Saponins from Vietnamese Ginseng, *Panax vietnamensis* HA *et* GRUSHV. Collected in Central Vietnam. II

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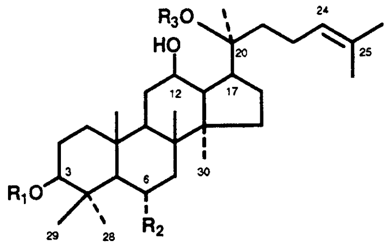
Further investigation on the saponin composition of rhizomes and roots of *Panax vietnamensis* HA *et* GRUSHV. has resulted in the isolation and structural elucidation of seven new dammarane saponins named vina-ginsenosides-R3 (12), -R4 (11), -R5 (16), -R6 (17), -R7 (6), -R8 (20), -R9 (22), together with the identification of six known saponins including 20-gluco-ginsenoside-Rf (10), ginsenoside-Rc (4), notoginsenoside-R6 (9), quinquenoside-R_1 (5), gypenoside XVII (2) and majoroside F1 (21). The structures of the novel saponins were established on the basis of chemical and spectral evidence. Vina-ginsenoside-R3 is the first naturally occurring glycoside of dammarenediol II, while vina-ginsenosides-R5 and -R6, two ocotillol-type saponins, are two other examples of saponins having the rare α-glucosyl linkage.

Keywords Vietnamese Ginseng; *Panax vietnamensis*; dammarane saponin; ginsenoside; vina-ginsenoside-R3, -R4, -R5, -R6, -R7, -R8, -R9; dammarenediol saponin

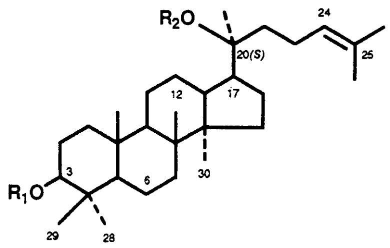
Vietnamese Ginseng, *Panax vietnamensis* HA *et* GRUSHV., is a new *Panax* species discovered in Central Vietnam in 1973, which has been used in the country as a tonic like the well-known Ginseng, *Panax ginseng* C. A. MEYER. It also showed an effectiveness in the treatment of sore throat, cough, *etc.* In our preceding paper,[1)] the isolation and identification of sixteen known saponins and two novel acetylated ocotillol-type dammarane saponins named vina-ginsenosides-R1 and -R2 from rhizomes and roots of *P. vietnamensis* was reported. We now deal with the structural elucidation of six other known saponins and seven new dammarane glycosides, vina-ginsenosides-R3, -R4, -R5, -R6, -R7, -R8, -R9 which were isolated from the same material.

The remaining crude saponin fractions from the separation and isolation procedure described previously[1)] were further subjected to column chromatography and preparative high performance liquid chromatography (HPLC) to afford six other known saponins along with seven new compounds. By comparison of optical rotation, thin layer chromatographic (TLC) behavior, ^{1}H- and ^{13}C-nuclear magnetic resonance (NMR) spectra, and mass spectrum (MS, as the trimethylsilyl ether) with those of a corresponding authentic sample or reported data, the known saponins were identified as 20-gluco-ginsenoside-Rf[2)] (**10**, 0.01% yield), ginsenoside-Rc[3)] (**4**, 0.013% yield), notoginsenoside-R6[4)] (**9**, 0.01% yield), quinquenoside-R_1[5)] (**5**, 0.012% yield), gypenoside XVII[6)] (**2**, 0.036% yield) and majoroside F1[7)] (**21**, 0.003% yield). All these known compounds have been isolated from other *Panax* spp., except for gypenoside XVII, which was first found in *Gynostemma pentaphyllum* MAKINO, Cucurbitaceae by Takemoto *et al.*[6)]

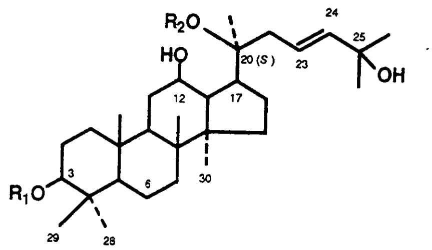
A new saponin, vina-ginsenoside-R3 (**12**), $C_{48}H_{82}O_{17}$, was obtained in yield of 0.009%. On acid hydrolysis, **12** gave glucose as the only sugar constituent. The electron impact mass spectrum (EI-MS) of the trimethylsilyl ether (TMSi) of **12** showed fragment ions at *m/z* 451 [terminal glucosyl $(TMSi)_4$] and 829 [glucosyl-glucosyl $(TMSi)_7$]. The ^{1}H-NMR spectrum of **12** displayed three anomeric protons at δ 4.96 (d, J=7.3 Hz), 5.12 (d, J=8.0 Hz) and 5.40 (d, J=7.3 Hz), which were found to correspond to the anomeric carbon signals at δ 105.1, 98.6 and 106.0, respectively, from a ^{13}C–^{1}H correlated spectroscopy (^{13}C-^{1}H COSY) experiment. The coupling constants of the anomeric protons, as well as the chemical shifts of the sugar carbon signals indicated that all the sugars are β-glucopyranosyl units. On enzymatic hydrolysis with crude hesperidinase, **12** gave an aglycone (**12a**) which was proved to be 20(*S*)-dammar-24-ene-3β, 20-diol (dammarenediol II)[8,9)] by physicochemical data in comparison with those of an authentic sample. Inspection of the spectral data revealed the glycosylation shifts for C-2 ($\Delta\delta$ −1.5 ppm) and C-3 ($\Delta\delta$+11.4 ppm); C-20 ($\Delta\delta$+8.2 ppm) and C-21 ($\Delta\delta$−3.7 ppm) on going from **12** to **12a**, demonstrating that **12** is a bisdesmoside of **12a** having two sugar linkages at the 3- and 20-hydroxyl groups (see Table I). When the ^{13}C-NMR spectrum of **12** and that of ginsenoside-Rd (**3**) were compared, carbon resonances assignable to the sugar moieties appeared at almost the same positions. It was reported that the glycosyl linkage at the C-20 hydroxyl group of dammarane saponins is unstable and readily hydrolyzed even under mild condition, yielding a C-20 epimeric mixture of the corresponding prosapogenin or sapogenin.[10)] On partial hydrolysis with 50% aqueous acetic acid, **12** afforded a prosapogenin (**12b**). The EI-MS of the trimethylsilyl ether of **12b** showed fragment ions at *m/z* 451 [terminal glucosyl $(TMSi)_4$] and 829 [glucosyl-glucosyl $(TMSi)_7$], indicating the presence of a glucosyl-glucosyl unit at C-3 which remained unhydrolyzed after the treatment. Further, the ^{13}C-NMR spectrum revealed that **12b** is a C-20 epimeric mixture with a β-sophorosyl residue attached to the 3-hydroxyl group (see Tables I and II). These results led to the formulation of **12** as 3-*O*-[β-D-glucopyranosyl-(1→2)-β-D-glucopyranosyl]-20-*O*-β-D-glucopyranosyl



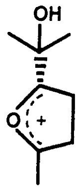
		R_1	R_2	R_3
1		-H	-H	-H
2		-Glc	-H	-Glc^6-Glc
3		-Glc^2-Glc	-H	-Glc
4		-Glc^2-Glc	-H	-Glc^6-Ara(f)
5		-Glc^2-Glc^6-Ac	-H	-Glc^6-Glc
6		-Glc^2-Glc^2-Xyl	-H	-Glc
6a	20*(R)*+20*(S)*	-Glc^2-Glc^2-Xyl	-H	-H
7		-Glc^2-Glc^2-Xyl	-H	-Glc^6-Glc
8		-H	-OH	-H
9		-H	-O-Glc	-Glc^6-α Glc
10		-H	-O-Glc^2-Glc	-Glc
11		-Glc^2-Glc	-OH	-Glc
11a	20*(R)*+20*(S)*	-Glc^2-Glc	-OH	-H



