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Steroid Saponins from *Paris polyphylla* Sm.—Supplement[1]

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Previously, four steroid glycosides (Pa—d) were isolated from the rhizomes of *Paris polyphylla* Sm. and characterized. In a continuation of our studies on this plant, a further five new steroid glycosides (**1**, **2**, **12**, **14** and **16**) together with three known compounds (**13**, **18** and **19**; the latter two have been obtained synthetically as acetyl derivatives) have been isolated and their structures established as follows. **1** and **2**: (22*ξ*,25*R*)-22-methoxyfurostanol 3,26-*O*-bisglycosides (proto-type saponins) corresponding to Pa [diosgein 3-*O*-α-L-ara·fur-(1→4)-[α-L-rha·pyr-(1→2)]-β-D-glc·pyranoside (**3**)] and Pb [diosgenin 3-*O*-α-L-rha·pyr-(1→4)-α-L-rha·pyr-(1→4)-[α-L-rha·pyr-(1→2)]-β-D-glc·pyranoside (**6**)], respectively; **12** and **13**: pennogenin oligosides having the same sugar moieties as **3** and **6**, respectively; **14** and **18**: pennogenin derivatives carrying 3-*O*-α-L-ara·fur-(1→4)-β-D-glc·pyranoside and hexaacetyl 3-*O*-α-L-rha·pyr-(1→2)-β-D-glc·pyranoside, respectively; **16** and **19**: diosgenin derivatives carrying the same sugar moieties as **14** and **18**, respectively.

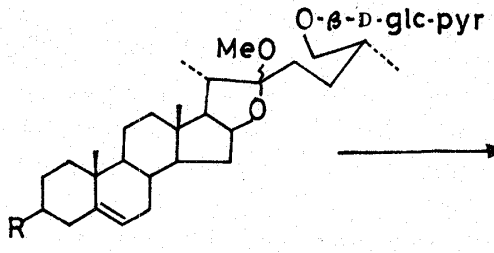
Keywords——*Paris polyphylla*; Liliaceae; furostanol glycoside; diosgenin glycoside; pennogenin glycoside; arabinofuranoside

In the previous paper,[1] we reported four steroid glycosides (Pa through Pd) from the rhizomes of Himarayan *Paris polyphylla* Sm. (Liliaceae), which is used as a folk medicine in some parts of Nepal. In China, this plant has been used to treat chronic bronchitis. In a continuation of our studies on the ingredients of this plant, we have obtained a further eight steroid glycosides (**1**, **2**, **12**, **13**, **14**, **16**, **18** and **19**) together with Pa (**3**) and Pb (**6**). This paper deals with their structure determinations.

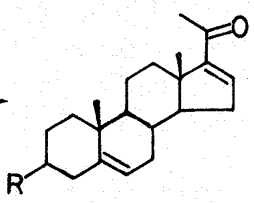
Separation of the MeOH extractive of the crude drug, commercial dried rhizomes of *Paris polyphylla* purchased in a market in China, was undertaken as described in Experimental, affording **1**, **2**, **12**, **13**, **14**, **16**, **18** and **19** in addition to Pa—d previously isolated.

Both **1**, an amorphous powder (mp 209—212°C), $[\alpha]_D$ −135.6°, and **2**, an amorphous powder (mp 174—177°C), $[\alpha]_D$ −87.0°, showed strong absorptions (3600—3200 cm^{-1}) due to hydroxyl groups, but not due to spiroketal functions[2] in their infrared (IR) spectra and were positive to Ehrlich reagent, which is a convenient reagent[3] to detect proto-type saponins such as protodioscin,[4] for example. They also showed methoxy signals[4] reminiscent of 22-methoxyfurostane derivatives in their ^1H nuclear magnetic resonance (^1H NMR) spectra. On the assumption that **1** and **2** were proto-type saponins, the compounds were hydrolyzed with β-glucosidase (almond emulsin) to afford spirostanol derivatives, **3**, colorless needles, mp 274—276°C, $[\alpha]_D$ −133.0°, identical (mp, $[\alpha]_D$, thin layer chromatography (TLC), ^{13}C nuclear magnetic resonance (^{13}C NMR)) with Pa, and **6**, colorless needles, mp 203—206°C, $[\alpha]_D$ −128.0°, identical (mp, $[\alpha]_D$, TLC, ^{13}C NMR) with Pb, respectively, both accompanied with D-glucose ($[\alpha]_D$, TLC). Furthermore, the identity of **3** and **6**, obtained by enzymic hydrolysis of **1** and **2**, with Pa and Pb, respectively, was substantiated by their transformation into the acetylated derivatives (**4**, colorless needles, mp 122—128°C, $[\alpha]_D$ −100.0°, and **7**, colorless needles, mp 148—153°C, $[\alpha]_D$ −74.0°, respectively) and methylated derivatives (**5**, colorless needles, mp 160—162°C, $[\alpha]_D$ −92.0°, and **8**, colorless needles, mp 135—139°C, $[\alpha]_D$ −110.0°, respectively). Therefore, **1** and **2** could be deduced to be proto-type saponins corresponding to Pa (**3**) and Pb (**6**), respec-

