Tirucallane-Type Triterpenoid Saponins from the Roots of *Sapindus mukorossi*

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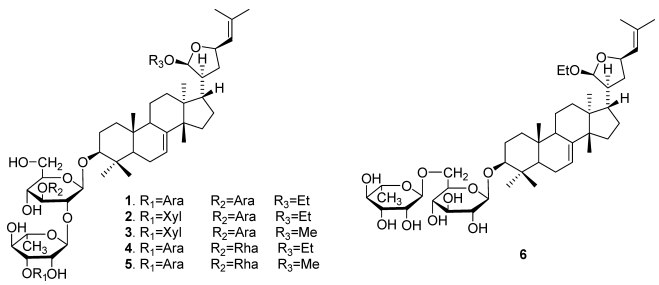
Six new tirucallane-type triterpenoid saponins, sapimukosides E—J (1—6) were isolated from the roots of *Sapindus mukorossi* GAERTN. Their structures were elucidated by a combination of spectral and chemical analysis.

Key words *Sapindus mukorossi*; Sapindaceae; triterpenoid saponin; sapimukoside

Sapindus mukurossi GAERTN. (Sapindaceae), which distributes in tropical and subtropical region of Asia, is an important economic agricultural product as a source of natural surfactants. Additionally, it has been used as expectorant, relieving cough, detoxification and defervescence in China.[1)] Previous phytochemical studies have identified several different types of saponins, containing sesquiterpene oligoglycoside,[2)] hederagenin saponins,[3,4)] dammarane-type triterpenoids[5)] and tirucallane-type triterpenoid saponins[6,7)] from the pericarp, stem and root. Our further phytochemical examination of the roots of this plant has led to the isolation of another six new saponins, sapimukosides E—J (**1**—**6**). This paper deals with the elucidation of their structures on the basis of MS, ^{1}H- and ^{13}C-NMR, and 2D NMR spectroscopic data and the results of hydrolytic cleavage.

The EtOH extract of the roots of *S. mukurossi* was partitioned between H_2O and *n*-BuOH, and the *n*-BuOH fraction was subjected to D-101 resin column, repeated silica gel column and RP-18 column chromatography to afford sapimukosides E—J (**1**—**6**).

Sapimukoside E (**1**) was obtained as an amorphous white powder, and its molecular formula was established to be $C_{54}H_{88}O_{20}$ from the negative-ion high-resolution fast atom bombardment mass spectrometry (HR-FAB-MS) (*m/z* 1055.5718 $[M-H]^-$) and ^{13}C-NMR and DEPT spectral data (Table 1) which suggested 11 degree of unsaturation. The IR spectrum exhibited the presence of hydroxyl (3418 cm^{-1}) and olefinic (1448, 1084 cm^{-1}) groups. The ^{1}H-NMR spectrum of the aglycone moiety of **1** showed signals for five tertiary methyls (δ_H 0.75, 0.99, 1.05, 1.29, 1.37), two allylic methyls (δ_H 1.68, 1.69), and three oxymethines (δ_H 3.47, 5.07, 5.16), suggesting the aglycone contained a triterpene skeleton. The ^{13}C-NMR and DEPT spectrum of the aglycone moiety of **1** indicated seven tertiary methyls, eight methylenes, four methine carbons [δ 48.9 (C-20), 49.0 (C-9), 49.2 (C-17), 51.9 (C-5)], one oxymethine [δ_C 89.3 (C-3)], four quaternary carbons [δ_C 35.0 (C-10), 39.8 (C-4), 44.2 (C-13), 51.6 (C-14)], two trisubstituted olefinic carbons [δ_C 118.5 (C-7), 145.9 (C-8)], characteristic of a tirucallane-euphane system having a double bond between C-7 and C-8, and a 3β hydroxyl group.[8—10)] In addition, the ^{1}H- and ^{13}C-spectra also indicated the presence of one hemiacetal carbon [δ_C 107.3 (C-21)], one oxymethine carbon [δ_C 75.8 (C-23)], two olefinic carbons [δ_C 129.4 (C-24), 133.4 (C-25)], along with a ethoxyl group [δ_C 15.8 (q), 63.1 (t)], which suggested the existence of a hemiacetal group and one double bond in the side chain. Double bonds were located at C-7 and C-23, and three oxymethines were assigned at C-3, C-21, and C-23, on the basis of the following the heteronuclear multiple bond correlations (HMBC) (Fig. 1) of H-3 with C-4, 28, 29, C-1_{glc}; H-7 with C-5, 9, 14; H-21 with C-17, 22, 23, OCH_2CH_3; H-23 with C-20, 21, 22, 25; H-24 with C-22, 23, 26, 27. Further comparison of the NMR data with those of sapimukoside A[6)] showed that the two structures were very similar except that **1** had an additional ethoxyl group and C-21 was shifted downfield from 98.0/102.0 to 107.3. These findings suggested that the additional ethoxyl group was linked at C-21 of the aglycone, which was further confirmed by the heteronuclear multiple bond correlation (HMBC) (Fig. 1) spec-



