compound **9**, identical (mp, $[\alpha]D$, ^{1}H nmr, and hreims) with the sesquiterpene bemarivolide of known absolute stereochemistry (15).

6α-Hydroxycinnamolide [**10**], mp 142.0–143.5°, $[\alpha]^{21}D$ +150.0° (c=0.15, MeOH), displays signals in the ^{1}H-nmr spectrum very similar to those of **8**. However, the coupling constants of C*H*OH (dt, J=9.3, 3.4) are compatible only with an α-hydroxyl group at C-6. This accounts for the downfield shift of H-5 (at 1.36 ppm) and of C-14 (at 36.1 ppm), relative to those of **8** (1.22 ppm and 32.7 ppm).

7α-Hydroxyconfertifolin [**11**], mp 216.0–218.0°, $[\alpha]^{21}D$ +34.4° (c=0.27, MeOH) is isomeric with **8** and **10**. The uv spectrum [214 nm (9900)], ir spectrum (1713 cm^{-1}), unsaturation number, and lack of olefinic hydrogen signals in the ^{1}H-nmr spectrum together with the presence of signals for two sp^2 carbon atoms in the ^{13}C-nmr spectrum (Table 2) show the presence of the unsaturated lactone ring at C-8–C-9–C-11–C-12. The location of a secondary hydroxyl group at C-7 is indicated by the chemical shift (4.59 ppm) and the coupling pattern (br d, J=4.6) of C*H*OH. To distinguish between the two feasible structures, one with the lactone carbonyl group at C-11, the other at C-12, the ^{1}H-nmr spectrum of **11** was compared with that of the known sesquiterpene futronolide [to which, erroneously, structure **11** was initially ascribed (10,16)]. Unlike futronolide, which possesses a carbonyl group at C-11 that strongly deshields H-1β (at ca. 2.5 ppm), **11** does not show signals in this region.

Preliminary testing shows that only peniopholide [1] has antifungal activity against *Ph. tremulae*.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Melting points are uncorrected and were determined on a Thomas model 40 melting point apparatus. Ft-ir spectra were recorded on a Nicolet 7199 FTIR interferometer. Uv spectra were obtained on a Hewlett Packard 8450A Diode Array spectrophotometer, and optical rotations were determined with a Perkin Elmer 241 polarimeter. Cd spectra were recorded on a Jasco Optical Rotatory Dispersion SS-20-2 recorder. Hreims were recorded on an AEI MS-50 mass spectrometer. Nmr spectra (^{1}H and ^{13}C) were obtained on Bruker WM-360 or Varian Unity 500 multinuclear spectrometers. Chemical shifts were referenced to residual hydrogen (7.26 ppm) or carbon (77.0 ppm) absorption of $CDCl_3$. Multiplicities of ^{13}C-nmr signals were determined from APT spectra. Flash chromatography was performed on Si gel 230–400 mesh, General Intermediates of Canada. Preparative tlc was performed on E. Merck precoated 20×20 glass plates of Si gel 60 F-254. Analytical tlc was carried out on cut sections of E. Merck precoated aluminium sheets of Si gel 60 F 254. Uv-active compounds were detected under a uv lamp (254 nm). Tlc plates were further visualized using iodine vapor or phosphomolybdic acid. All solvents were distilled prior to use. Skellysolve B (SKB) refers to Skelly Oil Company petroleum ether, bp 62–70°.

ISOLATION OF METABOLITES.—Cultures of *Pe. polygonia* (strain NOF 1494, UAMH 7006) were obtained from Dr. Y. Hiratsuka, Forestry Canada, Northern Forestry Centre, Edmonton. Eleven 2-liter Erlenmeyer flasks each containing 1 liter of V-8 juice liquid medium (10 g of glucose, 100 ml of V-8 juice filtered through Celite, and 900 ml of distilled H_2O) were inoculated with ca. 10 ml of a mycelial suspension of *Pe. polygonia* and were shaken at 23° for 49 days. The culture broth (11 liters) was filtered through cheesecloth, and the filtrate was concentrated under reduced pressure to 2 liters and extracted with EtOAc (3×800 ml). The organic extract was dried over $MgSO_4$ and the solvent removed to afford a crude extract (920 mg of red oil). This oil was subjected to flash chromatography on Si gel using gradient elution: SKB-EtOAc (90:10) (500 ml), SKB-EtOAc (84:16) (500 ml), SKB-EtOAc-CH_2Cl_2-MeOH (63:16:16:5) (500 ml), SKB-EtOAc-CH_2Cl_2-MeOH (36:36:18:10) (600 ml), and $CHCl_3$-MeOH (90:10) (600 ml), with fractions of 20 ml being collected.