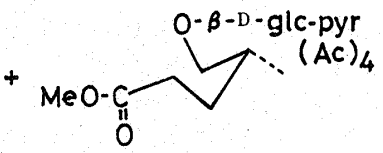
tively. To verify the structures of **1** and **2**, they were acetylated with Ac_2O–pyridine, refluxed with Ac_2O for pseudomerization into $\Delta^{20(22)}$ and oxidized with CrO_3 in AcOH (Marker's degradation[5]). After treatment of the oxidized products with alkali, **1** and **2** were finally decomposed respectively into 3β-hydroxy-pregna-5,16-dien-20-one glycosides, **9**, an amorphous powder (mp 214—216°C), $[\alpha]_D$ −42.0°, and **10**, colorless needles, mp 234—237°C, $[\alpha]_D$ −117.7°, respectively, both accompanied with a product formed from the steroidal side chain moiety, which on acetylation followed by methylation with CH_2N_2 afforded methyl 4(*R*)-methyl-5-hydroxy-



1 : R=S_1
2 : R=S_2



9 : R=S_1
10 : R=S_2



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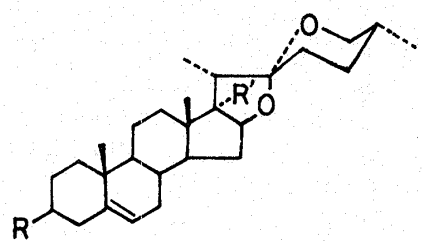
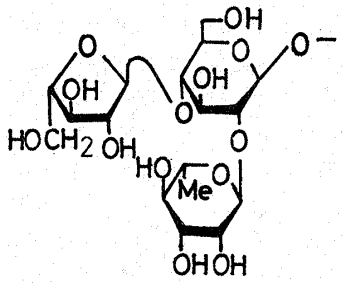


3: R=S_1, R′=H(=Pa)
4: R=$S_1(Ac)_8$, R′=H
5: R=$S_1(Me)_8$, R′=H
6: R=S_2, R′=H(=Pb)
7: R=$S_2(Ac)_{10}$, R′=H
8: R=$S_2(Me)_{10}$, R′=H
12: R=S_1, R′=OH
13: R=S_2, R′=OH
14: R=S_3, R′=OH
15: R=$S_3(Ac)_6$, R′=OH
16: R=S_3, R′=H
17: R=$S_3(Ac)_6$, R′=H
18: R=S_4, R′=OH
19: R=S_4, R′=H

S_1 :



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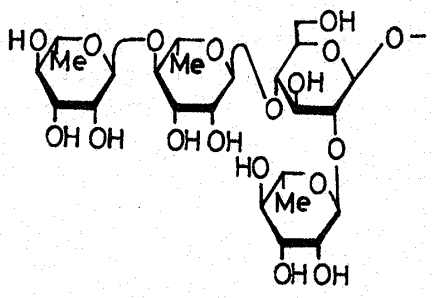
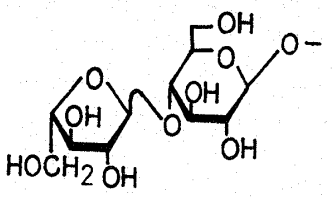
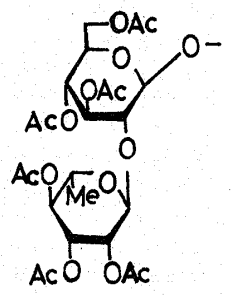


S_3 :



S_4 :



pentanoate-D-glucopyranoside tetraacetate (**11**)[4,5b] as a colorless oil. The structures of **9**, **10** and **11** were supported by their IR, mass (MS) and ^{1}H NMR spectra.

Consequently **1** and **2** can be defined as (22*ξ*, 25*R*)-22-methoxyfurost-5-ene-3,26-diol 26-*O*-*β*-D-glucopyranosides of 3-*O*-*α*-L-arabinofuranosyl-(1→4)-[*α*-L-rhamnopyranosyl-(1→2)]-*β*-D-glucopyranosyl and 3-*O*-*α*-L-rhamnopyranosyl-(1→4)-*α*-L-rhamnopyranosyl-(1→4)-[*α*-L-rhamnopyranosyl-(1→2)]-*β*-D-glucopyranosyl, respectively.

