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AFRICANE- AND MONOCYCLOFARNESANE-TYPE SESQUITERPENOIDS FROM THE LIVERWORT *PORELLA SUBOBTUSA*

FUMIHIRO NAGASHIMA, HIROMI IZUMO, ATSUSHI ISHIMARU, SHIORI MOMASAKI, MASAO TOYOTA, TOSHIHIRO HASHIMOTO and YOSHINORI ASAKAWA

Faculty of Pharmaceutical Sciences, Tokushima Bunri University, Yamashiro-cho, Tokushima 770, Japan

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Key Word Index—*Porella subobtusa*; Porellaceae; Hepaticae; 14-acetoxycaespitenone; swartzianin A; secoswartzianin A; dehydro-β-monocyclonerolidol; 8-hydroxy-9-methoxy-β-monocyclonerolidol; α-santalene; α-santalan-12(*S*),13-diol; africane-type; monocyclofarnesane-type; santalane-type; sesquiterpenoid.

Abstract—A new africane- and two monocyclofarnesane-type sesquiterpenoids have been isolated from the liverwort *Porella subobtusa*, along with five known sesquiterpenoids. The configuration of the hydroxyl group at C-12 of α-santalane-12,13-diol has been revised to *S*. Their structures were determined by extensive NMR techniques and chemical transformation. Copyright © 1996 Elsevier Science Ltd

INTRODUCTION

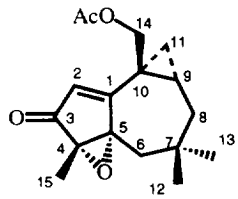
Porella species including *P. subobtusa* produce various sesquiterpenoids and are chemically divided into two types [1, 2]. One of which contains the intense pungent sesquiterpene dialdehyde, polygodial, and the other contains non-pungent substances [1, 2]. The chemical constituents of *P. subobtusa* belonging to the non-pungent group have not been reported yet. We report the isolation and characterization of a new africane-type (**1**) and two new monocyclofarnesane-type sesquiterpenoids (**2** and **3**), together with the known santalane- (**4** and **5**) and africane-type sesquiterpenoids (**6**–**8**). Furthermore, the configuration of the hydroxyl group at C-12 of **5** has been revised on the basis of the CD spectrum and modified Mosher's method.

RESULTS AND DISCUSSION

The CC on the ether extract of *P. subobtusa* yielded a new africane-type named 14-acetoxycaespitenone (**1**), two new monocyclofarnesane-type sesquiterpenoids, named dehydro-β-monocyclonerolidol (**2**) and 8-hydroxy-9-methoxy-β-monocyclonerolidol (**3**), together with two known santalan-type, α-santelane (**4**) [3], α-santalane-12(*S*), 13-diol (**5**) [4], three africane-type sesquiterpenoids, swartzianin A (**6**) [5], caespitenone (**7**) [6, 7] and secoswartzianin A (**8**) [8]. The known compounds were identified by the comparison of the spectral data of the authentic compounds and reference data.

The EI-mass spectrum of **1** showed *m/z* 290 $[M]^+$ and its molecular formula, $C_{17}H_{22}O_4$ (analyt 290.1511), was determined from the HR-mass spectrum. The IR spectrum showed the presence of carbonyl groups (1750, 1720 cm^{-1}). The ^1H NMR (Table 1) and ^{13}C NMR (Table 2) spectra of **1** closely resembled those of caespitenone (**7**) [6, 7], except for the presence of an acetoxyl group, indicating that **1** might be caespitenone (**7**) [6, 7] with the acetoxyl group at C-12, C-13, C-14 or C-15. The presence of the acetoxyl group at C-14 was confirmed by the ^1H–^1H and ^{13}C–^1H COSY (Fig. 1) and HMBC spectra (Fig. 2). Thus, the structure of **1** was cleared to be 14-acetoxycaespitenone. As no useful information about the stereochemistry of **1** was obtained by the difference NOE experiment, the determination of the stereostructure was carried out by the reported manner for the stereochemistry of **7** [6]. The reduction of **1** by Miyashita's reaction [9] afforded a monoalcohol **10**. In the NOESY spectrum of **10** in DMSO-d_6 solution, NOEs were observed between the hydroxyl group and H-11α, H-8α, H-6α and H-4α, respectively. Furthermore, in the ^1H NMR spectrum of **10** measured in pyridine-d_5 solution, the chemical shifts at H-4α, 6α, 8α and 11α shifted to downfield in comparison with those measured in $CDCl_3$ solution. Thus, the stereochemistry of 14-acetoxycaespitenone was depicted as shown in **1**. The absolute configuration of **1** was determined by the CD spectrum which exhibited the negative Cotton effect ($\Delta\varepsilon_{327} - 1.45$, $\Delta\varepsilon_{250} - 2.73$) as observed in caespitenone (**7**) [6, 7]. From the above evidence, the absolute configuration of 14-acetoxycaespitenone was established to be **1**.