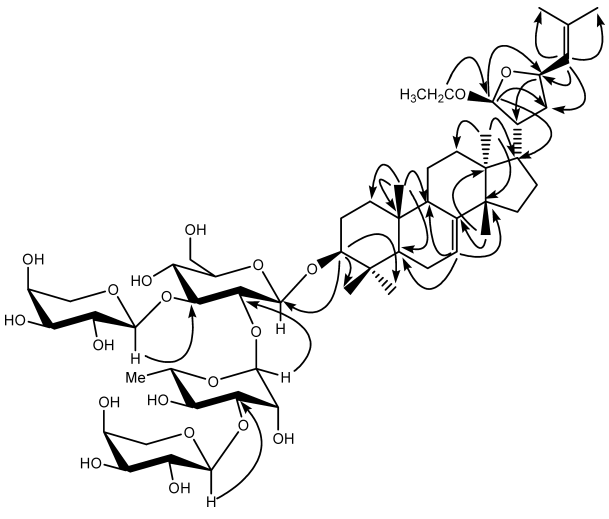


Fig. 1. Key HMBC Correlations of Compound **1**

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trum showing long range correlation between the methylene proton of the ethoxyl group [δ 3.97 (1H, dq, J=7.5, 9.8 Hz), 3.59 (1H, dq, J=7.5, 9.8 Hz)] and C-21. The β configuration of the 21-ethoxyl group and an α-orientation of H-23 were determined by NOE correlations of Me-18 with H-20, H-20 with H-21, H-23 in NOESY spectrum (Fig. 2). So the aglycone was determined to be 21, 23*R*-epoxyl tirucalla-7,24-diene-21β-ethoxyl-3β-ol.

Acid hydrolysis of **1** with 1 M HCl produced D-glucose, L-arabinose and L-rhamnose as sugar residues determined by GC analysis. There were four anomeric carbon signals [δ 107.2 (d, C-$1_{ara'}$), 105.1 (d, C-1_{glc}), 105.0 (d, C-1_{ara}), 101.4 (d, C-1_{rha})] and corresponding four anomeric proton signals [δ 4.87 (1H, d, J=7.7 Hz, H-1_{glc}), 4.85 (1H, d, J=7.3 Hz, H-1_{ara}), 5.38 (1H, d, J=6.9 Hz, H-$1_{ara'}$), 6.46 (1H, br s, H-1_{rha})] in the NMR spectra, indicating compound **1** contained four sugar units. Sugar proton and carbon signals in the NMR spectra were assigned by ^{1}H–^{1}H COSY, HMQC, and HMQC-TOCSY spectra. The linkage sites of each sugar were determined by HMBC spectrum (Fig. 1) which showed long range correlations between H-$1_{ara'}$ (δ_H 5.38) and C-3_{rha} (δ_C 82.3), H-1_{rha} (δ_H 6.46) and C-2_{glc} (δ_C 76.3), H-1_{ara} (δ_H 4.85) and C-3_{glc} (δ_C 88.6), and H-1_{glc} (δ_H 4.87) and C-3 of the aglycone (δ_C 89.4). Each sugar was a pyranosyl with the β configuration for glucosyl and the α configuration for both rhamnosyl and arabinosyl from the NMR data. On the basis of the above

