Sesquiterpenoid Constituents of the Liverwort *Frullania brotheri* STEPH.

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Two new sesquiterpene lactones, (+)-β-frullanolide and (+)-brothenolide, have been isolated from the liverwort *Frullania brotheri* STEPH. One of them, (+)-brothenolide, has an allergenic property. The absolute structures of these lactones have been established by spectroscopic analysis and chemical transformation.

It is well-known that a number of sesquiterpene lactones have been isolated from the liverwort belonging to Frullaniacea and that some of them have an allergenic property.[1] In the course of our investigation on terpene constituents of the liverwort, we examined the constituents of *Frullania brotheri* STEPH. collected in Wakayama Prefecture around Mt. Ohto in June 1976 and isolated two new sesquiterpene lactones, (+)-β-frullanolide (**1**) and (+)-brothenolide (**2**), one of which, **2**, had an allergenic property. This paper deals in detail with the structural determination of (+)-β-frullanolide and (+)-brothenolide.[2]

Column chromatographic separation on SiO_2 and $AgNO_3$-SiO_2 of the ether extract of the fresh material gave lactones **1** and **2**. (+)-β-Frullanolide (**1**), mp 165—167 °C, $[\alpha]_D$ +178°, was obtained as colorless needles. The molecular formula $C_{15}H_{20}O_2$ of **1** was determined by the appearence of a molecular ion peak at 232.1453 in the high resolution mass spectrum (HR-MS). The IR spectrum ($CHCl_3$) showed an α,β-unsaturated γ-lactone absorption bands at 1760 and 1670 cm^{-1} and an exocyclic methylene absorption band at 910 cm^{-1}. The ^1H NMR spectrum ($CDCl_3$) indicated the presence of one tertiary methyl group (δ 0.92), four exocyclic methylene protons (δ 4.92, 5.09, 5.52, and 6.07) and one methine proton (δ 4.58) adjacent to oxygen. In the decoupling experiments of **1**, irradiation at δ 5.09 (14-H_B) collapsed the proton of 5-H at δ 2.01 and the allylic methylene protons (3-$H_{A,B}$) at δ 2.1—2.5 to sharp peaks, respectively. Also irradiation of 6-H at δ 4.56 collapsed a doublet of 5-H to a singlet and collapsed a multiplet of 7-H to sharp peaks. Furthermore, irradiation at δ 2.88 (7-H) converted a pair of doublets at δ 5.52 and 6.07 due to exocyclic methylene protons (13-$H_{A,B}$) into a pair of singlets and doublet of doublets at δ 4.58 (6-H) into a doublet. These results indicated that β-frullanolide (**1**) had the partial structure **1a**. When the ^1H NMR spectrum of **1** was compared with those of eudesmanolides,[3] it was similar to them. From these results, we assumed that lactone **1** had an eudesmanolide skeleton, as shown by structure **1**. This assumption was confirmed by a partial synthesis from costunolide (**3**)[4] according to the following pathway (Chart 1).

Costunolide (**3**), isolated from costus root oil, was cyclized with thionyl chloride to give the cyclocostunolide mixture[5] from which β-cyclocostunolide **4** was separated by column chromatography on 10% $AgNO_3$-SiO_2. This compound was identified as β-cyclocostunolide **4** with authentic spectra.[5] Next, this compound was converted to **5** by ethanolysis with Na_2CO_3 in ethanol. The IR spectrum showed hydroxyl and ester

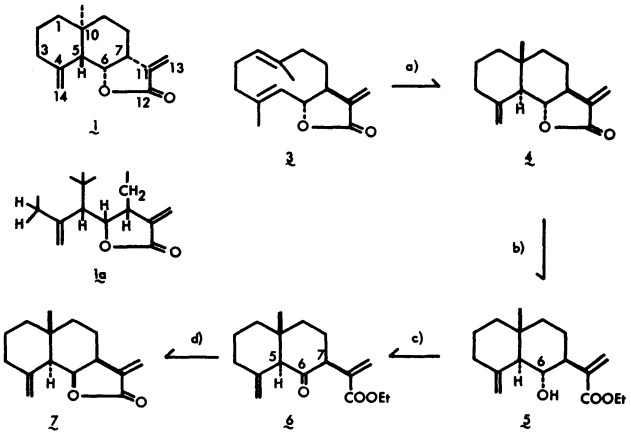

Chart 1. $SOCl_2$/pyridine, b) Na_2CO_3/EtOH, c) Jones reagent, d) $NaBH_4$/MeOH.