		R_1	R_2
12		-Glc^2-Glc	-Glc
12a		-H	-H
12b	20*(R)*+20*(S)*	-Glc^2-Glc	-H



	R_1	R_2
19	- Glc	- Glc
20	-Glc^2- Glc	- Glc



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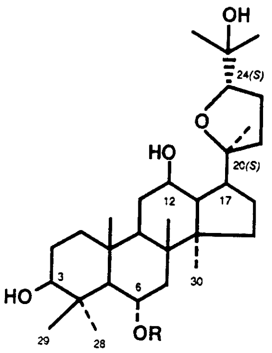
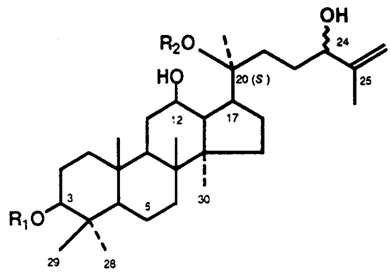


	R
14	-H
15	-Glc^2-Xyl
16	-Glc^2-Xyl^4-α Glc
17	-Glc^2— Xyl, ^{6}α Glc



		R_1	R_2
21	24 (*R*)	-Glc^2- Glc	- Glc
22	24 (*S*)	-Glc^2-Glc	- Glc

Glc; β-D-glucopyranosyl
α Glc; α-glucopyranosyl
Ac; acetyl
Xyl; β-D-xylopyranosyl
Ara(f); α-L-arabinofuranosyl

Chart 1

20(*S*)-dammar-24-ene-3*β*,20-diol. Dammarenediol II was particularly found in resinous exudates (dammar) from trees of the family Dipterocarpaceae, but **12** is its first naturally occurring glycoside. On the other hand, dammarenediol II has turned out to be the least oxygenated dammarane-type aglycone yet found in *Panax* plants.

Vina-ginsenoside-R4 (**11**), $C_{48}H_{82}O_{19}$, was isolated in yield of 0.004%. From the acid hydrolysate of **11**, glucose was obtained. The ^{1}H- and ^{13}C-NMR spectra showed the presence of three *β*-glucopyranosyl units. In the EI-MS of the trimethylsilyl ether of **11**, fragment ions at *m/z* 451 [terminal glucosyl (TMSi)$_4$] and 829 [glucosyl-glucosyl (TMSi)$_7$] were observed. Comparison of the ^{13}C-NMR spectrum of **11** with that of 20(*S*)-protopanaxatriol[8] (**8**) showed the glycosylation shifts for C-2 ($\Delta\delta-1.4$ ppm) and C-3 ($\Delta\delta+11.1$ ppm); C-20 ($\Delta\delta+10.4$ ppm) and C-21 ($\Delta\delta-4.6$ ppm) on going from **11** to **8** (see Table I). This demonstrated that **11** is a bisdesmoside of **8** whose hydroxyl groups at C-3 and C-20 are glycosylated. In addition, all the sugar carbon signals of **11** were essentially in harmony

TABLE I. ^{13}C-NMR Chemical Shifts of Aglycone Moieties (in C_5D_5N, δ)

	1	3	6	6a	7	12	12a	12b	19	20	21	22	8	11	11a	14	16	17
C-1	39.5	39.2	39.3	39.1	39.2	39.3	39.5	39.3	39.1	39.1	39.1	39.2	39.3	39.1	39.3	39.5	39.6	39.6
C-2	28.2	26.6	26.7	26.7	26.6	26.8	28.3	26.9	26.5	26.7	26.7	26.7	28.0	26.6	25.8	28.1	27.8	27.8
C-3	77.9	88.9	89.2	88.9	88.9	89.4	78.0	89.0	88.8	89.0	89.0	89.0	78.3	89.4	89.4	78.4	78.0	78.3
C-4	39.5	39.6	39.8	39.7	39.7	39.7	39.5	39.6	39.6	39.3	39.6	39.7	40.2	40.6	40.6	40.3	40.2	40.0
C-5	56.3	56.3	56.6	56.4	56.4	56.4	56.5	56.6	56.4	56.4	56.3	56.4	61.7	61.7	61.7	61.9	61.4	61.3
C-6	18.7	18.4	18.5	18.4	18.0	18.4	18.8	18.4	18.4	18.4	18.2	18.4	67.6	67.5	67.5	67.7	79.4	79.4
C-7	35.2	35.1	35.2	35.2	35.3	35.7	35.7	35.8	35.2	35.1	35.1	35.1	47.4	47.5	47.5	47.5	45.0	46.0
C-8	40.0	39.9	40.1	40.0	40.0	40.3	40.7	40.3	40.0	40.0	39.9	40.4	41.1	41.1	41.1	41.2	41.1	41.3
C-9	50.4	50.1	50.3	50.3	50.2	51.0	51.1	51.0	50.0	50.1	50.1	50.2	50.1	49.8	50.0	50.2	50.2	50.2
C 10	37.3	36.8	37.0	36.9	36.9	36.9	37.4	37.1	36.9	36.9	36.8	36.9	39.3	38.8	38.8	39.3	39.6	39.6
C-11	32.0	30.8	30.8	31.3	30.0[a]	21.9	21.9	21.9	30.7	31.0	30.8	30.7	31.9	30.7	31.4	32.2	32.2	32.2
C-12	70.9	70.2	70.3	70.7	70.2	25.5	25.8	25.8	70.6	70.5	70.4	70.3	70.9	70.2	70.9	70.8	70.8	70.9
C-13	48.5	49.3	49.5	48.5 (49.2)	49.4	42.5	42.6	42.6	49.3	49.5	49.3	49.3	48.1	49.1	48.9 (49.6)	49.1	49.1	49.2
C-14	51.6	51.3	51.5	51.7	51.4	50.6	50.6	50.8	51.4	51.5	51.5	51.5	51.6	51.3	51.6	52.2	52.2	52.3
C-15	31.3	30.8	31.0	32.0	30.7[a]	31.5	31.7	31.8	30.7	30.8	31.0	30.8	31.3	30.8	29.9	32.6	32.5	32.6
C-16	26.8	26.6	26.7	26.8	26.6	28.0	28.1	28.0	26.5	26.4	26.7	26.7	26.8	26.6	27.0	25.8	25.7	25.7
C-17	54.7	51.6	51.9	54.7 (50.6)	51.4	48.4	50.3	50.1 (49.9)	52.3	52.4	52.3	52.0	54.6	51.6	54.7 (51.7)	49.5	49.4	49.6
C-18	16.2[b]	16.2[b]	16.2[b]	16.3[b]	16.3[b]	16.6[b]	16.5[b]	16.7[b]	16.2	16.6[b]	16.6[b]	16.6[b]	17.5[b]	17.5[b]	17.6[b]	17.8[b]	17.8[b]	17.8[b]
C-19	15.8[b]	15.8[b]	16.0[b]	15.8[b]	16.0[b]	16.5[b]	16.3[b]	16.5[b]	15.9	16.3[b]	16.2[b]	16.2[b]	17.4[b]	17.4[b]	17.4[b]	17.2[b]	17.1[b]	17.3[b]
C-20	72.9	83.2	83.4	72.9 (73.0)	83.4	82.2	74.0	73.8 (—)	83.2	83.4	83.2	83.5	72.9	83.3	72.9 (73.0)	87.0	87.0	87.1
C-21	26.9	22.4	22.5	27.0 (22.7)	22.4	21.6	25.3	25.5 (—)	23.2	23.1	22.8	22.9	26.9	22.3	26.6 (22.7)	26.9[a]	26.9[a]	27.0[a]
C-22	35.8	35.9	36.2	35.9 (43.2)	36.2	40.6	41.9	41.9 (—)	39.6	39.7	32.4	32.5	35.7	36.1	35.7 (43.5)	32.6	32.6	32.6
C-23	22.9	23.2	23.3	22.9	23.2	23.2	23.3	23.3	122.5	122.8	31.0	31.0	22.9	23.2	23.0	28.7	28.6	28.8
C-24	126.2	125.8	125.9	126.3	125.9	126.1	126.0	126.2	141.9	142.1	76.1	75.6	126.2	125.9	126.3	88.4	88.4	88.3
C-25	130.6	130.8	130.9	130.7	131.0	130.6	130.6	130.8	69.9	69.9	149.5[c]	149.5[c]	130.6	130.9	130.8	70.0	70.0	70.0
C-26	25.8	25.7	25.6	25.8	25.8	25.8	26.1	25.8	30.5	30.6	110.3	109.9	25.8	25.7	25.8	26.6[a]	26.5[a]	26.5[a]
C-27	17.6	17.7[b]	17.7[b]	17.6[b]	17.4[b]	17.9[b]	17.7	17.8[b]	30.5	30.6	18.4	18.5	17.7	17.7[b]	17.7[b]	29.0[a]	28.9[a]	29.0[a]
C-28	28.6	28.0	28.1	28.1	28.1	28.7	28.7	28.4	28.1	28.1	28.1	28.1	31.9	31.3	31.9	31.9	31.6	32.0
C-29	16.4[b]	16.5[b]	16.6[b]	16.6[b]	16.6[b]	15.7[b]	15.8[b]	16.0[b]	17.1	15.9[b]	15.8[b]	15.9[b]	16.4[b]	16.7[b]	16.8[b]	16.5[b]	16.7[b]	17.2[b]
C-30	17.0	17.3[b]	17.4[b]	17.0[b]	17.1[b]	16.8[b]	16.9	16.7[b]	17.1	17.1[b]	17.2[b]	17.3[b]	17.0	17.3[b]	17.4[b]	18.1[b]	17.8[b]	17.9[b]