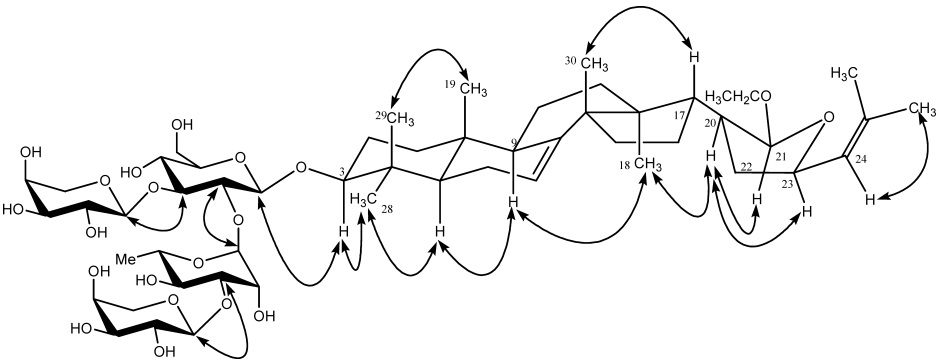

Fig. 2. Significant NOE Correlations of Compound **1**

Table 1. ^{13}C-NMR Spectral Data of the Aglycone Moieties of Compounds **1**—**6** (in C_5D_5N, 125 MHz)

C	1	2	3	4	5	6
1	37.5	37.7	37.4	37.6	37.7	37.3
2	27.4	27.3	27.4	27.3	27.3	27.3
3	89.4	89.4	89.4	89.3	89.3	89.6
4	39.8	39.7	39.7	39.7	39.7	39.7
5	51.9	51.9	51.9	51.9	51.8	51.6
6	24.4	24.3	24.3	24.3	24.3	24.3
7	118.5	118.5	118.7	118.5	118.5	118.5
8	146.0	145.8	145.8	145.9	145.9	146.0
9	49.0	49.0	48.9	49.0	48.8	48.9
10	35.0	34.9	35.0	34.9	34.9	35.1
11	18.1	18.1	18.1	18.1	18.1	18.3
12	32.8	32.8	32.7	32.8	32.8	32.8
13	44.2	44.2	44.2	44.2	44.2	44.2
14	51.6	51.5	51.9	51.5	51.5	51.6
15	34.3	34.3	34.3	34.3	34.3	34.3
16	28.1	28.1	28.1	28.1	28.1	28.0
17	49.2	49.2	49.2	49.2	49.2	49.2
18	23.1	23.5	23.0	23.1	23.0	23.3
19	13.5	13.4	13.4	13.4	13.4	13.4
20	48.9	48.9	48.9	48.9	48.9	48.8
21	107.3	107.3	108.7	107.3	107.2	107.2
22	37.7	37.7	37.7	37.5	37.4	37.3
23	75.8	75.7	75.7	75.6	75.7	75.7
24	129.4	129.4	129.3	129.4	129.2	129.4
25	133.4	133.3	133.5	133.3	133.5	133.3
26	25.9	25.8	25.8	25.8	25.8	25.8
27	18.0	17.9	18.0	18.0	17.9	18.1
28	27.9	27.8	27.8	27.9	27.8	27.9
29	16.4	16.4	16.4	16.3	16.3	16.1
30	27.4	27.3	27.4	27.4	27.3	27.3
1′	15.8	15.8	54.9	15.8	54.9	15.7
2′	63.1	63.1		63.1		

Table 2. ^{13}C-NMR Spectral Data of the Glycoside Moieties of Compounds **1**—**6** (in C_5D_5N, 125 MHz)