absorption bands at 3460 and 1715 cm^{-1}. In the ^1H NMR spectrum, the signals due to an ethyl group as the ester function at δ 1.30 and 4.23 were observed. The alcohol thus obtained was oxidized with Jones reagent to give keto ester **6**. The IR spectrum showed bands at 1718 cm^{-1} due to carbonyl groups and at 910 cm^{-1} due to an exocyclic methylene. In the ^1H NMR spectrum of **6**, the signals of 5-H and 7-H were observed at lower field (5-H: δ 3.08, s; 7-H: 3.67, t, J=10.0 Hz) than those of **5** (5-H: δ 1.90, d, J=11.0 Hz; 7-H: 2.60, dt, J=6.0 and 11.0 Hz). These results and the absence of a hydroxyl absorption in the IR and the signal of 6-H in the ^1H NMR of **6** indicated that hydroxyl group at C-6 was oxidized to keto group. Reduction of keto ester **6** with $NaBH_4$ in methanol gave a *cis* lactone **7**: $[\alpha]_D$ −161.8°.

Compound **7** was identified in every respect with (+)-β-frullanolide except for chiroptical property, CD (MeOH) $\Delta\varepsilon_{254}$+1.5 for **7** *vs.* $\Delta\varepsilon_{254}$−1.5 for **1** (Fig. 1). (+)-β-Frullanolide is fully represented by structure **1**.

(+)-Brothenolide (**2**), mp 113—114 °C, $[\alpha]_D$ +153.0°, CD (MeOH) $\Delta\varepsilon_{255}$−1.6, was obtained as colorless needles. The molecular formula $C_{15}H_{20}O_2$ of **2** was determined by HR-MS together with the ^{13}C and ^1H NMR data. The IR spectrum ($CHCl_3$) showed bands at 3040 cm^{-1} due to a cyclopropane ring, and at 1760 and 1660 cm^{-1} due to an α,β-unsaturated γ-lactone. The ^1H NMR spectrum ($CDCl_3$) showed the signals of two tertiary methyl groups (δ 1.11, s and 1.36, s), one proton on the cyclopropane ring (δ 0.38, t, J=3.6 Hz), two olefinic protons (δ 5.52, d, J=0.6 and 6.09, d, J=0.6 Hz), one allylic methine (δ 2.72, m) and one methine proton (δ 4.76, dd, J=3.0 and 4.8 Hz) adjacent to oxygen. The ^{13}C NMR spectrum ($CDCl_3$)

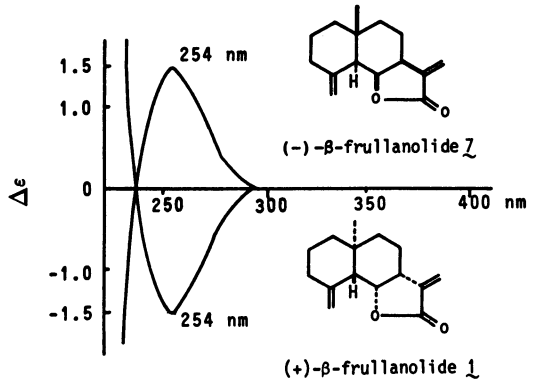

Fig. 1. CD spectra of (+)- and (−)-β-frullanolide.

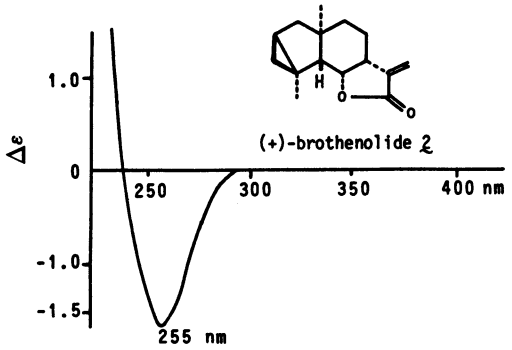

Fig. 2. CD spectrum of (+)-brothenolide.

also indicated the signals of α,β-unsaturated γ-lactone: C=O, δ 171.3; C-, 141.8; $=CH_2$, 119.5 and -O-CH, 77.8.