a, b) Interchangeable values in each vertical column. *c*) Signal overlapping with solvent signals but can be detected when run in CD_3OD (see Experimental). Values in parentheses are those for the 20 (*R*) epimer. (—) Expected sub-peaks not observed due to the low yield of **12b**.

with those of **3** and **12**, giving rise to the presence of a sophorosyl unit at C-3. On heating with aqueous acetic acid, **11** afforded a prosapogenin (**11a**) which was proved to be a C-20 epimeric pair of the saponins containing a β-sophorosyl moiety at the 3-hydroxyl group of the aglycone (see Tables I and II). The fragment ions at m/z 451 and 829 in the EI-MS of the trimethylsilylated **11a**, which are typical for [terminal glucosyl $(TMSi)_4$] and [glucosyl-glucosyl $(TMSi)_7$] ion peaks, respectively, supported the NMR data. On the above evidence, the structure of **11** was established as 3-*O*-[β-D-glucopyranosyl-(1→2)-β-D-glucopyranosyl]-20-*O*-β-D-glucopyranosyl 20(*S*)-protopanaxatriol. Since all protopanaxatriol bisdesmosides isolated from *Panax* spp. so far contain sugar chains at C-6 and C-20, **11** is the first example of saponins of this type which possesses glycosyl linkages at C-3 and C-20 of the aglycone.

Vina-ginsenosides-R5 (**16**) and-R6 (**17**) have the same molecular formula of $C_{47}H_{80}O_{19}$ and were isolated as white powder in yields of 0.008% and 0.006%, respectively. Glucose and xylose were identified in their acid hydrolysates. The EI-MS of their acetylated derivatives (**16a** and **17a**) exhibited a strong ion at m/z 143 due to fragment **18**, which is characteristic of the hydroxyisopropyltetrahydrofuran ring of ocotillol-type triterpenes and their saponins. A comparison of the ^{13}C-NMR spectra of **16** and **17** with that of 20(*S*),24(*S*)-epoxydammarane-3β,6α,12β,25-tetrol (**14**)[11] showed that signals due to the aglycone carbons of **16** and **17** were consistent with those of **14** except for the signals of C-6 and C-7, where glycosylation shifts were observed. This indicated that **16** and **17** are monodesmosides of **14** whose glycosyl linkage must be located at the 6α-hydroxyl group.

The EI-MS of the trimethylsillyl ether of **16** showed fragment ions at m/z 451 [terminal glucosyl $(TMSi)_4$], 727 [glucosyl-xylosyl $(TMSi)_6$] and 1105 [glucosyl-xylosyl-glucosyl $(TMSi)_9$], indicating that the sugar residue is a linear glucosyl-xylosyl-glucosyl unit. The ^{13}C-NMR spectrum of **16** exhibited three sugar units whose anomeric carbon atoms resonated at δ 102.3 ($^1J_{CH}=167$ Hz), 103.5 ($^1J_{CH}=159$ Hz) and 104.3 ($^1J_{CH}=166$ Hz). A ^{13}C–^{1}H COSY experiment revealed the location of the corresponding anomeric protons at δ 5.66 (d, $J=3.9$ Hz), 4.94 (d, $J=6.4$ Hz), 5.68 (d, $J=7.1$ Hz), respectively. The coupling constants of the anomeric protons and the carbon chemical shifts of the latter two monosaccharides are typical for β-linked sugars, but those of the former suggested that it possesses an α anomeric configuration. The sugar sequence and the interglycosidic linkage were then determined by a detailed analysis of the spectral data of the acetate of **16** (**16a**).

With the aid of a ^{1}H–^{1}H COSY experiment, the assignment of the sugar protons of **16a** was achieved as shown in Table III. Inspection of the ^{1}H-NMR data disclosed that the constituent monosaccharides consist of one β-glucopyranosyl unit (δ 5.04, d, $J=7.1$ Hz, H-1), one β-xylopyranosyl unit (δ 5.13, d, $J=7.8$ Hz, H-1) and one α-glucopyranosyl unit (δ 5.52, d, $J=3.9$ Hz, H-1). A phase sensitive rotating-frame nuclear Overhauser effect spectroscopy (PH-ROESY) experiment also afforded confirmations of the sugar anomeric configurations: intra-residue ROEs were detected between H-1 and H-3, H-1 and H-5 for β-glucose, between H-1 and $H5_b$ (axial, δ 3.62), H-3 and H-5_b for β-xylose, between the α-glucose H-1 and H-2.

TABLE II. ^{13}C-NMR Chemical Shifts of Sugar Moieties (in C_5D_5N, δ)