C	1	2	3	4	5	6
Glc 1	105.1	105.2	105.2	104.9	104.9	107.0
2	76.3	76.2	76.2	76.8	76.8	76.8
3	88.6	88.6	88.6	88.4	88.4	75.7
4	70.0	69.9	70.0	70.4	70.4	71.9
5	78.1	76.2	78.1	78.0	78.0	78.8
6	62.7	62.7	62.7	62.6	62.0	68.3
Rha 1	101.4	101.4	101.4	101.7	101.7	102.5
2	72.2	72.3	72.2	71.7	71.7	72.4
3	82.3	82.5	82.4	82.8	82.8	72.9
4	72.4	72.4	72.4	73.1	73.1	74.1
5	69.6	69.6	69.6	69.6	69.7	70.0
6	18.6	18.5	18.6	18.6	18.6	18.7
Ara 1	105.0	105.0	105.0			
2	73.1	73.0	73.0			
3	74.6	74.5	74.5			
4	69.5	69.4	69.4			
5	67.8	67.8	67.8			
Ara′ 1	107.2			107.2	107.2	
2	73.2			73.3	73.3	
3	74.5			74.6	74.6	
4	69.5			69.6	69.7	
5	67.1			67.3	67.3	
Rha′ 1				103.8	103.9	
2				72.5	72.5	
3				70.9	70.9	
4				73.6	73.6	
5				69.6	69.7	
6				18.5	18.5	
Xyl 1		107.5	107.5			
2		75.7	75.7			
3		78.5	78.5			
4		71.2	71.2			
5		67.4	67.4			

evidence, the structure of **1** was elucidated as 3-*O*-α-L-arabinopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[α-L-arabinopyranosyl-(1→3)]-β-D-glucopyranosyl-21,23*R*-epoxyl tirucalla-7,24-diene-21β-ethoxy-3β-ol (**1**), and was named sapimukoside E.

Sapimukoside F (**2**) and sapimukoside G (**3**) were assigned the molecular formulae of $C_{54}H_{88}O_{20}$ and $C_{53}H_{86}O_{20}$, respectively, by the negative-ion HR-FAB-MS. A careful comparison of the ^{1}H- and ^{13}C-NMR spectra of **2** with those of **1** showed that the two compounds were very similar except for the terminal sugar unit linked at C-3 of the rhamnosyl unit, which was determined to be β-D-xylopyranosyl by the NMR spectra and GC analysis. The structure of compound **3** was different from that of **2** only in the substitute group at C-21. There was a methoxyl group [δ_C 54.9 (q); δ_H 3.50 (3H, s)] in **3** other than an ethoxyl group in **2**. Furthermore, HMBC spectrum showed long range correlations between the methyl protons of the methoxyl group and C-21. Thus, the structures of sapimukosides F and G were formulated as 3-*O*-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[β-L-arabinopyranosyl-(1→3)]-β-D-glucopyranosyl-21,23*R*-epoxyl tirucalla-7,24-diene-21β-ethoxy-3β-ol, and 3-*O*-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[α-L-arabinopyranosyl-(1→3)]-β-D-glucopyranosyl-21,23*R*-epoxyl tirucalla-7,24-diene-21β-methoxy-3β-ol, respectively.

Sapimukoside H (**4**) and sapimukoside I (**5**) were showed to have the molecular formulae of $C_{55}H_{90}O_{20}$ and $C_{54}H_{88}O_{20}$, respectively, by the negative-ion HR-FAB-MS. The ^{1}H- and ^{13}C-NMR spectra of **4** were similar to those of **1** except for the sugar unit linked at C-3 of the glucopyranosyl unit, which was determined to be α-L-rhamnopyranosyl by the NMR spectra and GC analysis. The structure of compound **5** was different from that of **4** only in the substitute group at C-21. There was a methoxyl group [δ_C 54.9 (q); δ_H 3.49 (3H, s)] in **5** other than an ethoxyl group in **4**. So the structures of sapimukosides H and I were elucidated as 3-*O*-α-L-arabinopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyrano-syl-21,23*R*-epoxyl tirucalla-7,24-diene-21β-ethoxy-3β-ol, and 3-*O*-α-L-arabinopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-21,23*R*-epoxyltirucalla-7,24-diene-21β-methoxy-3β-ol, respectively.