The GC-mass spectra of **2** showed *m/z* 204 $[M]^+$ and its IR spectrum contained neither hydroxyl nor carbonyl absorption bands. The ^1H NMR (Table 1) and ^{13}C NMR (Table 2) spectra showed the presence of two



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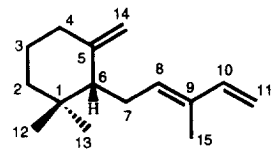

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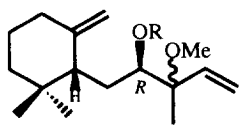
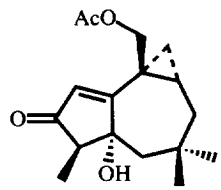


3 R=H
12 R=Ac
13a R=(*S*)-MTPA
13b R=(*R*)-MTPA



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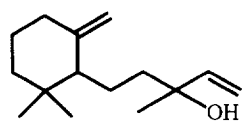

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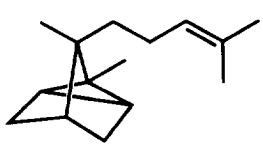

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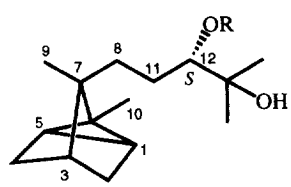
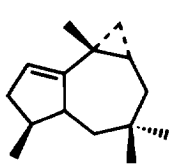


5 R=H
15a R=(R)-MTPA
15b R=(S)-MTPA



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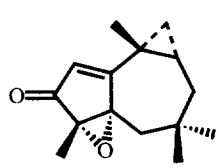

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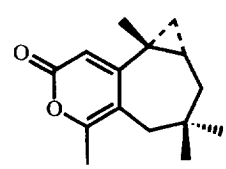

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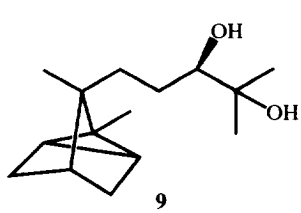

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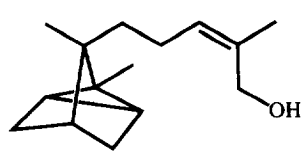

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Table 1. ^{1}H NMR spectral data of compounds **1**–**3** and **10** ($CDCl_3$, 400 MHz)

H	**1**	**2**	**3**	**10***
2	6.01 *s*	1.28 *m* 1.47 *m*	1.22 *m* 1.38–1.48 *m*	6.03 *s*
3		1.53, 2H, *m*	1.53 2H, *m*	
4		1.99 *m* 2.13 *m*	2.06 *dt*, *J* = 13.2, 4.4 Hz 2.21 *m*	2.51 *m*
6	2.31 *d*, *J* = 14.7 Hz, α 1.46 *d*, *J* = 14.7 Hz, β	1.83 *dd*, *J* = 11.2, 3.9 Hz	1.88 *dd*, *J* = 9.8, 3.9 Hz	1.58 *d*, *J* = 14.4 Hz, α 0.89 *d*, *J* = 14.4 Hz, β
7		2.20 *m* 2.35 *ddd*, *J* = 15.6, 6.8, 3.9 Hz	1.38–1.48 *m* 1.80 *dt*, *J* = 14.7, 3.4 Hz	
8	1.98 *dd*, *J* = 14.2, 3.4 Hz, α 1.22–1.32 *m*, β	5.40 *t*, *J* = 6.8 Hz	3.44 *dt*, *J* = 7.3, 2.9 Hz	2.21 *dd*, *J* = 14.2, 7.1 Hz, α 1.77 *dd*, *J* = 14.2, 8.8 Hz, β
9	1.36 *m*			1.51 *dddd*, *J* = 8.8, 8.8, 7.1, 7.1 Hz
10		6.34 *dd*, *J* = 17.6, 10.7 Hz	5.85 *dd*, *J* = 17.6, 10.7 Hz	
11	0.68 *t*, *J* = 4.9 Hz 1.22–1.32 *m*	4.87 *d*, *J* = 10.7 Hz 5.03 *d*, *J* = 17.6 Hz	5.22 *dd*, *J* = 18.1, 1.5 Hz 5.34 *dd*, *J* = 11.2, 1.5 Hz	2.09 *dd*, *J* = 7.1, 3.7 Hz, α 1.24 *dd*, *J* = 8.8, 3.7 Hz, β
12	1.14 3H, *s*	0.96 3H, *s*	0.91 3H, *s*	0.92 3H, *s*
13	1.02 3H, *s*	0.85 3H, *s*	0.83 3H, *s*	0.85 3H, *s*
14	3.91 *d*, *J* = 11.2 Hz 4.03 *d*, *J* = 11.2 Hz	4.50 *d*, *J* = 2.0 Hz 4.76 *s*	4.73 *d*, *J* = 2.4 Hz 4.78 *s*	4.02 *d*, *J* = 12.0 Hz 4.11 *d*, *J* = 12.0 Hz
15	1.46 3H, *s*	1.74 *d*, *J* = 1.0 Hz	1.23 3H, *s*	0.91 3H, *d*, *J* = 7.3 Hz
-OAc	2.07 3H, *s*			1.98 3HZ, *s*
-OMe			3.18 3H, *s*	
-OH				5.30 *s*