	3	6	6a	7	12	12b	19	20	21	22	11	11a
3-Glc												
1	104.9	104.6	104.8	104.7	105.1	105.1	106.7	105.1	105.1	105.1	105.3	105.3
2	83.2	82.8	82.8	82.8	83.4	83.4	75.5	83.2	83.2	83.4	83.2	83.4
3	78.0$^{a)}$	77.9$^{a)}$	77.7$^{a)}$	77.7$^{a)}$	78.0$^{a)}$	78.1$^{a)}$	78.5	78.1$^{a)}$	78.1$^{a)}$	78.0$^{a)}$	78.0$^{a)}$	78.1$^{a)}$
4	71.4	71.6$^{b)}$	71.8$^{b)}$	71.6	71.6$^{b)}$	71.6$^{b)}$	71.7	71.6$^{b)}$	71.6$^{b)}$	71.6$^{b)}$	71.6$^{b)}$	71.6$^{b)}$
5	78.0$^{a)}$	77.5$^{a)}$	77.7$^{a)}$	77.7$^{a)}$	77.9$^{a)}$	78.0$^{a)}$	78.1	78.2$^{a)}$	78.2$^{a)}$	78.2$^{a)}$	78.1$^{a)}$	78.0$^{a)}$
6	62.6	63.0$^{c)}$	62.9	62.8	62.8$^{c)}$	62.7$^{c)}$	62.9	62.7$^{c)}$	62.7$^{c)}$	62.9$^{c)}$	62.8$^{c)}$	62.8$^{c)}$
Glc												
1	105.7	103.2	103.1	103.1	106.0	106.0		106.0	105.9	106.0	105.9	106.0
2	76.8	84.5	84.5	84.4	77.1	77.1		77.1	77.1	77.1	77.0	77.0
3	79.0$^{a)}$	78.0$^{a)}$	78.3$^{a)}$	78.1$^{a)}$	78.2$^{a)}$	78.2$^{a)}$		78.9$^{a)}$	79.1$^{a)}$	78.3$^{a)}$	79.2$^{a)}$	78.4$^{a)}$
4	71.4	71.3$^{b)}$	71.1$^{b)}$	71.1	72.0$^{b)}$	71.7$^{b)}$		71.7$^{b)}$	71.6$^{b)}$	71.7$^{b)}$	71.7$^{b)}$	71.8$^{b)}$
5	78.0$^{a)}$	77.5$^{a)}$	77.9$^{a)}$	77.7$^{a)}$	77.9$^{a)}$	78.3$^{a)}$		78.3$^{a)}$	78.7$^{a)}$	79.0$^{a)}$	78.3$^{a)}$	78.1$^{a)}$
6	62.6	63.1$^{c)}$	62.9	62.8	63.1$^{c)}$	62.8$^{c)}$		62.8$^{c)}$	62.8$^{c)}$	62.9$^{c)}$	62.9$^{c)}$	62.9$^{c)}$
Xyl												
1		106.3	106.3	106.3								
2		75.7	75.9	75.7								
3		79.0	78.7	79.1								
4		70.7	71.1	70.6								
5		67.2	67.4	67.3								
20-Glc												
1	98.1	98.2		97.9	98.6		98.0	98.2	98.3	98.3	98.2	
2	74.9	75.1		75.0	75.6		75.0	75.3	75.3	75.2	75.1	
3	78.0$^{a)}$	78.0$^{a)}$		78.1$^{a)}$	79.0$^{a)}$		78.5	78.3$^{a)}$	78.8$^{a)}$	78.4$^{a)}$	78.1$^{a)}$	
4	71.4	72.1$^{b)}$		71.6	71.6$^{b)}$		71.3	71.6$^{b)}$	71.6$^{b)}$	71.5$^{b)}$	71.8$^{b)}$	
5	78.0$^{a)}$	78.6$^{a)}$		76.9$^{a)}$	78.3$^{a)}$		78.1	78.2$^{a)}$	77.9$^{a)}$	77.9$^{a)}$	78.0$^{a)}$	
6	62.6	63.1$^{c)}$		70.6	62.7$^{c)}$		62.9	62.8$^{c)}$	63.0$^{c)}$	63.0$^{c)}$	62.8$^{c)}$	
Glc												
1				105.2								
2				74.7								
3				78.1$^{a)}$								
4				71.6								
5				78.1$^{a)}$								
6				62.8								

	15	16	17
6-Glc			
1	103.5	103.5	103.6
2	80.4	80.2	80.3
3	78.8$^{a)}$	78.8$^{a)}$	78.3$^{a)}$
4	71.3$^{b)}$	71.7$^{b)}$	71.2$^{b)}$
5	79.9$^{a)}$	79.9$^{a)}$	76.7
6	62.9	62.9$^{c)}$	68.8
Xyl			
1	104.9	104.3	105.0
2	75.9	75.4$^{d)}$	75.7$^{c)}$
3	78.8$^{a)}$	77.2	78.7$^{a)}$
4	71.7$^{b)}$	79.8$^{a)}$	71.5$^{b)}$
5	67.3	65.4	67.2
α-Glc			
1		102.3	101.4
2		74.1	74.0
3		75.3$^{d)}$	75.2$^{c)}$
4		71.9$^{b)}$	72.3
5		74.3	74.0
6		62.7$^{c)}$	62.8

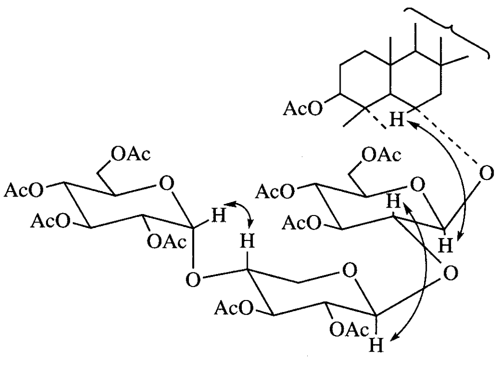
Glc, β-D-glucopyranosyl; Xyl, β-D-xylopyranosyl; α-Glc, α-glucopyranosyl. *a—d*) Interchangeable values in each vertical column.

In the ^{1}H-NMR spectrum of **16a**, the β-glucose H-2 and the β-xylose H-4 signals were shielded, suggesting that the two sugars were not acetylated at these positions and, therefore, giving rise to the presence of glycosyl linkages at the corresponding C-2 and C-4. From the PH-ROESY experiment, interglycosidic crosspeaks were observed between the anomeric proton of the β-glucosyl unit and H-6β of the aglycone, between that of the β-xylosyl unit and the inner glucose H-2, and between the terminal α-glucose H-1 and the middle xylose H-4. It follows that **16** can be formulated as 6-*O*-α-glucopyranosyl-(1→4)-β-D-xylopyranosyl-(1→2)-β-D-glucopyranosyl 20(*S*),24(*S*)-epoxydammarane-3β,6α,12β,25-tetrol.

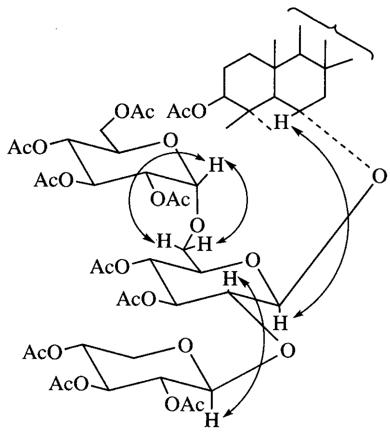
The EI-MS of the acetate and the trimethylsilyl ether of **17** exhibited fragment ions at *m/z* 259 [terminal xylosyl $(Ac)_3$], 331 [terminal glucosyl $(Ac)_4$] and 349 [terminal xylosyl $(TMSi)_3$], 451 [terminal glucosyl $(TMSi)_4$]. The MS data indicated the presence of a branched saccharidic chain with a terminal glucose and a terminal xylose unit. In the ^{1}H-NMR spectrum of **17**, three anomeric protons were observed at δ 5.05 (d, J=7.1 Hz), 5.54 (d, J=3.8 Hz) and 5.70 (d, J=7.2 Hz), which corresponded to the carbon signals at δ 103.6, 101.4 and 105.0, respectively, in the ^{13}C–^{1}H COSY spectrum. A non-decoupled insensitive nuclei enhanced by polarisation transfer (INEPTN) experiment determined their ^{13}C–^{1}H one bond coupling constants ($^1J_{CH}$) as 162, 169 and 162 Hz, respectively. The sugar residue whose anomeric proton atom resonated at δ 5.54 with a characteristic coupling constant (J=3.8 Hz) and the largest ^{13}C–^{1}H one bond coupling ($^1J_{CH}$=169 Hz) must be an α anomeric monosaccharide. The sequence of sugars and the interglycosidic linkage of **17** were determined by analyzing the NMR spectra of its acetate (**17a**) in the same manner applied to **16**.

Careful examination of the ^{1}H-NMR spectral data of the sugar protons as listed in Table III led to the identification of the constituent monosaccharides of **17a** as β-D-glucopyranose (δ 4.99, d, J=7.3 Hz, H-1), β-D-xylopyranose (δ 5.13, d, J=7.8 Hz, H-1) and α-glucopyranose (δ 5.60, d, J=3.9 Hz, H-1). The β-D-glucosyl unit has a shielded H-2 (δ 4.12) and two shielded H-$6_{a,b}$ (δ 3.98 and 4.15) signals while those of the other protons of the acetylated carbons were displaced downfield, disclosing that it is the inner sugar unit and substituted at C-2 and C-6 positions. The PH-ROESY experiment showed correlation between H-6β of the aglycone and H-1 of this inner glucosyl unit whose H-2 and two H-$6_{a,b}$ also displayed interglycosidic ROEs with the terminal xylose H-1 and the terminal α-glucose H-1, respectively. Thus, the structure of the sugar chain was identified and **17** was characterized as 6-*O*-[α-glucopyranosyl-(1→6)]-β-D-xylopyranosyl-(1→2)-β-D-glucopyranosyl 20(*S*),24(*S*)-epoxy-

Table III. ^{1}H-NMR Spectral Data of Sugar Moieties of **16a** and **17a** (400 MHz, in C_5D_5N, δ, Hz)