Sapimukoside J (**6**) was found to be $C_{44}H_{72}O_{12}$ by combined negative-ion HR-FAB-MS (*m*/*z* 791.4895 $[M-H]^-$) and ^{13}C-NMR spectroscopic analysis, together with DEPT data. The aglycone moiety of **6** was identical to that of **1** from the NMR spectra. Acid hydrolysis of **6** produced D-glucose and L-rhamnose as sugar residues determined by GC analysis. In the HMBC spectrum, long range correlations can be observed between H-1$_{rha}$ (δ_H 5.53) and C-6$_{glc}$ (δ_C 68.3), and H-1$_{glc}$ (δ_H 4.91) and C-3 of the aglycone (δ_C 89.6). Based on the above evidence, the structure of compound **6** was assigned as 3-*O*-α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranosyl-21,23*R*-epoxyl tirucalla-7,24-diene-21β-ethoxyl-3β-ol, and was named sapimukoside J.

Experimental

General Procedure Melting point: Koffler melting point apparatus (uncorrected). NMR: Bruker DRX-500 (500 MHz for ^{1}H-NMR) using TMS as internal standard, δ in ppm. Optional rotations: Japanese Fasco DIP-370 digital polarimeter. FAB-MS: VG Auto Spec-3000 spectrometer. Chromatography column (CC): D 101 resin (Tianjin Haiguang Chemical Co. Ltd. In China), silica gel (200—300 and 300—400 mesh, Qingdao Marine Chemical Factory in China), RP-18 LiChroprep (40—65 μm, Merck). TLC: GF254 (Qingdao Marine hemical Factory in China). Spots were detected on TLC under UV or by heating after spraying with 10% H_2SO_4 in EtOH.

Plant Material The roots of *Sapindus mukorossi* GAERTN. were collected from Yuxi, Yunnan Province of. China in July 1998. It was identified by Prof. Li Heng at Kunming Institute of Botany, Chinese Academy of Sciences.

Extraction and Isolation The air-dried roots (5.9 kg) were extracted with hot EtOH four times and then concentrated under reduced pressure. The concentrated extract was partitioned between *n*-BuOH and H_2O. The *n*-BuOH layer was subjected to D 101 resin column chromatography eluting with water and 80% EtOH, successively. Then the 80% EtOH fraction was applied to silica gel column chromatography with $CHCl_3$–MeOH (9 : 1 to 8 : 2 v/v) to afford four fractions (I—IV). Fraction I and IV were further repeatedly subjected to repeated silica gel column chromatography using mixtures of $CHCl_3$–MeOH–EtOAc–H_2O (5 : 4 : 2 : 0.2) as eluents and purified by RP-18 gel column with aqueous EtOH. As a result, fraction IV afforded **1** (346 mg), **2** (196 mg), **3** (180 mg), **4** (355 mg), **5** (263 mg) and fraction I yielded **9** (179 mg).