Fractions 23–48 (24 mg) were applied to 2 preparative tlc plates and developed three times with SKB-

EtOAc-CH_2Cl_2-MeOH (84:7:7:2). The material from the strongly uv-active zone was eluted with CH_2Cl_2-MeOH (10:1) and crystallized from SKB to afford cinnamolide [**6**] (3 mg). The weakly uv-active zone at a lower R_f afforded a mixture of **6** and *cis*-dihydroconfertifolin [**5**] (2 mg).

Fractions 49–52 (15 mg) gave peniopholide [**1**] (4.2 mg) on crystallization from SKB.

Fractions 64–90 (40 mg) were purified by preparative tlc, eluent SKB-EtOAc-CH_2Cl_2-MeOH (84:7:7:2) (fivefold development). The uv active zone at R_f 0.42 was eluted with CH_2Cl_2-MeOH (10:1), and the crude material (6 mg) was crystallized from SKB/Et_2O to give 6β-hydroxycinnamolide [**8**] (2.7 mg). The uv-active zone at R_f 0.20 afforded, after crystallization from SKB/Et_2O, pure 6α-hydroxycinnamolide [**10**] (1.5 mg).

Fractions 112–120 (140 mg) were crystallized from CH_2Cl_2 to give 3β-hydroxypeniopholide [**2**] (43 mg).

Fractions 95–112 (332 mg) were subjected to flash chromatography on Si gel using gradient elution: SKB-EtOAc (84:16) (500 ml), (80:20) (500 ml), (67:33) (2 liters), and (50:50) (1 liter). Fractions 45–55 (17 mg) afforded 7α-hydroxyconfertifolin [**11**] (2.7 mg), after crystallization from SKB/CH_2Cl_2. Fractions 92–120 (80 mg) were crystallized from SKB/Et_2O to give a mixture of 3β-hydroxydihydroconfertifolin [**5**] and 3β-hydroxycinnamolide [**7**] in a ratio of 3:2 (24 mg). This mixture was separated by preparative tlc, eluent SKB-EtOAc (75:25) (threefold development). The uv-active zone at R_f 0.38 was treated as above to give crude product (11.4 mg), which was crystallized from SKB/Et_2O to yield pure 3β-hydroxycinnamolide [**7**] (4.4 mg). The non-uv-active zone at R_f 0.43 was treated in the same way to afford pure 3β-hydroxydihydroconfertifolin [**5**] (7.1 mg). Fractions 150–165 (10 mg) were subjected to preparative tlc with toluene-EtOAc-MeOH (80:15:5) (threefold development). The non-uv-active zone at R_f 0.42 was eluted with CH_2Cl_2-MeOH (10:1), and the crude product (3.2 mg) was crystallized from SKB/Et_2O to give pure 3α-hydroxypeniopholide [**3**] (2.2 mg).