16a



17a

	16a	**17a**
[Aglycone H-6β:	*ca.* 4.27, overlapping	4.23 (ddd, 11.0, 11.0, 5.0)]
	Inner Glc	Inner Glc
H-1	5.04 (d, 7.1)	4.99 (d, 7.3)
H-2	4.21 (dd, 9.0, 7.1)	4.12 (dd, 9.0, 7.3)
H-3	5.69 (dd, 9.5, 9.0)	5.68 (dd, 9.5, 9.0)
H-4	5.36 (dd, 9.5, 9.5)	5.56 (dd, 9.5, 9.5)
H-5	4.10 (m)	4.02 (m)
H-6	4.48 (dd, 12.0, 5.0)	4.15 (dd, 12.0, 3.0)
	4.32 (dd, 12.0, 3.0)	3.98 (dd, 12.0, 5.0)
	Middle Xyl (1→2)	Terminal Xyl (1→2)
H-1	5.13 (d, 7.8)	5.13 (d, 7.8)
H-2	5.26 (dd, 9.5, 7.8)	5.38 (dd, 9.5, 7.8)
H-3	5.62 (dd, 9.5, 9.5)	5.65 (dd, 9.5, 9.5)
H-4	4.18 (ddd, 9.5, 9.5, 5.5)	5.29 (ddd, 10.0, 9.5, 5.5)
H-5	4.38 (dd, 11.5, 5.5)	4.25 (dd, 11.5, 5.5)
	3.62 (dd, 11.5, 9.5)	3.59 (dd, 11.5, 10.0)
	Terminal α-Glc (1→4)	Terminal α-Glc (1→6)
H-1	5.52 (d, 3.9)	5.60 (d, 3.9)
H-2	5.19 (dd, 10.0, 3.9)	5.30 (dd, 10.0, 3.9)
H-3	5.79 (dd, 10.0, 10.0)	5.96 (dd, 10.0, 10.0)
H-4	5.38 (dd, 10.0, 10.0)	5.48 (dd, 10.0, 10.0)
H-5	4.27 overlapping	4.33 (ddd, 10.0, 5.0, 3.0)
H-6	4.47 (dd, 12.0, 5.0)	4.54 (dd, 12.0, 5.0)
	4.32 (dd, 12.0, 3.0)	4.39 (dd, 12.0, 3.0)

dammmarane-3β,6α,12β,25-tetrol.

After establishing the structures, the remaining question of the assignments of the sugar carbon signals of **16** and **17** was then solved as shown in Table II by comparison of the ^{13}C-NMR spectra of these two compounds with the published data of methyl α-D-glucopyranoside[4,12] and majonoside-R2[11,13] (**15**), in addition to an analysis of the glycosylation shifts of the sugar moieties.

It is noteworthy that saponins containing α-linked glucose rarely occur in plants. Notoginsenoside-R6 (**9**), previously isolated from Sanchi Ginseng (*Panax notoginseng*) and now also from Vietnamese Ginseng, was reported as the first saponin having an α-glucosyl linkage.[4] Vina-ginsenosides-R5 and -R6 are two more examples of the rarely occurring glycosides. Moreover, the presence of this type of saponins in *P. vietnamensis* which has not been found in any *Panax* spp. other than Sanchi Ginseng seems to be significant of a botanical and taxonomical relationship between the two plants.

Vina-ginsenoside-R7 (**6**), $C_{53}H_{90}O_{22}$, 0.01% yield, gave glucose and xylose on acid hydrolysis. The ^{1}H-NMR and ^{13}C-NMR spectra of **6** showed the presence of four sugar units. From the coupling constants of the anomeric protons and the chemical shifts of the sugar carbons, all the constituent sugars must be β-linked. In the ^{13}C-NMR spectrum of **6**, as compared with those of 20(*S*)-protopanaxadiol (**1**),[8] the signals assignable to C-3 and C-20 were displaced downfield by 11.3 and 10.5 ppm, respectively, accompanied by upfield shifts of the signals due to C-2 and C-21 by 1.5 and 4.4 ppm, whereas other carbon signals of the aglycone remained almost unchanged (see Table I). This fact indicated that **6** is a bisdesmoside of **1** whose C-3 and C-20 are the sites of glycosidic linkages. The EI-MS of the trimethylsilyl ether of **6** exhibited fragment ions at m/z 349 [terminal xylosyl $(TMSi)_3$], 451 [terminal glucosyl $(TMSi)_4$], 727 [glucosyl-xylosyl $(TMSi)_6$] and 1105 [glucosyl-glucosyl-xylosyl $(TMSi)_9$], disclosing that one sugar chain is a glucosyl moiety and

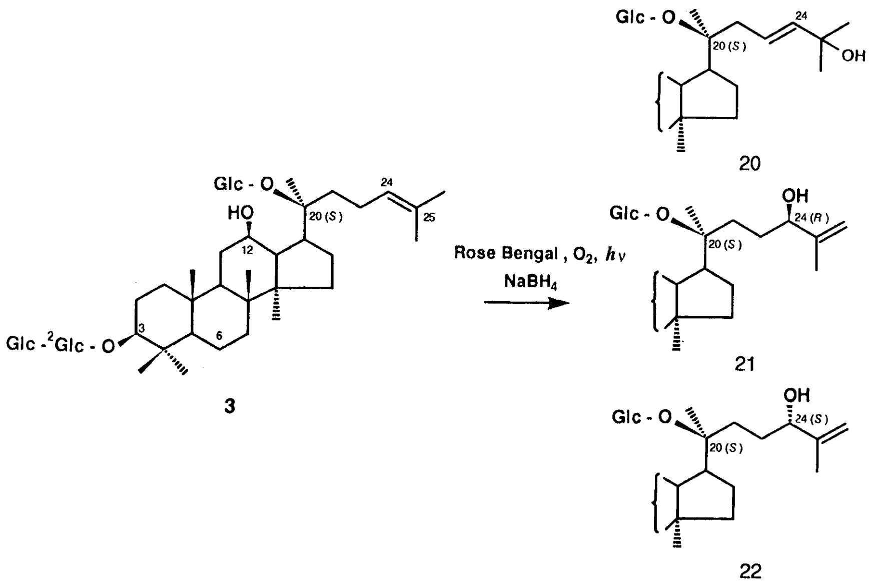


Chart 2

the other a linear xylosyl-glucosyl-glucosyl unit. When the ^{13}C-NMR spectra of **6** and notoginsenoside Fa (**7**), first isolated from leaves of *Panax notoginseng*,[14] were compared, all carbon signals of **6** were almost superimposable on those of **7**, except for the lack of a set of signals in **6** attributable to one terminal glucosyl unit in the sugar chain at C-20. On heating with aqueous acetic acid, **6** gave only glucose and a prosapogenin (**6a**) which was identical with that obtained from the same treatment of **7**. The EI-MS of the trimethylsilyl ether of **6a** exhibited characteristic fragment ions of a linear xylosyl-glucosyl-glucosyl unit (*vide supra*). In addition, the ^{13}C-NMR spectrum confirmed that **6a** is a C-20 epimeric mixture having the trisaccharide moiety at C-3 (see Tables I and II). These data allowed the formulation of **6** as 3-*O*-[β-D-xylopyranosyl-(1→2)-β-D-glucopyranosyl-(1→2)-β-D-glucopyranosyl]-20-*O*-β-D-glucopyranosyl 20(*S*)-protopanaxadiol.