* Measured by 600 MHz NMR (DMSO-d_6).

Table 2. ^{13}C NMR spectral data of compounds **1–3** ($CDCl_3$)

C	1	2	3
1	175.6	35.1	35.2
2	130.2	37.6	36.0
3	200.5	23.8	23.6
4	67.1	33.6	32.7
5	60.8	148.9	151.8
6	33.6	54.0	53.1
7	34.8	25.5	29.5
8	42.1	133.3	77.9
9	19.2	133.4	80.5
10	23.3	141.7	139.4
11	21.1	109.7	117.3
12	29.1	28.8	28.2
13	31.2	24.9	26.4
14	72.0	109.0	108.9
15	8.7	11.8	16.3
-OAc	20.9		
	170.7		
-OMe			50.2

tertiary methyls (δ_H 0.85, 0.96 each *s*), an olefinic methyl (δ_H 1.74 *d*), an *exo*-methylene (δ_H 4.50 *d*, 4.76 *s*, δ_C 109.0 *t*, 148.9 *s*), a trisubstituted olefin (δ_H 5.40 *t*, δ_C 133.3 *d*, 133.4 *s*) and a vinyl group (δ_H 4.87, 5.03 each *d*, 6.34 *dd*, δ_C 109.7 *t*, 141.7 *d*). The DEPT spectrum of **2** also indicated the presence of four methylenes, a methine and a quaternary carbon. The IR, ^{13}C NMR and GC-HR-mass spectra ($C_{15}H_{24}$ analyt. 204.1875) indicated that **2** was a monocyclic sesquiterpene hydrocarbon. The presence of four partial structures, (A) ■–C=CH_2, (B) ■–(CH_3)C=CH–CH_2–CH–■, (C) ■–CH=CH_2 and (D) ■–CH–CH_2–CH_2–■, were clarified by the analysis of the ^1H–^1H and ^{13}C–^1H COSY spectra. The connection of each partial structure was possible to correlate each segment by the HMBC spectrum. Two methyl groups (δ 0.85 and 0.96) were correlated with a quaternary carbon (δ 35.1), a methine carbon (δ 54.0) in segment B and a methylene carbon (δ 37.6) in segment D, respectively. The olefinic methyl (δ 1.74) in segment B was correlated with a methine carbon (δ 141.7) in segment C. Furthermore, the *exo*-methylenic protons (δ 4.50 and 4.76) in segment A was correlated with a methylene carbon (δ 33.6) in segment D and a methine carbon (δ 54.0) in segment B. Thus, the structure of **2** was determined to be a monocyclofarnesane-type sesquiterpene hydrocarbon. The *E*-configuration of the Δ^8-olefin was confirmed by the difference NOE spectrum in which NOEs were observed between (i) H-8 and H-10, and (ii) H-11 and H-15. NOEs were also observed between the tertiary methyl (δ 0.96) and H-2, H-6 and H-7, and another tertiary methyl (δ 0.85) and H-7, respectively. The above results presumed that the side chain at C-6 possessed *equatorial* configuration. The similar skeleton, β-monocyclonerolidol (**11**), has been isolated from the liverwort *Ptychanthus striatus* [10], whose stereochemistry has not been determined yet. The compound **2** is the dehydrated product of β-monocyclonerolidol (**11**). Thus, the structure of **2** was characterized as dehydro-β-monocyclonerolidol except for the absolute configuration.