Sapimukoside E (**1**): White amorphous powder. $[\alpha]_D^{24}$ −5.5° (*c*=0.55, MeOH). IR (KBr) ν_{max} cm^{-1}: 3418, 2968, 2932, 2883, 1448, 1384, 1350, 1084, 1009. Negative-ion FAB-MS *m*/*z*: 1055 $[M-H]^-$ (100), 924 $[M-132]^-$ (5), 777 $[M-132-146-H]^-$ (2); HR-FAB-MS *m*/*z*: 1055.5718 $[M-H]^-$ (Calcd for $C_{54}H_{87}O_{20}$: 1055.5791). ^{1}H-NMR (500 Hz, C_5D_5N) δ: 0.75 (3H, s, Me-19), 0.99 (3H, s, Me-30), 1.05 (3H, s, Me-18), 1.27 (3H, t, *J*=7.5 Hz, H-1′), 1.29 (3H, s, Me-29), 1.37 (3H, s, Me-28), 1.59 (3H, d, *J*=5.9 Hz, H-6$_{rha}$), 1.68 (3H, s, Me-27), 1.69 (3H, s, Me-26), 1.91 (1H, m, H-17), 3.47 (1H, dd, *J*=3.1, 11.6 Hz, H-3), 3.59 (1H, dq, *J*=9.8, 7.5 Hz, H-2′b), 3.97 (1H, dq, *J*=9.8, 7.5 Hz, H-2′a), 4.87 (1H, d, *J*=7.7 Hz, H-1$_{glc}$), 4.85 (1H, d, *J*=7.3 Hz, H-1$_{ara}$), 5.07 (1H, dd, *J*=6.9, 14.0 Hz, H-23), 5.16 (1H, br s, H-21), 5.30 (1H, br s, H-7), 5.38 (1H, d, *J*=6.9 Hz, H-1$_{ara'}$), 5.62 (1H, br d, *J*=8.6 Hz, H-24), 6.46 (1H, br s, H-1$_{rha}$); ^{13}C-NMR data: Tables 1 and 2.

Sapimukoside F (**2**): White amorphous powder. $[\alpha]_D^{24}$ −8.6° (*c*=0.30, MeOH). IR (KBr) ν_{max} cm^{-1}: 3424, 2967, 2932, 2883, 1448, 1384, 1084, 1045. Negative-ion FAB-MS *m*/*z*: 1056 $[M]^-$ (100), 924 $[M-132]^-$ (8), 777 $[M-132-146-H]^-$ (3); HR-FAB-MS *m*/*z*: 1055.5771 $[M-H]^-$ (Calcd for $C_{54}H_{87}O_{20}$: 1055.5793). ^{1}H-NMR (500 Hz, C_5D_5N) δ: 0.74 (3H, s, Me-19), 0.99 (3H, s, Me-30), 1.06 (3H, s, Me-18), 1.27 (3H, t, *J*=6.9 Hz, H-1′), 1.30 (3H, s, Me-29), 1.37 (3H, s, Me-28), 1.61 (3H, d, *J*=6.1 Hz, H-6$_{rha}$), 1.67 (3H, s, Me-27), 1.68 (3H, s, Me-26), 1.91 (1H, m, H-17), 3.46 (1H, dd, *J*=3.6, 11.9 Hz, H-3), 3.58 (1H, dq, *J*=9.7, 6.9 Hz, H-2′b), 3.98 (1H, dq, *J*=9.7, 6.9 Hz, H-2′a), 4.86 (1H, d, *J*=7.5 Hz, H-1$_{ara}$), 4.87 (1H, d, *J*=7.5 Hz, H-1$_{glc}$), 5.08 (1H, dd, *J*=6.2, 14.5 Hz, H-23), 5.16 (1H, d, H-21), 5.27 (1H, br s, H-7), 5.45 (1H, d, *J*=7.5 Hz, H-1$_{xyl}$), 5.63 (1H, dq, *J*=1.7, 7.5 Hz, H-24), 6.52 (1H, br s, H-1$_{rha}$); ^{13}C-NMR data: Tables 1 and 2.