Vina-ginsenoside-R8(**20**), 0.004% yield, has the molecular formula of $C_{48}H_{82}O_{19}$. From the acid hydrolysate of **20**, glucose was obtained. The ^{1}H- and ^{13}C-NMR spectral data revealed that **20** contains three β-glucopyranosyl units. The EI-MS of **20** as a trimethylsilyl ether exhibited fragment ions at *m*/*z* 451 [terminal glucosyl $(TMSi)_4$] and 829 [glucosyl-glucosyl $(TMSi)_7$]. Analysis of the spectral data showed that **20** is a protopanaxadiol-type saponin having a modified side-chain and bearing glycosyl linkages at both the 3- and 20-hydroxyl groups. The ^{13}C-NMR spectrum of **20** was in good coincidence with that of majoroside F4 (**19**), reportedly isolated from leaves of *Panax japonicus* var. *major*,[7] except for the presence of an additional set of carbon signals in **20** assignable to one more terminal glucose unit. Furthermore, the carbon resonances of the sugar moieties of **20** essentially matched those of **3**, **11** and **12**, accounting for the presence of a β-sophorosyl unit at C-3 position. Mild hydrolysis of **20** with aqueous acetic yielded glucose. Thus, **20** was characterized as 3-*O*-[β-D-glucopyranosyl-(1→2)-β-D-glucopyranosyl]-20-*O*-β-D-glucopyranosyl 20(*S*)-dammar-23-ene-3β,12β,20,25-tetrol.

Vina-ginsenoside-R9 (**22**), $C_{48}H_{82}O_{19}$, was obtained in 0.004% yield and gave glucose as the only sugar constituent. The ^{1}H- and ^{13}C-NMR spectra of **22** indicated the presence of three β-linked glucosyl moieties. The EI-MS of the trimethylsilylated **22** exhibited fragment ions at *m*/*z* 451 [terminal glucosyl $(TMSi)_4$] and 829 [glucosyl-glucosyl $(TMSi)_7$]. Treatment of **22** with 50% acetic acid gave glucose. A comparison of the ^{13}C-NMR spectrum of **22** with that of majoroside F1 (**21**),[7] which was also isolated from the studied material, showed a good agreement for all carbon signals due to the aglycone and sugar moieties, except for relatively significant upfield shifts of the C-24 and C-26 signals by $\Delta\delta = -0.5$ and -0.4 ppm, respectively, on going from **21** to **22**. This indicated that **22** is the C-24 epimer of **21**. In previous studies, C-24 epimeric mixtures of dammarane saponins having the similar side-chain structure named chikusetsusaponin-L_{3bc}, chikusetsusaponin-L_{9bc} and ginsenoside-M_{7cd} were isolated and their ^{13}C-NMR data were established.[15-17] Based on a ^{13}C-NMR assignment for ginsenoside-M_{7cd} in which the signals appearing at lower field than those of corresponding signals were ascribed to the 24(*R*) isomer, **21** was previously characterized as a 24(*R*) epimer.[7] Compound **22** was therefore deduced to be the corresponding 24(*S*) counterpart of **21** and was formulated as 3-*O*-[β-D-glucopyranosyl-(1→2)-β-D-glucopyranosyl]-20-*O*-β-D-glucopyranosyl 20(*S*)-dammar-25-ene-3β,12β,20,24(*S*)-tetrol.[18]

The formulation for **20** and **22** was finally confirmed by the preparation of **20**, **21** and **22** from ginsenoside-Rd (**3**) (see Chart 2). On photosensitized oxidation with Rose Bengal followed by reduction of the resultant hydroperoxides with $NaBH_4$[19] and separation by reversed-phase HPLC, **3** gave **22**, **20** and **21**, successively, which were proved to be vina-ginsenoside-R9, vina-ginsenoside-R8 and majoroside F1, respectively, by physicochemical evidence. Since saponins of the similar structures were found in fresh leaves of *P. japonicus*[15] and *P. ginseng*,[16] none of the three saponins seemed to be an artifact formed during the storage of the material or during the extraction process.

Experimental

General Procedures Melting points were measured on a Yanaco micro hot-stage and are uncorrected. Optical rotations were measured using a Union PM-101 automatic digital polarimeter. NMR spectra were recorded on JEOL JNM GX400 and JEOL JNM GX500 spectrometers in C_5D_5N, unless otherwise stated, using tetramethylsilane (TMS) as an internal standard. MS were obtained on a JEOL JMS-SX 102 spectrometer by the direct inlet method at an ionizing voltage of 70 eV. For gas chromatography (GC), a Shimadzu GC-8A apparatus was used. HPLC was carried out using a D-ODS-5 (20 mm i.d. × 25 cm) column with a Tosoh HLC 803D pump and a Tosoh RI-8 differential refractometer as detector.

For column chromatography, Kieselgel 60 (70—230 mesh, Merck), LiChroprep RP-8 (40—63 μm, Merck) were used.

Identification of the Known Saponins Each known saponin was identified by TLC on Silica gel 60 precoated plates, F-254 (Merck), with the solvent systems: $CHCl_3$–MeOH–H_2O (65 : 35 : 10, lower phase), $CHCl_3$–MeOH–H_2O (60 : 40 : 10, homogeneous), $CHCl_3$–1-BuOH–MeOH–H_2O (20 : 40 : 15 : 20, lower phase) and by HPTLC using RP-8 and/or RP-18 precoated plates, F-254s (Merck) using 60—80% MeOH as solvents (detection, 10% H_2SO_4), as well as by comparison of optical rotation, ^{1}H- and ^{13}C-NMR spectra and MS (as a trimethylsilyl ether) with those of a corresponding authentic sample or with the reported data in the case of **21**.

Trimethylsilylation and acetylation for EI-MS, acid hydrolysis and identification of resulting monosaccharides after hydrolysis: see the previous paper.[1,20]

Extraction and Separation of Saponins The dried rhizomes and roots of *P. vietnamensis* were extracted and separated as described.[1] A crude saponin obtained from column chromatography of fr.-III over LiChroprep RP-8 using 55—70% MeOH was further purified by HPLC using the solvent 75% MeOH to afford **12** in yield of 0.009%. Fraction IV was chromatographed on reversed-phase silica gel RP-8 using a gradient elution of 50—70% MeOH to afford **2** (0.036% yield), crude saponins **9**, **16**, **11** and **4**, and mixtures of **10** and **17**; **20**, **21** and **22**; **5** and **6**. **2**, a white powder, $[\alpha]_D^{22}$ +19.0° (c=1.05, MeOH). Crude **9**, **16** and **11** were purified by HPLC (solvent: 52% MeOH, 52% MeOH and 60% MeOH, respectively) to give the corresponding saponins **9** (0.01% yield), **16** (0.008% yield) and **11** (0.004% yield). **9**, a white powder, $[\alpha]_D^{22}$ +42.0° (c=1.0, MeOH). Crude **4** was subjected to silica gel column chromatography [solvent: $CHCl_3$–MeOH–H_2O (70 : 30 : 10, lower phase)] from which pure **4** was obtained as a white powder in yield of 0.013%, $[\alpha]_D^{22}$ +4.0° (c=1.0, MeOH).

The mixture of **10** and **17** was chromatographed on silica gel [solvent: $CHCl_3$–1-BuOH–MeOH–H_2O (20 : 40 : 15 : 20, lower phase)] to give **10** (0.01% yield) and crude **17**. The latter was purified by repeated HPLC using 49% MeOH, affording **17** in yield of 0.006%. **10**, a white powder, $[\alpha]_D^{22}$ +20.0° (c=0.9, MeOH).

The mixture of **20**, **21** and **22** was subjected to HPLC with 70% MeOH to give **22**, **20** and **21**, successively, in yields of 0.004%, 0.004% and 0.003% respectively. **21**, a white powder, $[\alpha]_D^{25}$ +9.3° (c=0.9, MeOH).

The mixture of **5** and **6** was chromatographed on silica gel [solvent: $CHCl_3$–MeOH–H_2O (70 : 30 : 10, lower layer)] and then subjected to HPLC using 70% MeOH as solvent to give **5** (0.012% yield) and **6** (0.01% yield). **5**, a white powder, $[\alpha]_D^{22}$+14.0° (c=1.0, MeOH).