The IR spectrum of **3** ($C_{16}H_{28}O_2$ analyt. *m/z* 252.2074) showed the presence of a hydroxyl group (3570 cm^{-1}). The ^1H NMR spectrum (Table 1) contained two tertiary methyls, a vinyl methyl, a methoxyl group (δ 3.18), an *exo*-methylene (δ 4.73 *d*, 4.78 *s*), a vinyl group (δ 5.22, 5.34, 5.85 each *dd*), and a methine proton with a hydroxyl group (δ 3.44 *dt*), respectively. Acetylation of **3** gave a monoacetate (**12**) (1760, 1250 cm^{-1}; CI-MS *m/z* 295 $[M+1]^+$; δ_H 1.94 3H *s*), and a methine proton at δ 3.44 observed in the ^1H NMR of **3** shifted to downfield at δ 4.85 in **12**. The ^{13}C NMR spectrum (Table 2) of **3** showed the presence of the oxygenated functional group bearing a methine (δ 77.9) and a quaternary carbon (δ 80.5) and three methyls, six methylenes, two methines and a further two quaternary carbons. Moreover, in comparison with the ^1H NMR and ^{13}C NMR spectra of **2**, the structure of **3** was presumed to be a monocyclofarnesane-type sesquiterpene alcohol. The ^1H–^1H and ^{13}C–^1H COSYs of **3** showed the presence of four segments: (i) ■–C(5)=CH_2(14); (ii) ■–(HO)CH(8)–CH_2(7)–CH(6)–■; (iii) ■–CH(10)=CH_2(11); and (iv) ■–CH_2(2)–CH_2(3)–CH_2(4)–■. The analysis of HMBC spectrum clarified the connectivity of each segment as shown in Table 3. NOEs were observed between H-12 and H-6

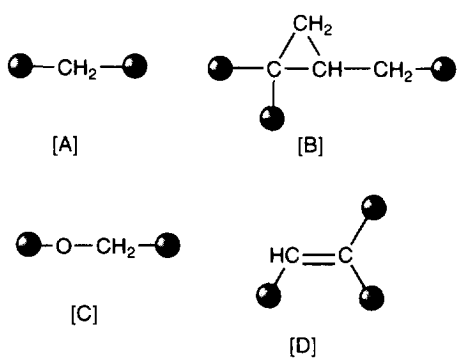

Fig. 1. Partial structures of compound **1**.

Table 3. Long-range ^1H–^{13}C correlations by the HMBC spectrum of compound **3**

H	C
4	2, 3, 5, 6, 14
6	1, 5, 7, 8, 14
7	1, 5, 6, 9
8	6, 9, 10
10	9, 15
11	9, 10
12	1, 2, 6, 13
13	1, 2, 6, 12
14	4, 6
15	8, 9, 10

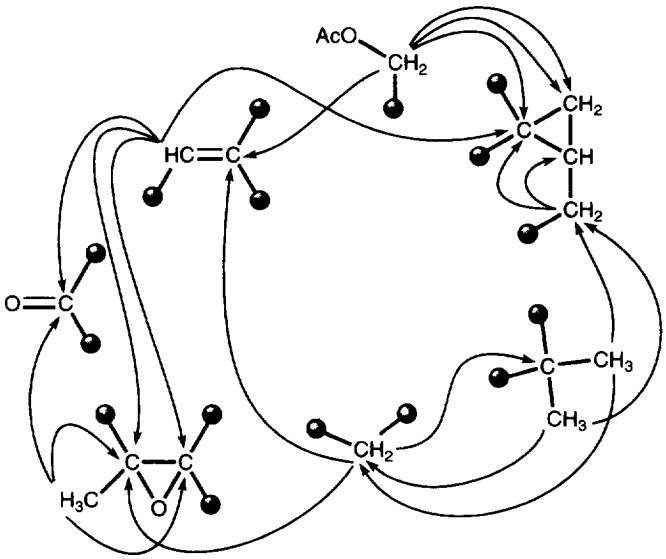

Fig 2. Long-range H–C correlations of compound **1**.

and H-7, and H-8 and H-6, indicating the side chain at C-6 was *equatorial*. Application of the modified Mosher's method [11], as shown in Fig. 3, supported that the configuration of the hydroxyl group at C-8 was *R*. Thus, the structure of **3** was established to be 8-hydroxy-9-methoxy-β-monocyclonerolidol.

The ^{1}H NMR and ^{13}C NMR spectra of **5** were completely identical with those of α-santalan-12(*R*), 13-diol (**9**) isolated from *Parella caespitans* var. *setigera* [4]. In our previous study, the absolute structure of **9** has been established by the comparison of the spectral data with those of the derivative from α-santalol (**14**) [4]. Furthermore, the configuration of the hydroxyl group at C-12 was decided by application of the empirical rule to the CD spectrum using Eu(FOD)$_3$ as chelating reagent [12, 13] which showed a negative Cotton effect. However, its value ($\Delta\varepsilon_{342} - 0.03$) was quite small in comparison with the reported data [12, 13] and therefore the CD spectrum of **5** was remeasured. Its spectrum showed the unexpected positive Cotton effect ($\Delta\varepsilon_{307} + 18.6$ in CCl_4 employing Eu(FOD)$_3$. In addition, application of the modified Mosher's method [10] (Fig. 4) using the (*R*)- and (*S*)-MTPA esters derived from **5** also supported the above result. Thus, the configuration of the hydroxyl group at C-12 was established to be *S*. Accordingly, the absolute structure of **9** was revised to α-santalan-12(*S*), 13-diol as shown in **5**. The error of the CD spectroscopic analysis of **5** might be caused by insufficient dryness of the solvent (CCl_4) and/or chelating reagent Eu(FOD)$_3$ [13].