Sapimukoside G (**3**): White amorphous powder. $[\alpha]_D^{24}$ −13.5° (*c*=0.37, MeOH). IR (KBr) ν_{max} cm^{-1}: 3421, 2932, 2883, 1450, 1385, 1084, 1044. Negative ion FAB-MS *m*/*z*: 1042 $[M]^-$ (100), 910 $[M-132]^-$ (12), 777 $[M-132-132-H]^-$ (7), 631 $[M-132-132-146-H]^-$ (3); HR-FAB-MS *m*/*z*: 1041.5674 $[M-H]^-$ (Calcd for $C_{53}H_{85}O_{20}$: 1041.5634). ^{1}H-NMR (500 Hz, C_5D_5N) δ: 0.74 (3H, s, Me-19), 0.98 (3H, s, Me-30), 1.03 (3H, s, Me-18), 1.30 (3H, s, Me-29), 1.37 (3H, s, Me-28), 1.60 (3H, d, *J*=6.0 Hz, H-6$_{rha}$), 1.67 (3H, s, Me-27), 1.68 (3H, s, Me-26), 1.91 (1H, m, H-17), 3.47 (1H, dd, *J*=4.4, 12.4 Hz, H-3), 3.50 (3H, s, H-1′), 4.85 (1H, d, *J*=7.5 Hz, H-1$_{ara}$), 4.86 (1H, d, *J*=7.5 Hz, H-1$_{glc}$), 5.06 (1H, d, H-21), 5.09 (1H, dd, *J*=6.9, 14.1 Hz, H-23), 5.27 (1H, br s, H-7), 5.44 (1H, d, *J*=7.2 Hz, H-1$_{xyl}$), 5.60 (1H, d, *J*=8.3 Hz, H-24), 6.50 (1H, br s, H-1$_{rha}$); ^{13}C-NMR data: Tables 1 and 2.

Sapimukoside H (**4**): White amorphous powder. $[\alpha]_D^{24}$ −36.7° (*c*=0.30, MeOH). IR (KBr) ν_{max} cm^{-1}: 3424, 2968, 2934, 1451, 1385, 1066. Negative-ion FAB-MS *m*/*z*: 1070 $[M]^-$ (100), 938 $[M-132]^-$ (8), 923 $[M-146-H]^-$ (6), 791 $[M-146-132-H]^-$ (3); HR-FAB-MS *m*/*z*: 1069.5904 $[M-H]^-$ (Calcd for $C_{55}H_{89}O_{20}$: 1069.5947). ^{1}H-NMR (500 Hz, C_5D_5N) δ: 0.75 (3H, s, Me-19), 0.98 (3H, s, Me-30), 1.01 (3H, s, Me-18), 1.30 (3H, s, Me-29), 1.41 (3H, s, Me-28), 1.27 (3H, t, *J*=6.8 Hz, H-1′), 1.60 (3H, d, *J*=5.9 Hz, H-6$_{rha}$), 1.68 (3H, s, Me-27), 1.69 (3H, s, Me-26), 1.92 (1H, m, H-17), 3.43 (1H, dd, *J*=3.6, 11.9 Hz, H-3), 3.59 (1H, dq, *J*=9.4, 6.8 Hz, H-2′b), 3.98 (1H, dq, *J*=9.4, 6.8 Hz, H-2′a), 4.83 (1H, d, *J*=7.6 Hz, H-1$_{glc}$), 5.07 (1H, dd, *J*=6.5, 14.2 Hz, H-23), 5.16 (1H, br s, H-21), 5.30 (1H, br s, H-7), 5.35 (1H, d, *J*=7.58 Hz, H-1$_{ara'}$), 5.62 (1H, d, *J*=8.3 Hz, H-24), 5.65 (1H, br s, H-1$_{rha'}$), 6.00 (1H, br s, H-1$_{rha}$); ^{13}C-NMR data: Tables 1 and 2.

Sapimukoside I (**5**): White amorphous powder. $[\alpha]_D^{24}$ −30.3° (c=0.33, MeOH). IR (KBr) ν_{max} cm^{-1}: 3423, 2965, 2933, 1450, 1385, 1045. Negative-ion FAB-MS m/z: 1056 $[M]^-$ (100), 924 $[M-132]^-$ (14), 910 $[M-146]^-$ (8), 778 $[M-132-132]^-$ (5), 631 $[M-132-132-132-146]^-$ (2); HR-FAB-MS m/z: 1055.5713 $[M-H]^-$ (Calcd for $C_{54}H_{87}O_{20}$: 1055.5791). ^{1}H-NMR (500 Hz, C_5D_5N) δ: 0.74 (3H, s, Me-19), 0.98 (3H, s, Me-30), 1.02 (3H, s, Me-18), 1.32 (3H, s, Me-29), 1.37 (3H, s, Me-28), 1.60 (3H, d, J=6.2 Hz, H-6_{rha}), 1.66 (3H, s, Me-27), 1.67 (3H, s, Me-26), 1.92 (1H, m, H-17), 3.43 (1H, dd, J=3.0, 11.6 Hz, H-3), 3.49 (1H, s, H-1′), 4.85 (1H, d, J=7.5 Hz, H-1_{glc}), 5.05 (1H, br s, H-21), 5.09 (1H, dd, J=6.2, 12.7 Hz, H-23), 5.29 (1H, br s, H-7), 5.37 (1H, d, J=6.5 Hz, H-$1_{ara'}$), 5.59 (1H, d, J=8.4 Hz, H-24), 5.67 (1H, br s, H-$1_{rha'}$), 6.04 (1H, br s, H-1_{rha}); ^{13}C-NMR data: Tables 1 and 2.