Hydrogenation of **2** with Pd/C in EtOH yielded a dihydro compound **8** (*m/z* 234). The ^1H NMR spectrum indicated the signal of a secondary methyl group at δ 1.06 instead of the protons of the exocyclic methylene in the spectrum of **2**. This compound was further reduced[6] with PtO_2 in EtOH-AcOH to give a tetrahydro compound **9** (*m/z* 236, $C_{15}H_{24}O_2$). The IR spectrum indicated the absence of a cyclopropane ring absorption and the presence of *gem*-dimethyl absorption bands at 1380 and 1370 cm^{-1}. In the ^1H NMR spectrum, a signal of a tertiary methyl group was observed in place of the protons on the cyclopropane ring. This fact showed that one of the two methyl groups in **2** should be attached to the cyclopropane ring. The presence of the partial structure **2a** was indicated by the above result, and the results of double irradiation experiments in the ^1H NMR spectrum of **2** with the aid of shift reagent. The signal of the proton at 7-H appeared as a multiplet which collapsed to a broad doublet of doublets (J=7.0 and 10.2 Hz) on irradiation of 6-H. In addition, irradiation of the signal of 6-H converted the doublet of 5-H into a singlet. The fact that 7-H is coupled to the 8α- and 8β-H's with J=7.0 and 10.2 Hz indicated that it is axial. Thus the $J_{6,7}$ value of 4.8 Hz shows that 6-H is equatorial. Therefore, the γ-lactone ring must be *cis*-fused at C-6 and C-7. The reduction of **9** with LAH afforded a diol **10**, the IR spectrum of which showed the presence of hydroxyl groups at 3500 and 3430 cm^{-1}. The ^1H NMR spectrum showed the signals of methylene protons adjacent to oxygen as primary alcohol at δ 3.44 and 3.56. The diol **10** was acetylated with acetic anhydride and pyridine at room temperature to give monoacetate **11**, the IR spectrum of which showed bands at 3510 and 1720 cm^{-1} due to the hydroxyl group and the carbonyl group, respectively. The ^1H NMR spectrum showed also the signals of the acetoxymethyl group at δ 2.06 and the $-C\underline{H}_2-OAc$ group at δ 3.94 and 4.24. The monoacetate **11** was oxidized with Jones reagent to yield a corresponding keto compound **12**. The IR spectrum indicated the presence of a 6-membered ring ketone at 1715 cm^{-1}. Furthermore, in the decoupling experiments of **2** using 360 MHz NMR, irradiation of 1-H_A at δ 0.83 collapsed the doublet of doublets of 1-H_B (δ 1.77) to a doublet, and a multiplet of 2-H (δ 1.22) to a sharp peak. In addition, irradiation of 3-H_A (δ 0.38) collapsed the doublet of doublets of 3-H_B (δ 0.93) to a doublet, and 2-H (δ 1.22) to sharp peak. This result indicated that brothenolide (**2**) had a partial structure **2b**. All these data show that the new sesquiterpene lactone **2** is most favorably represented by formula **2**. The fact that the CD Cotton effects of lactones **1** and **2** have the same signs (Figs. 1 and 2) shows that they belong to the same chiroptical series.[7]

Table 1. Chemical shift of the methyl groups (in δ)

	4-CH_3	4-CH_2D	10-CH_3
11-d_2	1.20	1.17	1.03
12-d_2	1.34	1.07	0.91

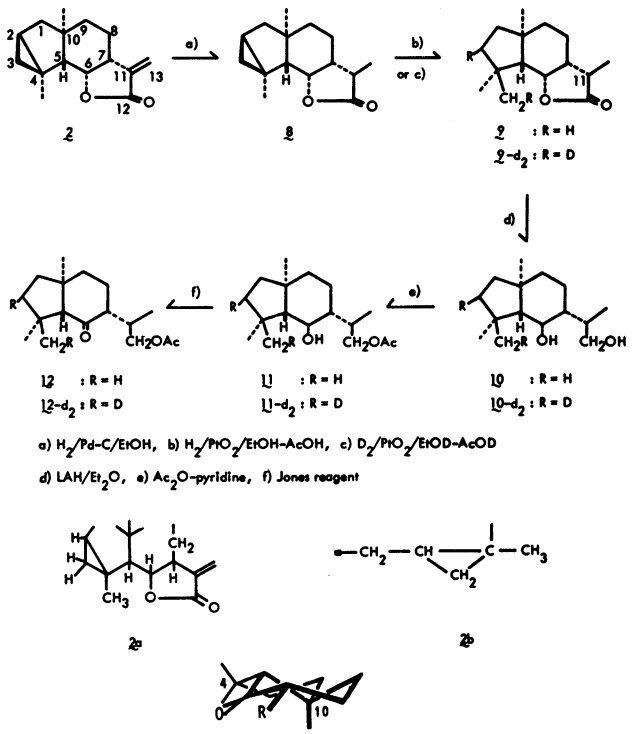

Chart 2.