Next, **12**, colorless needles, mp 274—278°C, $[\alpha]_D$ −108.9°, showed absorptions due to hydroxyl (3600—3200 cm^{-1}) and spiroketal side chain (980, 920, 900 and 890) moieties. The mass spectrum of **12** exhibited characteristic peaks,[6] *m/z* 412 ($C_{27}H_{40}O_3^+$), 155 ($C_9H_{15}O_2^+$), 153 ($C_9H_{13}O_2^+$) and 126 ($C_8H_{14}O^+$), derived from pennogenin. Moreover, the ^{13}C NMR spectrum of **12** revealed signals attributable to the sugar moiety superimposed on those of Pa (**3**), as described in Experimental. Accordingly, **12** was deduced to be pennogenin 3-*O*-*α*-L-arabinofuranosyl-(1→4)-[*α*-L-rhamnopyranosyl-(1→2)]-*β*-D-glucopyranoside, whose sugar moiety is identical with that of Pa (**3**).

13, colorless needles, mp 224—228°C (dec.), was identical (mp, TLC, IR) with Tg,[7] pennogenin tetraglycoside, having the same sugar residue as Pb (**6**) previously isolated from the fresh underground parts of *Trillium kamtschaticum* Pall.

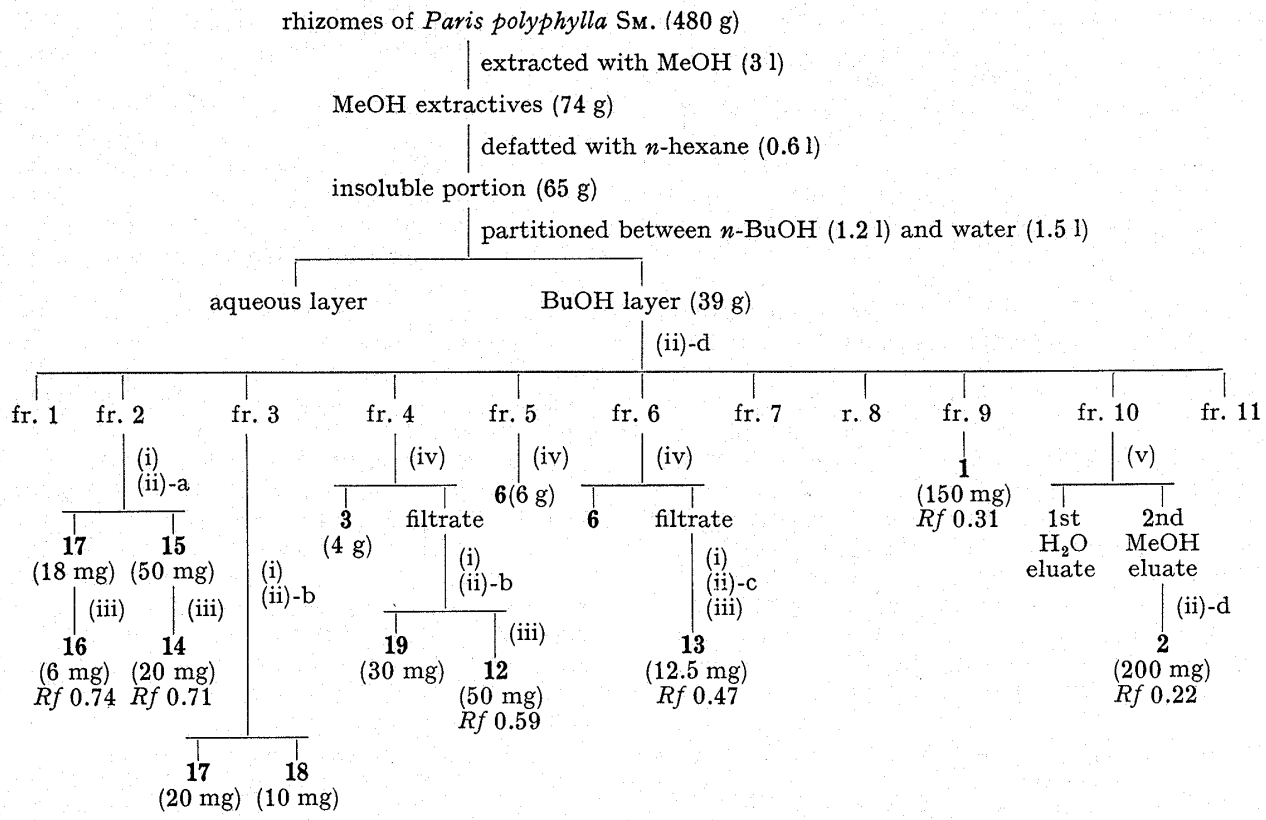
14, an amorphous powder, $[\alpha]_D$ −98.0°, and **16**, colorless needles, mp 239—242°C, were derived into the corresponding acetates, **15**, colorless needles, mp 172.5—176°C, $[\alpha]_D$ −44.6°, and **17**, colorless needles, mp 174—176°C, $[\alpha]_D$ −37.4°, respectively. Both acetates, **15** and **17**, showed peaks derived from the terminal peracetylated-pentosyl hexosyl cation (*m/z* 547)[6a] and -pentosyl cation (*m/z* 259)[6a] in the mass spectra. Furthermore, a comparison of the signals ascribable to the sugar moieties in the ^{13}C NMR spectra of **15** and **17** indicated that the moieties were almost identical. The hexamethyl ether derived from **14**, on acid hydrolysis with 2 N HCl–MeOH, gave two methylglycosides of 2,3,5-tri-*O*-methyl arabinofuranose and 2,3,6-tri-*O*-methyl glucopyranose besides aglycone derivatives (bethogenin and kryptogenin).[8] Since the mass spectra of **14** and **16** showed peaks at *m/z* [412, 155, 153, 126][6] and [396($C_{27}H_{40}O_2^+$), 282 ($C_{21}H_{30}^+$), 139 ($C_9H_{15}O^+$)][9] characteristic of pennogenin and diosgenin, respectively, and the chemical shifts of the signals due to the aglycones in the ^{13}C NMR spectra of **15** and **17** were in good agreement with those of pennogenin and diosgenin, respectively, **14** and **16** are pennogenin- and diosgenin-glycosides, respectively, having the same sugar moiety, 3-*O*-*α*-L-arabinofuranosyl-(1→4)-*β*-D-glucopyranoside, and correspond to prosapogenins of **12** and **3**, respectively.