Sapimukoside J (**6**): White amorphous powder. $[\alpha]_D^{24}$ −21.9° (c=0.32, MeOH). IR (KBr) ν_{max} cm^{-1}: 3426, 2967, 2933, 2883, 1449, 1385, 1071. Negative-ion FAB-MS m/z: 791 $[M-H]^-$ (100), 645 $[M-146-H]^-$ (7); HR-FAB-MS m/z: 791.4895 $[M-H]^-$ (Calcd for $C_{44}H_{72}O_{12}$: 791.4946). ^{1}H-NMR (500 Hz, C_5D_5N) δ: 0.73 (3H, s, Me-19), 0.99 (3H, s, Me-30), 1.01 (3H, s, Me-18), 1.30 (3H, s, Me-29), 1.43 (3H, s, Me-28), 1.23 (3H, t, J=7.0 Hz, H-1′), 1.64 (3H, d, J=6.0 Hz, H-6_{rha}), 1.66 (3H, s, Me-27), 1.68 (3H, s, Me-26), 1.89 (1H, m, H-17), 3.46 (1H, dd, J=3.5, 11.6 Hz, H-3), 3.53 (1H, dq, J=9.5, 7.0 Hz, H-2′b), 3.96 (1H, dq, J=9.5, 7.0 Hz, H-2′a), 4.91 (1H, d, J=7.6 Hz, H-1_{glc}), 5.08 (1H, dd, J=6.8, 13.3 Hz, H-23), 5.14 (1H, br s, H-21), 5.29 (1H, br s, H-7), 5.53 (1H, br s, H-1_{rha}), 5.61 (1H, br d, J=8.7 Hz, H-24); ^{13}C-NMR data: Tables 1 and 2.

Acid Hydrolysis of Compounds 1—6 Compound **1**, **4**, **5** (6 mg each) were refluxed with 1 M HCl–dioxane (1 : 1 v/v, 2 ml) on water bath for 4 h. The reaction mixture was neutralized with 1 M NaOH and filtered. The filtrate was extracted with $CHCl_3$ and H_2O. The H_2O-souble fraction was evaporated to dryness. The dried sugar residues was diluted in 1 ml pyridine without water and treated with 0.5 ml trimethyl-chlorsilan (TMCS) and stirred at 60 °C for 5 min. After drying the solution with a stream of N_2, the residue was extracted with ether (1 ml). The ether layer was analyzed by GC with the following conditions: HP AC-5 quartz capillary column (30 m× 0.32 mm); detector: FID (250 °C); injection temperature: 250 °C; column temperature: 180—280 °C; rate: 3 °C/min; and retention times (min): L-arabinose (3.73), L-rhamnose (3.83), and D-glucose (7.22).

By the same procedures carried out for compounds **2** and **3** (6 mg each). The derivatives of D-glucose, D-xylose, L-arabinose and L-rhamnose, were detected; t_R (min): L-arabinose (3.73), L-rnamnose (3.83), D-xylose (4.66), D-glucose (7.22).

Compound **6** (6 mg) was subjected to acid hydrolysis as described for **1** to give D-glucose and L-rhamnose moiety by GC analysis.

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