The β-configuration of the cyclopropane ring was determined as follows. Reduction of dihydrobrothenolide **8** with deuterium instead of hydrogen (see above) in EtOD-AcOD, followed by LAH treatment and acetylation gave **11**-d_2 (the dideuterio derivative of **11**).

The ^1H NMR spectrum of **11** showed three tertiary methyl signals at δ 1.17, 1.20, and 1.03, one of which, at δ 1.17, appeared as a CH_2D signal in **11**-d_2. Oxidation of **11**-d_2 with Jones reagent gave **12**-d_2, the ^1H NMR spectrum of which showed the signals of two tertiary methyl groups at δ 1.34 and 0.91, and a tertiary CH_2D group at δ 1.07. The chemical shifts of CH_3 and CH_2D groups of **11**-d_2 and **12**-d_2 are shown in Table 1. One methyl peak at δ 1.20 in **11**-d_2 underwent a low field shift to δ 1.34 in **12**-d_2 whereas the other methyl peaks at δ 1.17 and 1.03 in **11**-d_2 shifted to δ 1.07 and 0.91 in **12**-d_2. These shifts of methyl peaks were explained by the difference of anisotropic effects between 6α-OH and 6-keto function. The methyl signals at δ 1.17, 1.20, and 1.03 in **11**-d_2 were therefore assigned to 4β-Me, 4α-Me, and 10-Me groups, respectively.

Molecular model **12a** clearly shows that 4-Me group (δ 1.34) should have the α-configuration since it is in-plane in relation to the carbonyl group. The CH_2D group in **11**-d_2 is thus β-oriented, and therefore, the cyclopropane methylene group is also β-oriented in brothenolide (**2**).

Experimental

All melting points are uncorrected. IR spectra were measured with a Hitachi EPI-G2 spectrometer, ^{13}C NMR spectra with a JEOL FX-100 (25.0 Hz) spectrometer, ^1H NMR spectra with a JEOL FX-100 (100 MHz), a Nicolet NT-360 (360 MHz), or a Hitachi R-20B (60 MHz) spectrometer in deuteriochloroform solution containing tetramethylsilane as an internal standard, low resolution mass spectra with a Hitachi RMU-6 mass spectrometer and high resolution mass spectra with a JEOL-JMS 01SG-2, with direct inlet system operating at 70 eV. CD spectra were measured with a JASCO J-20 spectrometer. For column chromatography Kieselgel 60 (E. Merck, Darmstadt) was used. Thin-layer chromatography (TLC) was carried out on Kieselgel GF_{254} (E. Merck, Darmstadt) in 0.25 mm thickness.

Isolation. The fresh material (100 g) of *Frullania brotheri* Steph. collected in Wakayama Prefecture around Mt. Ohto in June 1976 was extracted with ether at room temperature. The ether extract (1.8 g) was chromatographed on SiO_2. Elution with $CHCl_3$ gave a lactone mixture that was subjected to further column chromatography on SiO_2 impregnated with 10% $AgNO_3$. Elution with hexane–Et_2O (20 : 1) yield a fraction of (+)-brothenolide. Next, elution with ether gave crude (+)-β-frullanolide.