18, colorless needles, mp 193—195°C, $[\alpha]_D$ −52.0°, and **19**, colorless needles, mp 203—207°C, $[\alpha]_D$ −60.0°, isolated after acetylation, were identical (mp, $[\alpha]_D$, TLC, MS) with Tb hexaacetate[7] and Ta hexaacetate,[7] respectively, previously obtained from the fresh underground parts of *T. kamtschaticum* Pall.

Experimental

All melting points were determined on a Yanagimoto micro melting point apparatus and are uncorrected. Optical rotations were measured with a Union Giken PM-201 automatic digital polarimeter at 20—25°C. IR and UV spectra were obtained with Hitachi IR-215 and Hitachi UV-124 machines, respectively. ^{13}C NMR and ^{1}H NMR spectra were taken with JEOL JNM-FX-90Q (22.5 MHz) and JEOL JNM-PS-100 (100 MHz) spectrometers, respectively. Chemical shifts are given on a *δ* (ppm) scale with tetramethylsilane as an internal standard. Mass spectra were taken with a JEOL JMS D-300 machine. Gas liquid chromatography (GLC) was run on a Shimadzu GC-6A unit with a flame ionization detector using a glass column (3 mm × 2.0 m) packed with neopentylglycol succinate. Paper partition chromatography (PPC) for sugar was conducted on Toyo Roshi No. 50 paper using the upper layer of *n*-BuOH–pyridine–water (6 : 2 : 3) + pyridine (1) as a solvent and aniline phthalate as a staining agent. TLC was performed on precoated silica gel plates 0.25 mm thick (Kieselgel 60, Merck) and detection was achieved by spraying 10% H_2SO_4 followed by heating. Column chromatography was performed on Kieselgel 60 (Merck, 70—230 mesh), Sephadex LH-20 (Pharmacia Fine Chem.) and Amberlite XAD-2 (Organo).

Extraction and Isolation of Steroid Saponins——Commercial dried rhizomes (480 g) purchased in a market



(i) acetylation with Ac_2O–pyridine
(ii) column chromatography on silica gel
using a. benzene–EtOH (30: 1)
b. *n*-hexane–AcOEt (1: 1)
c. benzene–acetone (1: 1)
d. $CHCl_3$–MeOH–water (7: 2: 0.2)
(iii) treated with 3% KOH–MeOH
(iv) crystallized from MeOH
(v) passed through Amberlite XAD-2
Rf values on TLC are for $CHCl_3$–MeOH–water (7: 3: 0.5) as a solvent.

Chart 1

in China were extracted with refluxing MeOH (3 l), and the extract was concentrated under reduced pressure to give a residue (74 g), which was treated as shown in Chart 1.

1——An amorphous powder (mp 209—212°C), $[\alpha]_D$ −135.6° (c=0.45, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3600—3300 (OH). Ehrlich reagent: positive. *Anal.* Calcd for $C_{50}H_{84}O_{22}\cdot H_2O$: C, 56.91; H, 8.11. Found: C, 56.51; H, 7.92. ^1H NMR (d_5-pyridine) δ (ppm): 3.24 (OMe).