Peniopholide [**1**].—Mp 119.5–120.5°; $[\alpha]^{21}D$ −5.2° (c=0.48, MeOH); Δε −1.4 (224 nm) (c=0.48, MeOH); ir ν max ($CHCl_3$) cm^{-1} 3500–3100 br, 2983, 2947, 2927, 2868, 2845, 1767; uv λ max (MeOH, ε) 224 nm (22); hreims *m/z* $[M]^+$ +252.1725(4) ($C_{15}H_{24}O_3$ requires 252.1725) 237(6) 234(2), 219(25), 208(29), $[M-C_2H_3O_2]^+$ 193(60), 191(11), 179(10), 166(10), 138(16), 137(19), 136(28), 125(63), 124(33), 123(59), 121(16), 111(16), 109(29), 107(14), 105(12), 97(18), 96(16), 95(43), 93(23), 91(18), 86(15), 82(44), 81(41), 69(100); ^1H nmr see Table 1; ^{13}C nmr see Table 2. HMBC (C/H's) 1/2ax, 2eq, 3ax, 3eq, 9, 15; 2/1ax, 1eq, 3ax, 3eq; 3/1ax, 1eq, 2ax, 2eq, 5, 13, 14; 4/2ax, 3ax, 3eq, 13, 14; 5/3ax, 3eq, 6ax, 6eq, 7ax, 7eq, 13, 14, 15; 6/7ax, 7eq; 7/6ax, 6eq; 8/7ax, 7eq, 9, 11β; 9/1ax, 5, 7eq, 11α, 11β; 10/1ax, 1eq, 5, 6ax, 6eq, 9, 11α, 11β, 15; 11/9; 12/7ax, 7eq, 9, 11β; 13/3ax, 14; 14/3ax, 13; 15/1ax, 5, 9.

3β-Hydroxypeniopholide [**2**].—Mp 204.5–206.0° $[\alpha]^{21}D$ −3.1° (c=0.68, MeOH); cd Δε −1.2 (226 nm) (c=0.68, MeOH); ir ν max ($CHCl_3$) cm^{-1} 3550–3100 br, 2966, 2949, 2935, 2869, 2856, 1765; uv λ max (MeOH, ε) 218 nmr (25); hreims *m/z* $[M]^+$ 268.1672(5) ($C_{15}H_{24}O_4$ requires 268.1674), 250(4), 232(2), 224(17), 222(15), 217(12), 210(14), $[M-C_2H_3O_2]^+$ 209(100), 207(8), 206(57), 204(10), 195(11), 193(21), 191(56), 189(16), 188(12), 177(14), 173(11), 168(7), 165(14), 164(14), 163(26), 154(15), 151(16), 149(25), 147(20), 139(40), 137(58), 135(39), 134(54), 124(26), 123(58), 122(26), 121(81), 119(32), 111(22), 110(25), 109(29), 107(49), 105(27), 97(24), 95(48), 93(62), 91(32), 83(26), 82(18), 81(60), 79(37), 77(23), 71(29), 70(25). ^1H nmr see Table 1; ^{13}C nmr see Table 2. HMBC (C/H's) 1/3, 9, 15; 2/1ax, leq; 3/1ax, 2ax, 2eq, 13, 14; 4/2ax, 2eq, 3, 5, 13, 14; 5/6ax, 7ax, 7eq, 9, 13, 14, 15; 6/5, 7ax, 7eq; 7/5, 6ax, 6eq; 8/7ax, 7eq, 9, 11β; 9/1ax, 5, 7eq, 11α,11β; 10/5, 9, 11α; 11/-; 12/7ax, 7eq, 9, 11β; 13/3, 5, 14; 14/3, 5, 13; 15/1ax, 5, 9.

3α-Hydroxypeniopholide [**3**].—Mp 178.0–179.5° $[\alpha]^{21}D$ −34.8° (c=0.21, MeOH); cd Δε −1.4 (224 nm) (c=0.21, MeOH); ir ν max ($CHCl_3$) cm^{-1} 3600–3100 br, 2950, 2926, 2860, 1763; uv λ max (MeOH, ε) 220 nm (23); hreims *m/z* $[M]^+$ 268.1675(12) ($C_{15}H_{24}O_4$ requires 268.1674), 250(20), 235(18), 212(16), $[M-C_2H_3O_2]^+$ 209(30), 206(24), 194(14), 191(54), 189(18), 177(16), 163(13), 149(19), 147(14), 137(75), 136(100), 134(64), 123(31), 121(92), 119(31), 107(44), 95(42), 93(59), 83(17), 82(16), 81(55), 79(36), 71(31), 69(66), 67(51); ^1H nmr see Table 1; ^{13}C nmr see Table 2. ^1H-nmr decouplings: irradiation at 3.48 ppm gave 1.93 (td), and ca 1.6 (changed). HMBC (C/H's): 1/3, 9, 15; 2/1ax; 3/1ax, leq, 13, 14; 4/5, 13, 14; 5/3, 7eq, 9, 13, 14, 15; 6/7ax; 7/5, 6ax, 6eq, 9; 8/7ax, 7eq, 9, 11β; 9/5, 7eq, 11β; 10/5, 9, 11α, 11β; 11/-; 12/not attempted; 13/5, 14; 14/5, 13; 15/1ax, 5, 9.