(+)-β-Frullanolide (1). The crude material was recrystallized from hexane–EtOAc to yield colorless needles (**1**, 40 mg), mp 165—167 °C, $[\alpha]_D$ +178.0° (*c* 1.17, $CHCl_3$); CD, $\Delta\varepsilon_{254}$ −1.5 (*c* 0.012, MeOH); IR ($CHCl_3$) 1760, 1670, and 910 cm^{-1}; ^1H NMR δ 0.92 (3H, s, 10-CH_3), 4.58(1H, dd, J=4.5 and 3.0 Hz, 6-H), 4.92 (1H, br. s, 14-H_A), 5.09 (1H, br. s, 14-H_B), 5.52 (1H, d, J=0.6 Hz, 13-H_A), and 6.07 (1H, d, J=0.6 Hz, 13-H_B); ^{13}C NMR δ 18.3 (q), 23.0 (t), 24.6 (t), 34.7 (t), 37.5 (t), 38.8 (t), 41.4 (d), 43.0 (t), 50.9 (d), 77.8 (d), 109.7 (t), 119.0 (t), 141.7 (s), 145.7 (s) and 170.8 (s); MS *m/z* (%) 232 (93, M^+), 217 [100, $(M-CH_3)^+$], 176 (35), 171 (34), 91 (46), and 79 (37). Found: *m/z* 232.1453. Calcd for $C_{15}H_{20}O_2$: M, 232.1445.

Isolation of (+)-Brothenolide (2). The crude fraction was recrystallized from hexane–EtOAc to give colorless needles (**2**, 270 mg), mp 113—114 °C, $[\alpha]_D$ +153.0° (*c* 1.14, $CHCl_3$); CD, $\Delta\varepsilon_{255}$ −1.6 (*c* 0.012, MeOH); IR ($CHCl_3$) 3040, 1760, 1660 cm^{-1}; ^1H NMR (360 MHz) δ 0.38 (1H, t, J=3.6 Hz, 3-H_A), 0.83 (1H, br. dd, J=3.6 and 12.5 Hz, 1-H_A), 0.93 (1H, dd, J=3.6 and 8.7 Hz, 3-H_B), 1.06 (1H, d, J=3.0 Hz, 5-H), 1.11 (3H, s, 10-CH_3), 1.22 (1H, m, 2-H), 1.22 (1H, m, 9-H_B), 1.36 (3H, s, 4-CH_3), 1.47 (1H, m, 8-H_A), 1.60 (1H, ddd, J= 3.0, 4.2, and 12.6 Hz, 9-H_B), 1.72 (1H, dd, J=7.5 and 12.5 Hz, 1-H_B), 2.72 (1H, m, 7-H), 4.76 (1H, dd, J=3.0 and 4.8 Hz, 6-H), 5.52 (1H, d, J=0.6 Hz, 13-H_A), and 6.09 (1H, d, J=0.6 Hz, 13-H_B); ^{13}C NMR δ 20.4 (q), 20.9 (q), 25.5 (t), 25.9 (d), 26.6 (s), 34.1 (t), 35.7 (t), 40.5 (d), 46.3 (t), 50.4 (s), 56.5 (d), 77.8 (d), 119.5 (t), 141.8 (s), and 171.3 (s); MS *m/z* (%) 232 (8, M^+), 217 [100, $(M-CH_3)^+$], 199 (39), 171 (44), 145 (31), 119 (45), 91 (41), and 79 (28). Found: *m/z* 232.1472. Calcd for $C_{15}H_{20}O_2$: M, 232.1445.

Isolation of Costunolide (3). Costus root oil (3 g) was chromatographed on SiO_2, and eluted with $CHCl_3$ to give costunolide (**3**, 270 mg), colorless needles, mp 104—105 °C (recrystallized from hexane), $[\alpha]_D$ +127.0° (*c* 1.05, $CHCl_3$). The IR, NMR, and mass spectra, and $[\alpha]_D$ value of **3** were found to be identical with those of known (+)-costunolide.[4]

Cyclization of Costunolide (3). A solution of costunolide (270 mg) in $CHCl_3$ (50 ml) containing $SOCl_2$ (0.1 ml) was kept at room temperature for 1 h. After removal of solvent under vacuum, the resulting residue was chromatographed on 10% $AgNO_3$–SiO_2. Elution with hexane–EtOAc (5 : 1) gave β-cyclocostunolide (160 mg), colorless needles, mp 67—68 °C (recrystallized from EtOH), $[\alpha]_D$ +175.3° (*c* 0.93, $CHCl_3$). The IR, NMR, and mass spectra, and $[\alpha]_D$ value were identical with those of known (+)-β-cyclocostunolide.[5]