Enzymic Hydrolysis of 1——A solution of **1** (60 mg) in dist. water (5 ml) was incubated with almond emulsin (Sigma Chem. Co.) (20 mg) at 37°C for 8 h, then the reaction mixture was evaporated to dryness *in vacuo* to give a residue, to which MeOH was added. The mixture was filtered. The MeOH solution was evaporated to dryness to give a residue, which was subjected to Sephadex LH-20 column chromatography using MeOH as an eluent to give a glycoside (**3**), colorless needles (from MeOH), mp 274—276°C, $[\alpha]_D$ −133.0° (c=0.56, MeOH), identical with Pa (30 mg), and a sugar identical with D-glucose (12 mg), *Rf* 0.46 (on PPC), $[\alpha]_D$ +59.5° (c=0.51, water). **3**: *Anal.* Calcd for $C_{44}H_{72}O_{16}\cdot H_2O$: C, 60.39; H, 8.52. Found: C, 60.37; H, 8.45. ^{13}C NMR (in d_5-pyridine) δ (ppm): aglycone (diosgenin);[10] 37.4, 30.0, 77.6, 40.4, 140.8, 121.6, 32.2, 31.6, 50.3, 37.0, 21.0, 39.8, 40.4, 56.6, 32.1, 81.1, 62.4, 16.3, 19.3, 41.9, 14.9, 109.3, 31.6, 29.1, 30.4, 66.8, 17.2 (C_{1-27}), glucose moiety; 100.0, 78.2, 77.2, 76.2, 77.6, 62.7 ($C_{1'-6'}$), rhamnose moiety; 101.6, 72.4, 71.9, 73.6, 69.2, 18.3 ($C_{1''-6''}$), arabinofuranose moiety; 109.4, 82.5, 77.6, 86.1, 61.2 ($C_{1'''-5'''}$).

Octaacetate (4) of 3——**3** (85 mg) was acetylated with Ac_2O–pyridine (each 4 ml) at 80°C for 1 h to give a peracetate (**4**), colorless needles (from MeOH, 68 mg), mp 122—128°C, $[\alpha]_D$ −100.0° (c=0.50, $CHCl_3$). MS m/z: 777, 396 ($C_{27}H_{40}O_2^+$), 282 ($C_{21}H_{30}^+$), 273 ($C_{12}H_{17}O_7^+$), 259 ($C_{11}H_{15}O_7^+$), 253, 139 ($C_9H_{15}O_2^+$), 115 ($C_6H_{11}O_2^+$).

Octamethylether (5) of 3——**3** (50 mg) was methylated by Hakomori's method[11] to give a permethylate (**5**), colorless needles (from MeOH, 26 mg), mp 160—162°C, $[\alpha]_D$ −92.0° (c=0.50, $CHCl_3$). MS m/z: 553, 396, 282, 253, 189 ($C_9H_{17}O_4^+$), 175 ($C_8H_{15}O_4^+$), 139, 115. ^1H NMR (in $CDCl_3$) δ (ppm): 0.80 (3H, s, 18-CH_3),

0.85 (3H, d, J=6 Hz, 27-CH_3), 0.98 (3H, d, J=6 Hz, 21-CH_3), 1.02 (3H, s, 19-CH_3), 3.53—3.70 (OMe), 4.33 (1H, d, J=7 Hz, glucosyl anomeric proton), 5.17 (1H, s, rhamnosyl anomeric proton), 5.22 (1H, broad, s, arabinofuranosyl anomeric proton).