cis-*Dihydroconfertifolin* [**4**].—Mixture with **6** in a ratio of 1:1. Ir ν max ($CHCl_3$) cm^{-1} 1770; hreims *m/z* $[M]^+$ 236.1776 (24) ($C_{15}H_{24}O_2$ requires 236.1776); ^1H nmr (500 NHz, $CHCl_3$, signals after subtraction of the signals of **6**) 0.82, 0.85, 0.89 (each 3H, s, 3×Me), 1.10–1.25 (2H, m), 1.35–1.72 (8H, m), 2.08 (1H, dd, J=8.0, 5.4, H-9), 2.32 (1H, br dd, J=13.5, 5.5, H-7eq), 2.55 (1H, t, 8.0, H-8), 4.11 (1H, dd, J=9.7, 5.4, H-11α), 4.22 (1H, d, J=9.7, H-11β).

3β-Hydroxydihydroconfertifolin [**5**].—Mp 176.0–178.0°; $[\alpha]^{21}D$ −4.3° (*c*=0.37, MeOH); cd Δε +1.5 (216 nm) (*c*=0.37, MeOH); ir ν max ($CHCl_3$) cm^{-1} 3580–3350 br, 2980, 2942, 2925, 2872, 2860, 1740; uv λ max (MeOH, ε) ca. 215 nm sh (90); hreims *m/z* $[M]^+$ 252.1727(16) ($C_{15}H_{24}O_3$ requires 252.1725), 237(9), 234(81), 219(54), 195(41), 193 (17), 191(22), 173(16), 153(82), 150(67), 149(38), 148(12), 147(14), 139(39), 137(21), 136(23), 135(38), 133(18), 125(17), 123(45), 122(26), 121(98), 109(42), 107(99), 95(57), 93(64), 91(41), 86(100), 85(52), 81(78), 79(54), 69(56), 67(67), 55(68); ^1H nmr (500 MHz, $CDCl_3$) 0.80 (3H, s, H-13), 0.83 (1H, dd, *J*=12.0, 2.0, H-5), 0.85 (3H, s, H-15), 1.02 (3H, s, H-14). 1.09 (1H, qd, *J*=13.0, 4.0, H-2ax), 1.34 (1H, td, *J*=12.5, 5.0, H-1ax), 1.38 (1H, dd, *J*=12.5, 5.0), 1.54–1.69 (4H, m), 1.75 (1H, dt, *J*=13.0, 3.5, H-1eq), 2.07 (1H, dd, *J*=8.0, 5.2, H-9), 2.35 (1H, br dd, *J*=14.0, 5.0, H-7eq), 2.56 (1H, t, *J*=8.0, H-8), 3.25 (1H, dd, *J*=11.5, 4.5, H-3), 4.13 (1H, dd, *J*=9.8, 5.2, H-11α), 4.22 (1H, d, *J*=9.8, H-11β); ^{13}C nmr see Table 2.

Cinnamolide [**6**].—Mp 121.0–124.0° [lit. (4) mp 126°]; $[\alpha]^{21}D$ −22.2° (*c*=0.18, $CHCl_3$) [lit. (4) $[\alpha]D$ −29° (*c*=1, $CHCl_3$)]; cd Δε +4.0 (249 nm), −6.3 (ca. 210 nm) (*c*=0.028, MeOH); ir ν max ($CHCl_3$) cm^{-1} 1763; uv λ max (MeOH, ε) 222 nm (7360); ^1H nmr (360 MHz, $CHCl_3$) 0.82, 0.92, 0.95 (each 3H, s, 3×Me), 1.15–1.30 (2H, m, H-1ax, H-3ax), 1.40 (1H, dd, *J*=11.6, 5.4, H-5), 1.45–1.62 (4H, m, H-1eq, H-2, H-3eq), 2.12 (1H, dddd, *J*=20.0, 11.6, 4.6, 3.5, H-6ax), 2.42 (1H, ddt, *J*=20.0, 5.1, 3.5, H-6eq), 2.78–2.85 (1H, m, H-9), 4.04 (1H, t, *J*=9.1, H-11β), 4.38 (1H, t, *J*=9.1, H-11α), 6.88 (1H, dt, *J*=3.5, 3.5, H-7); ^{13}C nmr see Table 2.