Ethanolysis of β-Cyclocostunolide (4). 0.05 M (1 M=1 mol dm^{-3}) Na_2CO_3 aqueous solution (0.2 ml) was added to a solution of β-cyclocostunolide (150 mg) in EtOH (10 ml), and the mixture was kept at room temperature for 2 d. The reaction mixture was diluted with H_2O, evaporated to remove EtOH under reduced pressure and extracted with ether. Ether layer was washed with H_2O and dried over Na_2SO_4. After removal of solvent under reduced pressure, the resulting residue was chromatographed on SiO_2. Elution with $CHCl_3$ gave hydroxy ester **6**, colorless needles, mp 95—96 °C (recrystallized from hexane), $[\alpha]_D^{20}$ +25.2° (*c* 0.92, $CHCl_3$); IR ($CHCl_3$) 3460 and 1715 cm^{-1}; ^1H NMR δ 0.83 (3H, s, 10-CH_3), 1.30 (3H, t, J=8.0 Hz, $OCH_2C\underline{H}_3$), 1.90 (1H, d, J=11.0 Hz, 5-H), 2.60 (1H, dt, J=6.0 and 11.0 Hz, 7-H), 3.92 (1H, t, J= 11.0 Hz, 6-H), 4.23 (2H, q, J=8.0 Hz, $OC\underline{H}_2CH_3$), 4.68 (1H, br. s, 14-H_A), 4.96 (1H, br. s, 14-H_B), 5.69 (1H, br. s, 13-H_A), and 6.27 (1H, br. s, 13-H_B); MS *m/e* (%) 278 (trace, M^+), 232 [53, $(M-EtOH)]^+$], 217 (33), 123 (61), and 81 (100). Found: *m/z* 278.1864. Calcd for $C_{17}H_{26}O_3$: 278.1869.

Oxidation of 5 with Jones Reagent. A few drops of Jones reagent were added slowly to a stirred solution of **5** (80 mg) in acetone (10 ml) at 0 °C, and the mixture was further stirred for 2 d at room temperature. The reaction mixture was treated in the usual way to give a residue which was chromatographed on SiO_2. Elution with $CHCl_3$ gave keto compound **6**, colorless needles, mp 64 °C (recrystallized from hexane), $[\alpha]_D^{22}$ +10.6° (*c* 1.0, $CHCl_3$); IR ($CHCl_3$) 1718, 1640, and 910 cm^{-1}; ^1H NMR δ 0.83 (3H, s, 10-CH_3), 1.27 (3H, t, J=8.0 Hz, $OCH_2C\underline{H}_3$), 3.08 (1H, s, 5-H), 3.63 (1H, t, J=10.0 Hz, 7-H), 4.18 (1H, q, J=8.0 Hz, $OC\underline{H}_2CH_3$), 4.98 (1H, br. s, 14-H_A), 5.54 (1H, br. s, 13-H_A), 5.88 (1H, br. s, 14-H_B), and 6.32 (1H, br. s, 13-H_B); MS *m/z* (%) 276 (55, M^+), 230 [92, $(M-EtOH)^+$], 175 (89), 135 (100), and 107 (97). Found: *m/z* 276.1714. Calcd for $C_{17}H_{24}O_3$: M, 276.1706.

Reduction of 6 with $NaBH_4$. After $NaBH_4$ (4.4 mg) was added to an ice-cooled solution of **6** (26 mg) in MeOH (0.3 ml), the mixture was stirred at room temperature for 2 h. The

reaction mixture was diluted with H_2O and extracted with CH_2Cl_2. The CH_2Cl_2 layer was treated in the usual way to give a residue (20 mg) which was chromatographed on TLC (solvent system; hexane–EtOAc=10 : 1). Colorless needles (**7**, 8 mg) were obtained by recrystallization from hexane–EtOAc, mp 164—165 °C, $[\alpha]_D$ −161.8° (*c* 0.64, $CHCl_3$); CD, $\Delta\varepsilon_{254}$ +1.5 (*c* 0.012, MeOH). Found: *m/z* 232.1449. Calcd for $C_{15}H_{20}O_2$: M, 232.1445. The IR, NMR and mass spectra were found to be identical with those of (+)-β-frullanolide (**1**).