Vina-ginsenoside-R3 (12) Colorless needles from MeOH–H_2O, mp 254—256 °C (dec.), $[\alpha]_D^{18}$ −15.0° (c=0.6, MeOH). HR-FAB-MS (negative): Found m/z: 929.5419; $[C_{48}H_{82}O_{17}-H]^-$ requires m/z 929.5474. FAB-MS (negative) m/z: 929 $[M-H]^-$, 767 $[(M-H)-Glc]^-$, 605 $[(M-H)-Glc-Glc]^-$. EI-MS (acetate deriv.) m/z: 619 [Glc–Glc $(Ac)_7$], 331 [Glc $(Ac)_4$]; (TMSi deriv.) m/z: 829 [Glc-Glc $(TMSi)_7$], 739 [829−TMSiOH], 451 [Glc $(TMSi)_4$], 361 [451−TMSiOH]. ^{1}H-NMR (500 MHz) δ: 0.8, 1.14, 1.31, 1.54, 1.69, 1.70 (each 3H), 1.00 (6H) (each s, *tert*-Me × 8), 3.31 (1H, dd, J=11.5, 4.0 Hz, H-3), 5.33 (1H, t, J=7.0 Hz, H-24), 4.96 (1H, d, J=7.3 Hz, H-1 of inner 3-Glc), 5.12 (1H, d, J=8.0 Hz, H-1 of 20-Glc), 5.40 (1H, d, J=7.3 Hz, H-1 of teminal Glc). ^{13}C-NMR: see Tables I and II.

Enzymatic Hydrolysis of **12**: Compound **12** (20 mg) was suspended in 5% MeOH (20 ml). After addition of crude hesperidinase (100 mg) and a few drops of toluene to prevent contamination, the mixture was incubated at 37 °C for 10 d. The reaction mixture was diluted with an equivalent volume of H_2O and then extracted with EtOAc. The residue obtained after evaporation of the solvent was chromatographed on silica gel using benzene–acetone (8 : 2) as the eluting solvent to give the aglycone **12a** (5 mg).

Compound 12a (Dammarenediol II) Colorless crystal from MeOH–H_2O, mp 65—67 °C, $[\alpha]_D^{25}$ +29.6° (c=0.8, $CHCl_3$) [lit.,[9] mp. 75—76 °C (dimorphic), $[\alpha]_D$ +33° ($CHCl_3$)]. ^{1}H-NMR (400 MHz) δ: 0.88, 0.98, 1.00, 1.06, 1.25, 1.43, 1.66, 1.70 (each 3H, s, *tert*-Me × 8), 3.47 (1H, dd, J=11.0, 7.0 Hz, H-3), 5.35 (1H, t, J=7.0 Hz, H-24). ^{13}C-NMR: see Table I.

Vina-ginsenoside-R4 (11) A white powder, $[\alpha]_D^{18}$ +28.4° (c=0.7, MeOH). HR-FAB-MS (negative): Found m/z: 961.5385; $[C_{48}H_{82}O_{19}-H]^-$ requires m/z 961.5372. FAB-MS (negative) m/z: 961 $[M-H]^-$, 799 $[(M-H)-Glc]^-$, 637 $[(M-H)-Glc-Glc]^-$, 475 $[(M-H)-Glc-Glc-Glc]^-$. EI-MS (acetate deriv.) m/z: 619 [Glc–Glc $(Ac)_7$], 331 [Glc $(Ac)_4$]; (TMSi deriv.) m/z: 829 [Glc–Glc $(TMSi)_7$], 739 [829−TMSiOH], 451 [Glc $(TMSi)_4$], 361 [451−TMSiOH]. ^{1}H-NMR (400 MHz) δ: 0.92, 0.99, 1.05, 1.51, 1.61, 2.00 (each 3H), 1.60 (6H) (each s, *tert*-Me × 8), 3.36 (1H, dd, J=11.9, 4.8 Hz, H-3), 4.18 (1H, m, H-12), *ca.* 4.37 (1H, overlapped, H-6), 5.07 (1H, br s, H-24), 4.97 (1H, d, J=7.2 Hz, H-1 of inner 3-Glc), 5.18 (1H, d, J=7.6 Hz, H-1 of 20-Glc), 5.42 (1H, d, J=7.7 Hz, H-1 of terminal Glc). ^{13}C-NMR: see Tables I and II.

Vina-ginsenoside-R5 (16) A white powder, $[\alpha]_D^{18}$ +38.0° (c=1.0, MeOH). HR-FAB-MS (negative): Found m/z: 947.5275; $[C_{47}H_{80}O_{19}-H]^-$ requires m/z 947.5215. FAB-MS (negative) m/z: 947$[M-H]^-$, 785 $[(M-H)-Glc]^-$, 653 $[(M-H)-Glc-Xyl]^-$, 491 $[(M-H)-Glc-Xyl-Glc]^-$. EI-MS (acetate deriv.) m/z: 835 [Glc–Xyl–Glc $(Ac)_9$], 547 [Glc–Xyl $(Ac)_6$], 331 [Glc$(Ac)_4$], 143 (fragment ion **18**); (TMSi deriv.) m/z: 1105 [Glc–Xyl–Glc $(TMSi)_9$], 727 [Glc–Xyl $(TMSi)_6$], 637 [727−TMSiOH], 451 [Glc$(TMSi)_4$], 361 [451−TMSiOH]. ^{1}H-NMR (400 MHz) δ: 0.79, 1.01 1.22, 1.29, 1.33, 1.45, 1.46, 2.04 (each 3H, s, *tert*-Me × 8), 3.51 (1H, dd, J=11.0, 5.0 Hz, H-3), 3.74 (1H, ddd, J=11.0, 11.0, 4.8 Hz, H-12), 4.09 (1H, dd, J=9.7, 3.8 Hz, H-24), *ca.* 4.35 (1H, overlapped, H-6), 4.94 (1H, d, J=6.4 Hz, H-1 of inner 6-Glc), 5.66 (1H, d, J=3.9 Hz, H-1 of terminal α-Glc), 5.68 (1H, d, J=7.1 Hz, H-1 of middle Xyl). ^{13}C-NMR: see Tables I and II.

Vina-ginsenoside-R6 (17) A white powder, $[\alpha]_D^{25}$+20.0° (c=0.4, MeOH). HR-FAB-MS (negative): Found m/z: 947.5260; $[C_{47}H_{80}O_{19}-H]^-$ requires m/z 947.5216. EI-MS (acetate deriv.) m/z: 835 [Glc–(Glc)–Xyl $(Ac)_9$], 331 [Glc$(Ac)_4$], 259 [Xyl$(Ac)_3$], 143 (fragment ion **18**); (TMSi deriv.) m/z: 451 [Glc $(TMSi)_4$], 361 [451−TMSiOH], 349 [Xyl$(TMSi)_3$], 259 [349−TMSiOH]. ^{1}H-NMR (400 MHz) δ: 0.83, 0.97, 1.28, 1.29, 1.45, 2.07 (each 3H), 1.36 (6H) (each s, *tert*-Me × 8), 3.49 (1H, dd, J=11.0, 5.0 Hz, H-3), 3.75 (1H, m, H-12), 4.15 (1H, dd, J=9.8, 3.6 Hz, H-24), *ca.* 4.36 (1H, overlapped, H-6), 5.05 (1H, d, J=7.1 Hz, H-1 of inner 6-Glc), 5.54 (1H, d, J=3.8 Hz, H-1 of terminal α-Glc), 5.70 (1H, d, J=7.2 Hz, H-1 of terminal Xyl). ^{13}C-NMR: see Tables I and II.

Acetylation of 16 and 17 Each of the saponins [**16** (8 mg), **17** (10 mg)] was dissolved in Ac_2O–C_5D_5N (1 : 1.2 ml) and left at room temperature overnight. The reaction mixture was subjected to azeotropic distillation with toluene to afford the corresponding acetylated saponin [**16a** (10 mg), **17a** (12 mg)]. The acetylated saponin was monitored on TLC [solvent A: $CHCl_3$–MeOH (9 : 1)] and HPTLC (solvent B: 80% MeOH) which revealed only a single spot [**16**: Rf_A=0.60, Rf_B=0.27; **17**: Rf_A=0.56, Rf_B=0.33). ^{1}H-NMR: see Table III.