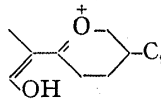
Oxidation Products (9 and 11) of 1——The peracetate (72 mg) of **1** in Ac_2O (10 ml) was refluxed for 45 min, then the solution was cooled and EtOH was added. The whole was evaporated to dryness *in vacuo*, yielding a pale brown residue (60 mg) which included the acetyl $\Delta^{20(22)}$ compound of **1**. AcOH (1.6 ml) and AcONa (32 mg) were added to the residue, and the resulting solution was cooled to 12°C. CrO_3 solution (65 mg in 0.5 ml of 40% AcOH) was added with stirring, and the mixture was allowed to stand for 1 h at room temperature. The reaction mixture was diluted with water (50 ml) and shaken with $CHCl_3$ to extract oxidation products. Evaporation yielded a syrup (78 mg), which was mixed with an aqueous solution (1.2ml) containing KOH (165 mg). After further addition of *t*-BuOH (4 ml), the mixture was incubated at 30°C for 3.5 h. The *t*-BuOH was distilled off, and the reaction mixture was extracted with *n*-BuOH. The extract was evaporated to dryness *in vacuo* to give a residue, which was chromatographed on silica gel using $CHCl_3$–MeOH–H_2O (7 : 3 : 0.2) to give a pregnenolone glycoside (**9**, sugar moiety=S_1), a white powder (mp 214—216°, 25 mg), $[\alpha]_D$ −42.0° (c=0.50, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3600—3200 (OH), 1650, 1580 (α,β-unsaturated ketone). UV λ_{max}^{EtOH} nm: 239 (ε=9800). ^1H NMR (in d_5-pyridine) δ (ppm): 0.95 (3H, s, 19-CH_3), 1.05 (3H, s, 18-CH_3), 1.75 (3H, d, J=6 Hz, rhamnosyl 5-CH_3), 2.25 (3H, s, 21-CH_3), 5.80 (1H, d, J=9 Hz, glucosyl anomeric proton), 6.18 (1H, s, arabinofuranosyl anomeric proton), 6.63 (1H, br s, rhamnosyl anomeric proton). The aqueous layer, after being adjusted to pH 3.0 with 3 N HCl, was shaken in turn with *n*-BuOH and $CHCl_3$, neutralized with 2 N NaOH, and concentrated to afford a salt mixture. The salt were acetylated in a usual way and its acetate was taken up in MeOH. This solution was treated with diazomethane in ether. After removal of the solvent, the residue was chromatographed on a silica gel column with *n*-hexane–acetone (1 : 1) to give methyl 4(*R*)-methyl-5-hydroxypentanoate β-D-glucopyranoside tetraacetate (**11**), a colorless oil (11 mg), $[\alpha]_D$ −16.8° (c=0.42, $CHCl_3$). MS m/z: 331 ($C_{14}H_{19}O_9^+$), 243, 242, 200, 169, 157, 145, 140, 129 ($C_7H_{13}O_2^+$), 115, 109, 103, 98, 97 ($C_6H_9O^+$). ^1H NMR (in $CDCl_3$) δ (ppm): 0.90 (3H, d, J=7 Hz, *sec*. CH_3), 2.00—2.08 (12H, 4×OAc), 3.62 (3H, s, 1×OMe), 4.45 (1H, d, J=7 Hz, glucosyl anomeric proton).

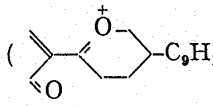
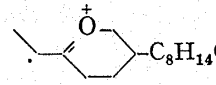
2——An amorphous powder (mp 174—177°C), $[\alpha]_D$ −87.0° (c=1.00, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3600—3200 (OH). Ehrlich reagent: positive. *Anal.* Calcd for $C_{57}H_{90}O_{25}\cdot 3H_2O$: C, 55.40; H, 7.99. Found: C, 55.35; H, 8.10. ^1H NMR (in d_5-pyridine) δ (ppm): 3.21 (OMe). Enzymic hydrolysis of **2** (42 mg) with almond emulsin (15 mg) in the manner described for **1** and **2** yielded a glycoside (**6**), colorless needles (12 mg), mp 203—206°C, $[\alpha]_D$ −128.0° (c=0.50, MeOH), identical with Pb, and a sugar (11 mg) identical with D-glucose, *Rf* 0.45 (on PPC), $[\alpha]_D$ +60.0° (c=0.30, water). **6**: *Anal.* Calcd for $C_{51}H_{82}O_{20}\cdot 3H_2O$: C, 57.29; H, 8.30. Found: C, 57.49; H, 8.00. ^{13}C NMR (in d_5-pyridine) δ (ppm): aglycone; 37.4, 30.0, 77.2, 38.8, 140.7, 121.6 32.2, 31.6, 50.2, 37.0, 21.0, 39.8, 40.4, 56.6, 32.0, 81.0, 62.6, 16.3, 19.3, 41.8, 14.9, 109.3, 31.6, 29.0, 30.4, 66.8, 17.2 (C_{1-27}), glucose moiety; 100.1, 80.0, 76.4, 77.6, 78.1, 61.1 ($C_{1'-6'}$), rhamnose moiety attached to C-2 of glucose; 102.0, 72.4, 71.9, 73.6, 69.3, 18.4 ($C_{1''-6''}$), rhamnose moiety attached to C-4 of glucose; 102.7, 72.4, 70.0, 78.1, 68.2, 18.3 ($C_{1'''-6'''}$), rhamnose moiety attached to C-4 of rhamnose; 102.0, 72.4, 71.9, 73.6, 69.3, 18.1 ($C_{1''''-6''''}$).

Decaacetate (7) of 6——**6** (30 mg) was acetylated with Ac_2O–pyridine (each 2 ml) at 80°C for 1 h to give **7** (30 mg) as colorless needles (from MeOH, 21 mg), mp 148—153°C, $[\alpha]_D$ −74.0° (c=1.00, $CHCl_3$). MS m/z: 503 ($C_{22}H_{31}O_{13}^+$), 414, 396, 282, 273, 253.