Hydrogenation of Brothenolide (2). Lactone **2** (25 mg) and palladium charcoal (10 mg) in EtOH (2 ml) were stirred under hydrogen atmosphere at room temperature for 3 h. The catalyst was removed by filtration and the solvent was evaporated to give a residue (25 mg), which was purified by recrystallization from hexane (**8**, 18 mg). Colorless needles, mp 105—106 °C; IR ($CHCl_3$) 3040 and 1770 cm^{-1}; ^1H NMR δ 0.32 (1H, t, *J*=3.6 Hz, 3-H_A), 1.06 (3H, d, *J*=7.0 Hz, 11-CH_3), 1.18 (3H, s, 10-CH_3), 1.36 (3H, s, 4-CH_3), and 4.53 (1H, dd, *J*=3.0 and 4.8 Hz, 6-H); MS *m/z* (%) 234 (7, M^+), 219 [100, $(M-CH_3)^+$], 161 (57), 145 (71), and 107 (45). Found: *m/z* 234.1613. Calcd for $C_{15}H_{22}O_2$: M, 234.1608.

Hydrogenation of Dihydro Lactone (8). Dihydro lactone **8** (20 mg) and platinum catalyst (prepared from platinum oxide; 10 mg) in EtOH (1.5 ml) and AcOH) (0.3 ml) were stirred under hydrogen atmosphere at room temperature for 8 h. The reaction mixture was filtered and the filtrate was evaporated under reduced pressure to give a residue (21 mg). Tetrahydro lactone **9** was obtained by recrystallization from hexane, (**9**, 15 mg), colorless needles, mp 138—140°C; IR (CCl_4) 1780, 1380, and 1370 cm^{-1}; ^1H NMR δ 1.04 (3H, s), 1.07 (3H, s), 1.17 (3H, s), 1.20 (3H, d, *J*=6.6 Hz, 11-CH_3), and 4.52 (1H, dd, *J*=3.0 and 4.8 Hz, 6-H); MS *m/z* (%) 236 (1.5, M^+), 221 [16, $(M-CH_3)^+$], 177 (43), 147 (62), 123 (40), 109 (100), and 107 (69). Found: *m/z* 236.1762. Calcd for $C_{15}H_{24}O_2$: M, 236.1751.

Reduction of Tetrahydro Lactone (9) with Lithium Aluminium Hydride. After a solution of LAH (10 mg) in dry Et_2O (2 ml) was added to an ice-cooled solution of tetrahydro lactone **9** (12 mg) in dry ether (2 ml), the mixture was kept at room temperature for 3 h. The reaction mixture was treated in the usual way to give a residue (15 mg) which was chromatographed on SiO_2. Elution with $CHCl_3$–MeOH (15 : 1) gave a diol compound (**10**, 10 mg), colorless viscous oil, IR (film) 3500, 3430, and 1045 cm^{-1}; ^1H NMR δ 0.98 (3H, d, *J*=7.0 Hz, 11-CH_3), 1.02 (3H, s), 1.19 (6H, s), 3.44 (1H, dd, *J*=5.0 and 12.0 Hz, $\underline{H_A}CH$–OH), 3.56 (1H, dd, *J*=6.5 and 12.0 Hz, $HC\underline{H_B}$–OH), and 4.14 (1H, br. s, 6-H); MS *m/z* (%) 240 (trace, M^+, $C_{15}H_{28}O_2$), 222 [43, $(M-H_2O)^+$], 209 (100).

Acetylation of Diol Compound (10). A solution of diol **10** (10 mg) in dry pyridine (2 ml) and Ac_2O (1 ml) was kept at room temperature for 3 h. The reaction mixture was treated in the usual way to give a residue (13 mg), which was purified by SiO_2 column chromatography using $CHCl_3$ to give monoacetate, (**11**, 8 mg), colorless viscous oil, IR (film) 3510, 1720, and 1370 cm^{-1}; ^1H NMR δ 1.03 (3H, s, 10-CH_3), 1.06 (3H, d, *J*=7.0 Hz, 11-CH_3), 1.17 (3H, s, 4-CH_3), 1.20 (3H, s, 4-CH_3), 2.06 (3H, s, $COCH_3$), 3.94 (1H, dd, *J*=7.5 and 11.0 Hz, 12-H_A), 4.18 (1H, br. s, 6-H), and 4.24 (1H, dd, *J*=2.1 and 11.0 Hz, 12-H_B); MS *m/z* (%) 282 (5, M^+, $C_{17}H_{30}O_3$), 142 (58), 141 (100), and 140 (42).