Vina-ginsenoside-R7 (6) A white powder, $[\alpha]_D^{25}$+17.8° (c=1.07, MeOH). HR-FAB-MS (negative): Found m/z: 1077.5880; $[C_{53}H_{90}O_{22}-H]^-$ requires m/z 1077.5842. FAB-MS (negative) m/z: 1077 $[M-H]^-$, 945 $[(M-H)-Xyl]^-$, 915 $[(M-H)-Glc]^-$, 783 $[(M-H)-Xyl-Glc]^-$, 621 $[(M-H)-Xyl-Glc-Glc]^-$, 459 $[(M-H)-Xyl-Glc-Glc-Glc]^-$. EI-MS (TMSi deriv.) m/z: 1105 [Xyl–Glc–Glc $(TMSi)_9$], 1015 [1105−TMSiOH], 727 [Xyl–Glc $(TMSi)_6$], 637 [727−TMSiOH], 451 [Glc $(TMSi)_4$], 361 [451− TMSiOH], 349 [Xyl $(TMSi)_3$], 259 [349−TMSiOH]. ^{1}H-NMR (400 MHz) δ: 0.79, 0.94, 0.95, 1.11, 1.28, 1.63 (each 3H), 1.60 (6H) (each s, *tert*-Me × 8), 3.29 (1H, dd, J=11.7, 4.4 Hz, H-3), 4.11 (1H, m, H-12), 5.25 (1H, t, J=6.5 Hz, H-24), 4.93 (1H, d, J=7.7 Hz, H-1 of inner 3-Glc), 5.19 (1H, d, J=7.7 Hz, H-1 of 20-Glc), 5.40 (1H, d, J=6.7 Hz, H-1 of teminal Xyl), 5.52 (1H, d, J=7.7 Hz, H-1 of middle Glc). ^{13}C-NMR: see Tables I and II.

Vina-ginsenoside-R8 (20) A white powder, $[\alpha]_D^{25}$ +14.0° (c=1.0, MeOH). HR-FAB-MS (negative): Found m/z: 961.5392; $[C_{48}H_{82}O_{19}-$

H]$^-$ requires *m/z* 961.5372. FAB-MS (negative) *m/z*: 961 [M−H]$^-$, 799 [(M−H)−Glc]$^-$, 637 [(M−H)−Glc−Glc]$^-$, 475 [(M−H)−Glc−Glc−Glc]$^-$. EI-MS (acetate deriv.) *m/z*: 619 [Glc–Glc (Ac)$_7$], 331 [Glc (Ac)$_4$]; (TMSi deriv.) *m/z*: 829 [Glc–Glc (TMSi)$_7$], 739 [829−TMSiOH], 451 [Glc (TMSi)$_4$], 361 [451−TMSiOH]. ^{1}H-NMR (400 MHz) δ: 0.84, 0.89, 1.00, 1.11, 1.29, 1.54 (each 3H), 1.53 (6H) (each s, *tert*-Me × 8), 3.26 (1H, dd, J=11.6, 4.4 Hz, H-3), 4.02 (1H, m, H-12), 6.25 (1H, td, J=15.5, 7.5 Hz, H-23), 6.03 (1H, d, J=15.5 Hz, H-24), 4.92 (1H, d, J=7.7 Hz, H-1 of inner 3-Glc), 5.19 (1H, d, J=7.6 Hz, H-1 of 20-Glc), 5.36 (1H, d, J=7.7 Hz, H-1 of terminal Glc). ^{13}C-NMR: see Tables I and II.

Vina-ginsenoside-R9 (22) A white powder, $[\alpha]_D^{25}$ +10.5° (c=0.6, MeOH). HR-FAB-MS (negative): Found *m/z*: 961.5420; $[C_{48}H_{82}O_{19}-H]^-$ requires *m/z* 961.5372. FAB-MS (negative) *m/z*: 961 [M−H]$^-$, 799 [(M−H)−Glc]$^-$, 637 [(M−H)−Glc−Glc]$^-$, 475 [(M−H)−Glc−Glc−Glc]$^-$. EI-MS (TMSi deriv.) *m/z*: 829 [Glc–Glc (TMSi)$_7$], 739 [829−TMSiOH], 451 [Glc(TMSi)$_4$], 361 [451−TMSiOH]. ^{1}H-NMR (400 MHz) δ: 0.79, 0.92, 0.94, 1.10, 1.28, 1.63, 1.89 (each 3H, s, *tert*-Me × 7), 3.26 (1H, dd, J=12.0, 4.0 Hz, H-3), 4.07 (1H, m, H-12), 4.42 (1H, dd, J=12.0, 5.0 Hz, H-24), 4.91 and 5.68 (each 1H, br s, >C=CH$_2$), 4.92 (1H, d, J=7.4 Hz, H-1 of inner 3-Glc), 5.22 (1H, d, J=7.7 Hz, H-1 of 20-Glc), 5.37 (1H, d, J=7.5 Hz, H-1 of terminal Glc). ^{13}C-NMR (in C_5D_5N): see Tables I and II; (in CD_3OD, 100 MHz, δ): aglycone moiety: C-1, 40.3; C-2, 27.3; C-3, 91.2; C-4, 40.5; C-5, 57.5; C-6, 19.2; C-7, 35.9; C-8, 41.0; C-9, 51.1; C-10, 37.9; C-11, 31.6; C-12, 71.9; C-13, 49.2; C-14, 52.2; C-15, 31.1; C-16, 27.3; C-17, 53.3; C-18, 17.3; C-19, 16.7; C-20, 84.9; C-21, 22.9; C-22, 32.4; C-23, 30.7; C-24, 76.9; C-25, 148.9; C-26, 111.3; C-27, 18.0; C-28, 28.4; C-29, 16.2; C-30, 17.3; sugar moieties: inner 3-Glc (C-1 → C-6), 104.5, 81.1, 78.2, 71.5, 77.6, 62.6; terminal Glc (C-1 → C-6), 105.3, 76.3, 78.2, 71.1, 77.9, 62.6, 20-Glc (C-1 → C-6), 98.2, 75.3, 78.4, 71.5, 77.6, 63.1.

Partial Acid Hydrolysis of 6, 11, 12 Each saponin (15 mg) was dissolved in 50% acetic acid (5 ml) and heated at 70 °C for 4 h. After dilution with H_2O, the reaction mixture was extracted with 1-BuOH saturated with H_2O. The aqueous layer was neutralized by Amberlite MB-3 and concentrated to give a residue in which glucose was identified by TLC over silica gel [solvent: $CHCl_3$–MeOH–H_2O (60 : 40 : 10)]. The BuOH layer was washed with H_2O and concentrated to dryness. This residue was purified by silica gel column chromatography [solvent: $CHCl_3$–MeOH–H_2O (70 : 30 : 10, lower layer)] to give the corresponding prosapogenin **6a** (8 mg), **11a** (10 mg) and **12b** (6 mg).

Photosensitized Oxidation of 3 To a solution of **3** (400 mg) in 2-PrOH (60 ml) was added Rose Bengal (25 mg) and the mixture was stirred and irradiated by a 230 W Hg lamp for 5 h. The reaction mixture was treated with active charcoal to remove the pigment and then evaporated to dryness *in vacuo*. After addition of AcOH and H_2O, the reaction mixture was extracted with 1-BuOH saturated with H_2O. The BuOH layer was concentrated to dryness and the residue was subjected to HPLC using an ODS column with 70% MeOH afford **22** (46 mg), **20** (85 mg) and **21** (33 mg), successively. The obtained saponins were identified by optical rotation, TLC behavior, and NMR spectral data.

References and Notes

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18) From a research in a series of chemical studies on *Panax* spp., S. Yahara (reference 17 above) previously isolated **20** and an epimeric mixture of **21** and **22** as the acetate derivatives from the leaves of *P. japonicus* C. A. Meyer and termed them ginsenoside-F_{6a} and F_{6bc}, respectively. However, separation of the C-24 epimeric mixture, ginsenoside-F_{6bc}, was unsuccessful and low yields of the saponins prohibited a concrete structural elucidation. The 20(*R*) isomer of ginsenside-F_{6bc} was later isolated and identified as majoroside F1,[7] and from this work ginsenoside-F_{6a} and the 24(*S*) isomer of ginsenoside-F_{6bc} were characterized as two novel saponins, vina-ginsenosides-R8 and -R9, respectively.
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