The detection and isolation of santalane-type sesquiterpenoids have been reported only in *Plagiochila yokogurensis* [1] and *P. caespitans* var. *setigera* [4]. This is the third isolation from liverworts. The africane-type sesquiterpenoids have already been isolated from soft coral [14, 15], ascomycete fungus [16], Verbenaceae [17] and Compositae [18]. In liverworts, the isolation of africane-type sesquiterpenoids was reported only in two species, *P. caespitans* var. *setigera* [4] and *P. swartziana* [5–8]. Thus, the isolation of africanes is the third report from a liverwort. The present species is chemically very close to *P. caespitans* var. *setigera* [4], because both species produce africane- and santalane-type sesquiterpenoids. β-Monocyclonerolidol and pinguisane-type sesquiterpenoids have been found in the

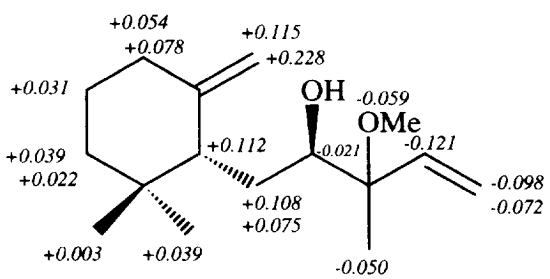

Fig. 3. $\Delta\delta$ values obtained for the MTPA ester of compound **3**.

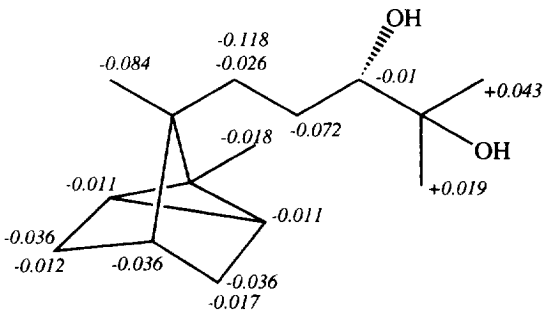

Fig. 4. $\Delta\delta$ values obtained for the MTPA ester of compound **5**.

Lejeuneaceae family (Hepaticae) and the Porellaceae, thus both families might originate from a common ancestor.

EXPERIMENTAL

The solvents used for spectral measurements were TMS-$CDCl_3$ [^{1}H- (600 and 400 MHz) and ^{13}C- (150, 100 and 50 MHz) NMR]; $CDCl_3$ ($[\alpha]_D$); UV and CD (MeOH). TLC was carried out as previously reported [19]. GC-MS was carried out on a 30 m × 0.25 mm, DB-17 column, carrier gas He, temp. 80–250° at 15° min^{-1}, detection EI at 70 eV.

Plant material. Porella subobtusa (Steph.) Hatt. was collected in Kisawa, Tokushima, Japan in Nov., 1992. The voucher specimen was deposited at the Institute of Pharmacognosy, Tokushima Bunri University.

Extraction and isolation. Air-dried *P. subobtusa* (37 g) was extracted with Et_2O and its crude extract (8 g) was chromatographed on silica gel (*n*-hexane–EtOAc gradient) to give 13 frs. Repeated chromatography of silica gel and silica gel impregnated with 10% $AgNO_3$ (benzene–*n*-hexane 1:99, 2:98 or *n*-hexane) of fr. 2 gave **2** (121 mg), **4** (30 mg) [3] and **6** (32 mg) [5].

Dehydro-β-monocyclonerolidol (**2**). $[\alpha]_D$ +9.5 (*c* 10.1); GC-HR-MS: Found $[M]^+$ 204.1875 $C_{15}H_{24}$ requires 204.1878; FTIR ν_{max} cm^{-1}: 1640, 1605, 1440, 1380, 1360; ^{1}H and ^{13}C NMR: Tables 1 and 2; GC-MS *m/z* (rel. int.): 204 $[M]^+$ (26), 189 (25), 161 (6), 148 (22), 133 (28), 123 (52), 109 (22), 81 (100), 79 (38), 53 (14), 41 (20).