3β-Hydroxycinnamolide [**7**].—Mp 164.5–167.0°; $[\alpha]^{21}D$ −22.6° (c=0.42, MeOH); cd Δε +3.4 (250), −5.4 (ca. 210 nm) (*c*=0.042, MeOH); ir ν max ($CHCl_3$) cm^{-1} 3462, 3065, 2930, 2920, 2900, 2855, 1763, 1734, 1683: uv λmax (MeOH, ε) 222 nm (7000); hreims *m/z* $[M]^+$ 250.1573(0.6), $C_{15}H_{22}O_3$ requires 250.1568), 232(5), 217(3), 149(7), 140(10), 139(8), 125(16), 122(87), 111(10), 107(15), 105(7), 97(9), 96(100) (C_7H_{12}), 91(13), 81(10), 79(8); ^1H nmr (360 MHz, $CDCl_3$) 0.81, 0.92, 1.05 (each 3H, s, 3×Me), 1.38 (1H, dd, *J*=11.5, 5.5, H-5), 1.20–1.40 (2H, m), 1.45–1.70 (3H, m), 2.20 (1H, dddd, *J*=20.2, 11.5, 5.0, 3.5, H-6ax), 2.44 (1H, ddt, *J*=20.2, 5.5, 3.5, H-6eq), 2.79 (1H, m, H-9), 3.31 (1H, dd, *J*=10.5, 4.5, H-3), 4.05 (1H, t, *J*=9.1, H-11β), 4.39 (1H, t, *J*=9.1, H-11α), 6.90 (1H, dt, *J*=3.5, 3.5, H-7); ^{13}C nmr see Table 2.

6β-Hydroxycinnamolide [**8**].—Mp 190.0–191.0°; $[\alpha]^{21}D$ −162.7° (*c*=0.22, MeOH); cd Δε −6.3 (232) (*c*=0.022, MeOH); ir ν max ($CDCl_3$) cm^{-1} 3504, 2942, 2925, 2864, 1742, 1694; uv λ max (MeOH, ε) 220 nm (7550); hreims *m/z* $[M]^+$ 250.1569 (12) ($C_{15}H_{22}O_3$ requires 250.1568), 235(3), 232(5), 217(5), 189(3), 180(5), 153(8), 127(21), 126(27), 124(49), 109(100) (C_8H_{13}), 91(14), 81(16), 77(11), 69(19), 67(11), 55(15); ^1H nmr (360 MHz, $CDCl_3$) 1.06, 1.12, 1.35 (each 3H, s, 3×Me), 1.22 (1H, d, *J*=5.8, H-5), 1.15–1.70 (7H, m), 2.70 (1H, tdd, *J*=9.3, 3.4, 2.7, H-9), 4.11 (1H, t, *J*=9.3, H-11β), 4.44 (1H, t, *J*=9.3, H-11α), 4.71 (1H, br s, H-6), 6.81 (1H, t, *J*=3.6, H-7). Decouplings: irradiation at 2.70 ppm gave 4.11 (d), 4.44 (d), 4.71 (t), and 6.81 (d); irradiation at 4.71 ppm gave 1.22 (s), 2.70 (td), and 6.81 (d). ^{13}C nmr see Table 2.