Decamethyl Ether (8) of 6——**6** (50 mg) was methylated by Hakomori's method to give **8** (10 mg) as colorless needles (from MeOH, 12 mg), mp 135—139°C, $[\alpha]_D$ −110.0° (c=0.50, $CHCl_3$). MS m/z: 1154 (M^+), 414, 396, 363 ($C_{17}H_{31}O_8^+$), 282, 253, 189 ($C_9H_{17}O_4^+$). ^1H NMR (in $CDCl_3$) δ (ppm): 0.79 (3H, s, 18-CH_3), 0.85 (3H, d, J=7 Hz, 27-CH_3), 0.97 (3H, d, J=6 Hz, 21-CH_3), 1.03 (3H, s, 19-CH_3), 1.20, 1.22, 1.24 (each 3H, d, J=6 Hz, rhamnosyl 5-CH_3), 3.35—3.75 (OMe), 4.38 (1H, d, J=7 Hz, glucosyl anomeric proton), 5.02 (1H, s, rhamnosyl anomeric proton), 5.20 (2H, s, 2×rhamnosyl anomeric proton).

Oxidation Products (10 and 11) of 2——The peracetate (150 mg) of **2** was oxidized in the manner described for **1** to give a pregnenolone glycoside (**10**, sugar moiety=S_2), colorless needles (from MeOH, 62 mg) mp 234—237°C, $[\alpha]_D$ −117.7° (c=0.51, MeOH) and methyl 4(*R*)-methyl-5-hydroxypentanoate β-D-glucopyranoside tetraacetate (**11**), a colorless oil (30 mg), $[\alpha]_D$ −18.2° (c=0.92, $CHCl_3$). **10**: IR ν_{max}^{KBr} cm^{-1}: 3600—3000 (OH), 1650, 1580 (α,β-unsaturated ketone). UV λ_{max}^{EtOH} nm: 235 (ε=10800). ^1H NMR (in d_5-pyridine) δ (ppm): 0.95 (3H, s, 19-CH_3), 1.06 (3H, s, 18-CH_3), 1.60 (6H, d, J=6 Hz, 2×rhamnosyl 5-CH_3), 1.77 (3H, d, J=6 Hz, rhamnosyl 5-CH_3), 2.24 (3H, s, 21-CH_3), 5.75 (1H, d, J=7 Hz, glucosyl anomeric proton), 6.22, 6.35, 6.56 (each 1H, s, rhamnosyl anomeric proton). **11**: Mass and ^1H NMR (in $CDCl_3$) spectra were identical with those of the product of oxidation of **1**.

12——Colorless needles (from MeOH), mp 274—278°C (dec.), $[\alpha]_D$ −108.9° (c=0.73, MeOH). $[M]_D$ −949.6°, $\Delta[M]_D$(**12**−Tb) −80.2°, $\Delta[M]_D$(**12**−**14**) −257.7°. IR ν_{max}^{KBr} cm^{-1}: 3600—3200 (OH), 980, 920, 900, 890 (intensity 900>920, 25*R*-spiroketal side chain). *Anal.* Calcd for $C_{44}H_{72}O_{17}\cdot H_2O$: C, 59.31; H, 8.37. Found. C, 58.96; H, 8.27. MS m/z: 430 ($C_{27}H_{42}O_4^+$), 412 ($C_{27}H_{40}O_3^+$), 155 (), 153

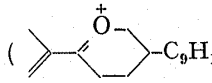
( -$C_9H_{13}O_2^+$), 126 ( -$C_8H_{14}O^+$). ^{13}C NMR (in d_5-pyridine) δ (ppm): aglycone (pennogenin);[12] 37.4, 30.0, 77.6, 38.8, 140.7, 121.6, 32.2, 31.6, 50.1, 37.0, 20.8, 37.0, 45.0, 52.9, 32.2, 89.9, 90.1, 17.1, 19.3, 44.7, 9.3, 109.8, 32.0, 28.6, 30.3, 66.7, 17.2 (C_{1-27}), glucose moiety; 100.0, 78.2, 77.2, 76.2, 77.6, 62.4 ($C_{1'-6'}$), rhamnose moiety; 101.6, 72.4, 71.9, 73.6, 69.2, 18.3 ($C_{1''-6''}$), arabinofuranose moiety; 109.4, 82.4, 77.6, 86.1, 61.3 ($C_{1'''-6'''}$).

13——Colorless needles (from MeOH), mp 224—228°C (dec.), $[\alpha]_D$ −132.0° (c=0.50, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3600—3200 (OH), 980, 920, 900, 890 (intensity 900—920, 25*R*-spiroketal side chain). *Anal.* Calcd for $C_{51}H_{82}O_{21}\cdot 3H_2O$: C, 56.44; H, 8.17. Found: C, 56.21; H, 8.08. Identical with Tg in terms of mp, *Rf* value on TLC and IR.