Fr. 4 was chromatographed on Sephadex LH-20 (CH_2Cl_2–MeOH 1:1) and silica gel (*n*-hexane–Et_2O, 19:1) to give 8-hydroxy-9-methoxy-β-monocyclonerolidol (**3**) (18 mg), $[\alpha]_D$ +62.7 (*c* 1.75); HR-EI-MS: Found $[M]^+$ 252.2074; $C_{16}H_{28}O_2$ requires 252.2089; FTIR ν_{max} cm^{-1}: 3570 (OH); ^{1}H and ^{13}C NMR: Tables 1 and 2; EI-MS *m/z* (rel. int): 252 $[M]^+$ (0.5), 234 (1), 220 (2), 202 (3), 187 (4), 167 (5), 149 (8), 137 (4), 123 (25), 115 (17), 109 (7), 86 (100), 81 (15), 69 (15), 55 (20), 41 (9).

CC on Sephadex LH-20 (MeOH) of fr. 6 gave caespitenone (**7**) (762 mg) [6, 7]. Fr. 8 was chromatographed on Sephadex LH-20 ($CHCl_3$–MeOH, 1:1) and silica gel (*n*-hexane–EtOAc 5:1) to give frs A–C. The prep. HPLC (Cosmosil 5C_{18}, MeOH) of fr. C gave secoswartzianin A (**8**) (5 mg) [8]. Fr B was rechromatographed on silica gel (CH_2Cl_2–Et_2O 97:3) to give 14-acetoxycaespitenone (**1**) (42 mg). $[\alpha]_D$ −315.7 (*c* 1.48); HR-EI-MS: Found $[M]^+$ 290.1511 $C_{17}H_{22}O_4$ requires 290.1518; FTIR ν_{max} cm^{-1}: 1750, 1720, 1290, 1240 (C=O, OAc); UV λ_{max} nm (log ε): 276 (3.17), 210 (3.49) (*c* 4.1×10^{-4}); ^{1}H and ^{13}C NMR: Tables 1 and 2; EI-MS *m/z* (rel. int.): 290 $[M]^+$ (87), 275 (10), 262 (1), 248 (12), 231 (15), 215 (13), 202 (49), 187 (47), 174 (24), 159 (46), 146 (27), 131 (14), 117 (13), 105 (13), 91 (15), 77 (10), 69 (7), 55 (9), 43 (100); CD: $\Delta\varepsilon_{327}$ −1.45, $\Delta\varepsilon_{250}$ −2.73 (*c* 4.1×10^{-4}).

Fr. 10 was rechromatographed on Sephadex LH-20 ($CHCl_3$–MeOH 1:1) and silica gel (*n*-hexane–Et_2O gradient) to give α-santalan-12(*S*), 13-diol (**5**) (2.4 g) [4]. CD: $\Delta\varepsilon_{307}$ +18.6, $\Delta\varepsilon_{283}$ −12.7 (employing 6.6×10^{-3} M Eu(FOD)$_3$, CCl_4, *c* 7.5×10^{-3}).