Oxidation of Acetate (11) with Jones Reagent. A drop of Jones reagent was added to a solution of **11** (10 mg) in acetone (1 ml) at 0 °C, and the mixture was further stirred for 3 h at room temperature. The reaction mixture was treated in the usual way to give a residue (10 mg), which was chromatographed on TLC using $CHCl_3$ to afford keto acetate, (**12**, 6 mg), colorless viscous oil, IR (film) 1735 and 1715 cm^{-1}; ^1H NMR δ 0.91 (3H, s, 10-CH_3), 0.98 (3H, d, *J*=7.0 Hz, 11-CH_3), 1.07 (3H, s, 4-CH_3), 1.34 (3H, s, 4-CH_3), 2.03 (3H, s, $COCH_3$), 2.12 (1H, s, 5-H), 2.24 (1H, m, 7-H), 3.93 (1H, dd, *J*=7.1 and 11.0 Hz, 12-H_A), and 4.09 (1H, dd, *J*=2.1 and 11.0 Hz, 12-H_B); MS *m/z* (%) 280 (17, M^+, $C_{17}H_{28}O_3$), 265 [36, $(M-CH_3)^+$], and 220 [100, $(M-60)^+$].

Preparation of D-Labelled Compounds (9-d_2), (11-d_2), and (12-d_2). Dihydro lactone **8** (10 mg) and platinum catalyst (prepared from platinum oxide; 5 mg) in EtOD (1.5 ml) and AcOD (0.3 ml) were stirred under deuterium atmosphere at room temperature for 8 h. The reaction mixture was filtered and the filtrate was evaporated *in vacuo* to give a residue which was chromatographed on TLC using $CHCl_3$ to give **9**-d_2 (7 mg), MS *m/z* (%) 238 (1, M^+, $C_{15}H_{22}D_2O_2$), and 110 (100); IR (CCl_4) 1780, 1380, and 1370 cm^{-1}; ^1H NMR δ 1.04 (3H, s, 10-CH_3), 1.08 (2H, s, 4-CH_2D), 1.16 (3H, s, 4-CH_3), 1.20 (3H, *J*=6.5 Hz, 11-CH_3), and 4.51 (1H, br. s, 6-H).

Conversion of 9-d_2 into 11-d_2. A mixture of **9**-d_2 (7 mg) and LAH (10 mg) in dry ether (3 ml) was stirred at room temperature for 3 h. The reaction mixture was treated in the usual way. The residue was treated with acetic anhydride (0.5 ml) and pyridine (1 ml) at room temperature for a day and then worked up in the usual way. The resulting residue was chromatographed on TLC using $CHCl_3$ to yield **11**-d_2, (4 mg); MS *m/z* (%) 284 (54, M^+, $C_{17}H_{28}D_2O_3$), 269 [31, $(M-CH_3)^+$] and 209 (100); IR (film) 3500, 1720 and 1380 cm^{-1}; ^1H NMR δ 1.03 (3H, s, 10-CH_3), 1.06 (3H, d, *J*=7.0 Hz, 11-CH_3), 1.17 (2H, s, 4-CH_2D), 1.20 (3H, s, 4-CH_3), 2.06 (3H, s, $COCH_3$), 3.94 (1H, dd, *J*=7.5 and 11.0 Hz, 12-H_A), 4.18 (1H, br. s, 6-H), and 4.24 (1H, dd, *J*=2.1 and 11.0 Hz, 12-H_B).

Oxidation of 11-d_2 with Jones Reagent. To a solution of **11**-d_2 (4 mg) in acetone (1 ml) was added a drop of Jones reagent at 0 °C. The mixture was stirred for a day at room temperature, and then worked up in the usual way. The residue was chromatographed on TLC using $CHCl_3$ to afford **12**-d_2 (2 mg), MS *m/z* (%) 282 (32, M^+, $C_{17}H_{26}D_2O_3$), 267 [28, $(M-CH_3)^+$], and 222 [100, $(M-60)^+$]; IR (film) 1735 and 1715 cm^{-1}; ^1H NMR δ 0.91 (3H, s, 10-CH_3), 0.98 (3H, d, *J*=7.0 Hz, 11-CH_3), 1.07 (2H, s, 4-CH_2D), 1.34 (3H, s, 4-CH_3), 2.03 (3H, s, $COCH_3$), 2.13 (1H, s, 5-H), 2.24 (1H, m, 7-H), 3.93 (1H, dd, *J*=7.1 and 11.0 Hz, 12-H_A), and 4.09 (1H, dd, *J*=2.1 and 11.0 Hz, 12-H_B).

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