14——A white powder (mp 220—223°C), $[\alpha]_D$ −98.0° (c=0.50, MeOH), $[M]_D$ −691.9°. IR ν_{max}^{KBr} cm^{-1}: 3600—3100 (OH), 980, 915, 898, 890 (intensity 898>915, 25*R*-spiroketal side chain). MS *m/z*: 430, 412, 394, 155, 153, 127, 126. *Anal.* Calcd for $C_{38}H_{60}O_{12}$: C, 64.38; H, 8.53. Found: C, 64.12; H, 8.41.

Hexaacetate (15) of 14——Colorless needles (from MeOH), mp 172.5—176°C, $[\alpha]_D$ −44.6° (c=0.92, $CHCl_3$). MS (*m/z*): 684, 547, 412, 394, 282, 259, 157, 139, 115, 109. ^{13}C NMR (in $CDCl_3$) δ (ppm): aglycone (pennogenin); 37.2, 29.5, 76.6, 39.0, 140.5, 121.7, 32.0, 31.6, 49.8, 36.8, 20.6, 36.8, 43.8, 52.9, 31.6, 91.0, 90.1, 17.1, 19.3, 44.6, 8.0, 110.0, 31.2, 28.1, 30.1, 66.8, 17.2 (C_{1-27}), sugar moiety; 107.4, 99.5, 81.6, 81.1, 79.9, 76.6, 73.5, 72.7, 72.1, 63.1, 62.4.

Methanolysis of Hexamethyl Ether of 14——**14** (10 mg) was methylated by Hakomori's method to give a permethyl ether (6 mg) of **14**, a white solid, *Rf* 0.41 (on TLC, solv. *n*-hexane–acetone (2: 1)), MS *m/z*: 412, 394, 376, 175, 101. The hexamethyl ether (4 mg) of **14** was subjected to methanolysis under reflux with 1 N HCl–MeOH (5 ml) for 2 h to give bethogenin and kryptogenin (*Rf* 0.62 and 0.12, respectively, on TLC, solv. *n*-hexane–AcOEt (1: 1)) and methylglycosides, which were examined by GLC comparison with authentic samples. GLC, column temp. 108°C, N_2 1.25 kg/cm^2; t_R, 5.5, 7.4 min (methyl 2,3,5-tri-*O*-methyl α- and β-arabinofuranoside): column temp. 180°C, t_R, 1.60, 2.05 min (methyl 2,3,6-tri-*O*-methyl α- and β-glucopyranoside).

16——Colorless needles (from MeOH), mp 239—242°C. IR ν_{max}^{KBr} cm^{-1}: 3600—3100 (OH), 980, 918, 8.98, 860 (intensity 898>918, 25*R*-spiroketal side chain), *Anal.* Calcd for $C_{38}H_{60}O_{13}\cdot H_2O$: C, 61.43; H, 8.41. Found: C, 61.56; H, 8.38. MS *m/z*: 396 ($C_{27}H_{40}O_2^+$), 282 ($C_{21}H_{30}^+$), 253, 139 ( -$C_9H_{15}O^+$).

Hexaacetate (17) of 16——Colorless needles (from MeOH), mp 174—176°C, $[\alpha]_D$ −37.4° (c=0.54, $CHCl_3$), MS (*m/z*): 684, 547, 396, 282, 259, 253, 139, 115. ^{13}C NMR (in $CDCl_3$) δ (ppm): aglycone (diosgenin); 37.2, 30.7, 76.6, 39.0, 140.5, 121.8, 32.1, 31.5, 50.2, 36.9, 21.0, 39.8, 40.3, 56.6, 32.0, 80.8, 62.3, 16.3, 19.4, 41.7, 14.5, 109.2, 31.5, 28.9, 29.7, 66.9, 17.1 (C_{1-27}), sugar moiety; 107.4, 99.5, 81.7, 81.2, 79.8, 76.0, 73.5, 72.8, 72.1, 63.2, 62.3.

18——Colorless needles (from MeOH), mp 193—195°C, $[\alpha]_D$ −52.0° (c=0.50, $CHCl_3$), MS (*m/z*): 561, 412, 394, 273, 153, 139, 126, 111.

19——Colorless needles (from MeOH), mp 203—207°C, $[\alpha]_D$ −60.0° (c=1.00, $CHCl_3$), IR ν_{max}^{KBr} cm^{-1}: 1750 (OAc), 980, 920, 900, 860 (intensity 900>920, 25*R*-spiroketal side chain), MS (*m/z*): 684, 561, 414, 396, 282, 273, 253, 139. *Anal.* Calcd for $C_{51}H_{74}O_{18}$: C, 62.81; H, 7.65. Found: C, 62.50; H, 7.60.

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