Reduction of **1** *with Miyashita's reaction.* A soln of diphenyldiselenide (53 mg) in ethanol (4.5 ml) was treated with $NaBH_4$ (12 mg) under argon and stirred for 10 min at room temp. To this soln was added one drop of acetic acid and a solution of **1** (26.7 mg) in ethanol (2 ml), and then stirred at room temp. for 1.5 hr. To the reaction mixt. was added H_2O and extracted with EtOAc. The organic solution was washed with satd NaCl soln dried with $MgSO_4$, and evapd to afford a residue, which was chromatographed on Sephadex LH-20 (CHCl–MeOH, 1:1) and silica gel (Et_2O–CH_2Cl_2, 7:3) to give an alcohol **10** (12 mg). $[\alpha]_D$ −119.3 (*c* 1.15); HR-EI-MS: Found $[M]^+$ 292.1668; $C_{17}H_{24}O_4$ requires 292.1674; FTIR ν_{max} cm^{-1}: 3400 (OH), 1740, 1700, 1250, 1230 (C=O, OAc); ^{1}H NMR ($CDCl_3$ 400 MHz): δ 0.90 (3H, *s*, H-13), 0.99 (3H, *s*, H-12), 1.09 (3H, *d*, *J* = 7.3 Hz, H-15), 1.12 (1H, *d*, *J* = 14.7 Hz, H-6β), 1.25 (1H, *dd*, *J* = 8.8, 3.9 Hz, H-11β), 1.55 (1H, *dddd*, *J* = 8.8, 8.8, 6.8, 6.8 Hz, H-9), 1.63 (1H, *d*, *J* = 14.7 Hz, H-6α), 1.88 (1H, *dd*, *J* = 14.7, 8.8 Hz, H-8β), 2.04 (3H, *s*, -OCOMe), 2.13 (1H, *dd*, *J* = 6.8, 3.9 Hz, H-11α), 2.19 (1H, *dd*, *J* = 14.7, 6.8 Hz, H-8α), 2.53 (1H, *q*, *J* = 7.3 Hz, H-4), 4.02 (1H, *d*, *J* = 12.2 Hz, H-14), 4.17 (1H, *d*, *J* = 12.2 Hz, H-14), 6.17 (1H, *s*, H-2); ^{13}C NMR ($CDCl_3$, 100 MHz): δ 7.7 (*q*), 20.8 (*t*), 25.6 (*d*), 27.9 (*s*), 28.4 (*q*), 32.3 (*s*), 34.6 (*q*), 36.5 (*t*), 50.5 (*t*), 58.8 (*d*), 71.6 (*t*), 81.5 (*s*), 129.0 (*d*), 171.1 (*s*), 178.6 (*s*), 204.0 (*s*); ^{1}H NMR (C_5D_5N, 400 MHz): δ 0.94 (3H, *s*, H-13), 1.00 (3H, *s*, H-12), 1.23 (1H, *d*, *J* = 14.6 Hz, H-6β), 1.28 (3H, *d*, *J* = 7.3 Hz, H-15), 1.30 (1H, *dd*, *J* = 8.8, 3.4 Hz, H-11β), 1.60 (1H, *dddd*, *J* = 8.8, 8.8, 7.3, 7.3 Hz, H-9), 1.85 (1H, *dd*, *J* = 14.2, 8.8 Hz, H-8β), 1.95 (1H, *q*, *J* = 14.6 Hz, H-6α), 2.00 (3H, *s*, -OCOMe), 2.53–2.59 (2H, *q like*, $W_{1/2}$ = 10.7 Hz, H-8α and 11α), 3.01 (1H, *q*, *J* = 7.3 Hz, H-4), 4.28 (1H, *d*, *J* = 11.7 Hz, H-14), 4.32 (1H, *d*, *J* = 11.7 Hz, H-14), 6.56 (1H, *s*, H-2), 6.99 (1H, *s*, OH); ^{13}C NMR (C_5D_5N, 100 MHz): δ 8.3 (*q*), 20.6 (*q*), 23.6 (*t*), 25.9 (*d*), 28.4 (*q*), 28.7 (*s*), 32.5 (*s*), 34.6 (*q*), 36.6 (*t*), 50.8 (*t*), 59.0 (*d*), 71.8 (*t*), 81.1 (*s*), 128.7 (*d*), 170.8 (*s*), 180.0 (*s*), 204.0 (*s*); EI-MS *m/z* (rel. int): 292 $[M]^+$ (3), 277 (6), 250 (4), 232 (100), 217 (72), 204 (11), 187 (19), 176 (40), 161 (22), 147 (26), 133 (21), 120 (39), 105 (19), 91 (22), 83 (6), 77 (11), 69 (7), 55 (11), 43 (46).

Acetylation of **3**. Compound **3** (7.5 mg) in Ac_2O (0.5 ml) and pyridine (0.5 ml) was kept overnight at room temp. Work-up as usual gave a mono acetate **12** (6 mg). $[\alpha]_D$ +35.0 (*c* 0.01); CI-MS (*iso*-butane): 295 $[M+1]^+$; FTIR ν_{max} cm^{-1}: 1760, 1250 (OAc); ^{1}H NMR (400 MHz): δ 0.86 (3H, *s*), 0.89 (3H, *s*), 1.21 (1H, *m*), 1.22 (3H, *s*), 1.41–1.54 (3H, *m*), 1.63 (1H, *dt*, *J* = 14.7, 9.3 Hz), 1.79 (1H, *dd*, *J* = 9.3, 3.9 Hz), 1.94 (3H, *s*, -OCOMe), 1.98 (2H, *m*), 2.13 (1H, *m*), 3.18 (3H, *s*, -OMe), 4.53 (1H, *d*, *J* = 2.0 Hz), 4.64 (1H, *s*), 4.85 (1H, *dd*, *J* = 9.3, 2.4 Hz), 5.19 (1H, *dd*, *J* = 17.6, 1.0 Hz), 5.26 (1H, *dd*, *J* = 11.2, 1.0 Hz), 5.76 (1H, *dd*, *J* = 17.6, 11.2 Hz); ^{13}C NMR (100 MHz): δ 16.5, 21.2,

23.6, 26.6, 27.1, 28.0, 32.2, 35.3, 35.8, 50.5, 52.5, 78.4, 79.6, 108.6, 117.0, 140.4, 149.8, 170.6; EI-MS *m/z* (rel. int): 234 $[M - CH_3COOH]^+$ (6), 219 (3), 202 (8), 187 (8), 162 (3), 149 (11), 123 (5), 111 (22), 98 (7), 85 (100), 69 (9), 55 (19), 43 (30).