Benarivolide [**9**].—Compound **8** (1.8 mg) in Ac_2O (0.1 ml), and pyridine (0.1 ml) was kept overnight at room temperature. Toluene (0.4 ml) was added and the solvent removed under reduced pressure. The residue was purified by preparative tlc using SKB-EtOAc-CH_2Cl_2-MeOH (84:7:7:2). The crude product was crystallized from SKB to afford **9** (1.0 mg): mp 129.0–132.0° [lit. (15) mp 137–138°]; $[\alpha]^{21}D$ −162° (c=0.1, $CDCl_3$) [lit. (15) $[\alpha]D$ −258° (*c*=1, $CHCl_3$)]; ir ν max ($CHCl_3$) cm^{-1} 2975, 2930, 2870, 2850, 1760, 1737; hreims *m/z* $[M]^+$ 292.1672(13) ($C_{17}H_{24}O_4$ requires 292.1674), 250(69), 232(24), 217(17), 189(10), 176(29), 169(25), 164(20), 149(25), 131(23), 124(34), 109(100) (C_8H_{13}), 91(23), 69(21); ^1H nmr (360 MHz, $CDCl_3$) 1.02, 1.06, 1.16 (each 3H, s, 3×Me), 1.15–1.70 (7H, m), 2.08 (3H, s, OAc), 2.74 (1H, tt, *J*=9.1, 3.6, H-9), 4.12 (1H, t, *J*=9.1, H-11β), 4.45 (1H, t, *J*=9.1, H-11α), 5.82 (1H, br s, H-6), 6.71 (1H, t, *J*=3.6, H-7).

6α-Hydroxycinnamolide [**10**].—Mp 142.0–143.5° $[\alpha]^{21}D$ +150.0° (*c*=0.15, MeOH); cd Δε +10.5 (258), −11.4 (ca. 210) (*c*=0.015, MeOH); ir ν max ($CDCl_3$) cm^{-1} 3540–3150 br, 2924, 2907, 2870, 1762, 1751; uv λ max (MeOH, ε) 212 nm, (8800); hreims *m/z* $[M]^+$ 250.1568(12) ($C_{15}H_{22}O_3$ requires 250.1568), 235(6), 232(6), 217(6), 189(8), 180(12), 176(9), 153(17), 149(6), 131(14), 126(38), 124(37), 109(100) (C_8H_{13}), 105(11), 98(15), 95(12), 93(11), 91(21), 81(26), 79(16), 77(16); ^1H nmr (360 MHz, $CDCl_3$) 0.86, 1.07, 1.17 (each 3H, s, 3×Me), 1.2–1.8 (7H, m), 1.36 (1H, d, *J*=9.3, H-5), 2.92 (1H, tt, *J*=9.1, 3.4, H-9), 4.04 (1H, t, *J*=9.1, H-11β), 4.39 (1H, t, *J*=9.1, H11α), 4.50 (1H, dt, *J*=9.3, 3.4, H-6), 6.73 (1H, t, *J*=3.4, H-7); ^{13}C nmr see Table 2.

7α-Hydroxyconfertifolin [**11**].—Mp 216.0–218.0°; $[\alpha]^{21}D$ +34.4° (*c*=0.27, MeOH); cd Δε +1.1 (244), −3.1 (220) (*c*=0.027, MeOH); ir ν max ($CHCl_3$) cm^{-1} 3457, 2926, 1713; uv λ max (MeOH, ε) 214 nm (9900); hreims *m/z* $[M]^+$ 250.1566(100) ($C_{15}H_{22}O_3$ requires 250.1568), 235(6), 232(17), 217(32), 206(21), 191(8), 189(10), 176(14), 173(10), 163(12), 161(15), 153(20), 149(13), 135(12), 131(14),

127(45), 123(16), 121(12), 109(35), 105(20), 95(14), 91(31), 81(20), 79(22), 69(30), 67(22); ^{1}H nmr (360 MHz, $CDCl_3$) 0.93, 0.99, 1.14 (each 3H, s, 3×Me), 1.28 (1H, td, J=13.0, 4.0, H-1ax or H-3ax), 1.42 (1H, td, J=12.1, 3.5, H-3ax or H-1ax), 1.45–1.75 (5H, m), 1.79 (1H, td, J=13.5, 5.2, H-6ax), 1.99 (1H, br d, J=13.5, H-5), 2.19 (1H, br s, OH), 4.59 (1H, br d, J=4.6, H-7), 4.72 (1H, d, J=17.0, H-11α), 4.77 (1H, dd, J=17.0, 1.5, H-11β). Irradiation at 4.59 gave 1.79 (t); irradiation at 1.79 gave 1.99 (br s) and 4.59 (br s); ^{13}C nmr see Table 2.

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