(S) *and* (R)-*MTPA ester of* **3**. To each soln of **3** (4 mg) in $CHCl_2$ (0.5 ml) was added (*S*)- or (*R*)-MTPA (each 34 mg), dicyclohexyl carbodiimide (DCC) (20 mg) and 4-dimethylaminopyridine (DMAP) (8 mg) and allowed to stand at room temp. for 2 days. Prep. TLC of the reaction mixture gave a (*S*)- and (*R*)-MTPA ester, **13a** (2 mg) and **13b** (2 mg), respectively.

Compound **13a**. ^{1}H NMR (600 MHz): δ 1.241 (1H, *m*, H-2), 1.408 (1H, *m*, H-2), 1.505 (2H, *m*, H-3), 1.945 (1H, *m*, H-4), 2.059 (1H, *m*, H-4), 1.907 (1H, *m*, H-6), 1.582 (1H, *ddd*, J = 14.9, 14.9, 7.6 Hz, H-7), 1.999 (1H, *dt like*, H-7), 5.243 (1H, *dd*, J = 7.6, 3.9 Hz, H-8), 5.655 (1H, *dd*, J = 17.8, 11.0 Hz, H-10), 5.143 (1H, *dd*, J = 17.8, 1.2 Hz, H-11), 5.224 (1H, *dd*, J = 11.0, 1.0 Hz, H-11) 0.847 (3H, *s*, H-12), 0.945 (3H, *s*, H-13), 4.574 (1H, *s*, H-14), 4.723 (1H, *s*, H-14), 1.220 (3H, *s*, H-15), 3.083 (3H, -OMe).

Compound **13b**. ^{1}H NMR (600 MHz): δ 1.202 (1H, *m*, H-2), 1.386 (1H, *m*, H-2),1.474 (2H, *m*, H-3), 1.891 (1H, *m*, H-4), 1.981 (1H, *m*, H-4), 1.795 (1H, *br t*, H-6), 1.474 (1H, *m*, 14.9, 7.6 Hz, H-7), 1.924 (1H, *m*, H-7), 5.264 (1H, *dd*, J = 7.6, 4.2 Hz, H-8), 5.776 (1H, *dd*, J = 17.8, 11.0 Hz, H-10), 5.215 (1H, *dd*, J = 17.8, 1.2 Hz, H-11), 5.322 (1H, *dd*, J = 11.0, 1.0 Hz, H-11), 0.844 (3H, *s*, H-12), 0.906 (3H, *s*, H-13), 4.346 (1H, *d*, J = 1.2 Hz, H-14), 4.608 (1H, *s*, H-14), 1.270 (3H, *s*, H-15), 3.142 (3H, -OMe).

(R)- *and* (S)-*MTPA ester of* **5**. To each soln of **5** (10 mg) in pyridine (0.5 ml) was added (*R*)- or (*S*)-MTPA (each 50 mg) and DMAP (10 mg) and stirred at room temp. for 2 hr. Each reaction mixture was partitioned between H_2O and $CHCl_3$. The organic layer was washed in 1 N HCl, 5% $NaHCO_3$, dried with $MgSO_4$, and evapd to afford (*R*)- and (*S*)-MTPA esters, respectively. Each ester was then purified by CC on silica gel (*n*-hexane–EtOAc system) to give (*R*)- and (*S*)-MTPA esters, **15a** (6 mg) and **15b** (7 mg), respectively.

Compound **15a**. ^{1}H NMR (400 MHz): δ 0.73 (3H, *s*, H-9), 0.84 (2H, *s*, H-1 and 5), 0.94 (3H, *s*, H-10), 1.04 (1H, *d*, J = 10.3 Hz, H-4), 1.07 (1H, *d*, J = 10.8 Hz, H-2), 1.15 (3H, *s*, H-14), 1.19 (3H, *s*, H-15), 1.21 (1H, *m*, H-8), 1.52 (3H, *m*, H-8′ and 11), 1.57 (3H, *m*, H-2, 3 and 4) 4.94 (1H, *dd*, J = 9.3, 2.4 Hz, H-12).

Compound **15b**. ^{1}H NMR (400 MHz): δ 0.65 (3H, *s*, H-9), 0.83 (2H, *s*, H-1 and 5), 0.92 (3H, *s*, H-10), 1.03 (1H, *m*, H-4), 1.05 (1H, *m*, H-2), 1.09 (1H, *m*, H-8), 1.17 (3H, *s*, H-14), 1.23 (3H, *s*, H-15), 1.43 (2H, *m*, H-11), 1.48 (1H, *m*, H-8), 1.52 (3H, H-2, 3 and 4), 4.93 (1H, *dd*, J = 9.8, 2.9 Hz, H-12).

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