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Saponins from Leaves of *Acanthopanax senticosus* HARMS., Ciwujia: Structures of Ciwujianosides B, C_1, C_2, C_3, C_4, D_1, D_2 and E

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Eleven triterpenoid saponins were isolated from the leaves of a Chinese folk medicine, *Acanthopanax senticosus* (RUPR. *et* MAXIM) HARMS. Of these compounds, three were identified as known saponins. The structures of eight new saponins named ciwujianosides B (**1**), C_1, (**2**), C_2 (**3**), C_3 (**4**), C_4 (**5**), D_1 (**6**), D_2 (**7**), and E (**8**) were elucidated to be as follows: (**1**), 3-*O*-α-rhamnopyranosyl-(1→2)-β-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-rhamnopyranosyl-(1→4)-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester; (**2**), 3-*O*-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-rhamnopyranosyl-(1→4)-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester; (**3**), 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester; (**4**), 3-*O*-α-arabinopyranosyl oleanolic acid 28-*O*-α-rhamnopyranosyl-(1→4)-β-glucopyranosyl(1→6)-β-glucopyranosyl ester; (**5**), 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl oleanolic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester; (**6**), 3-*O*-α-arabinopyranosyl oleanolic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl(1→6)-β-glucopyranosyl ester; (**7**), 3-*O*-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester; (**8**), 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid.

Keywords——*Acanthopanax senticosus*; Araliaceae; saponin; Chinese folk medicine; ciwujianoside; oleanolic acid glycoside; noroleanolic acid glycoside; 30-norolean-12,20(29)-dien-28-oic acid; ciwujia

The roots of *Acanthopanax senticosus* (RUPR. *et* MAXIM) HARMS (刺五加, Chinese name: ciwujia, Japanese name: ezoukogi) (Araliaceae) have been prescribed in Chinese medicine as a tonic, antirheumatic, and prophylactic for chronic bronchitis, hypertension and ischemic heat disease.[1] Isolation and structure studies of several oleanane-saponins from leaves of this plant were reported previously.[2] As a part of our continuing chemical studies on the glycosides of the Acanthopanax species, we have re-investigated the triterpenoid saponins of leaves of this plant collected in Jilin, North-East district of China. The present paper describes the isolation and structural elucidation of eight new saponins named ciwujianosides and the identification of three known saponins.

The leaves was extracted with methanol and then the residue was extracted with water. The methanol extract was subjected to chromatography on highly porous polymer and the resulting crude saponin fraction was separated by repeated chromatography to give eleven saponins, **1**—**11**. The water extract was separated in the same way as the methanol extract to give saponins **1**—**10**. The combined yields of these saponins, **1**—**11** were 0.05, 0.03, 0.13, 0.03, 0.04, 0.02, 0.07, 0.01, 0.08, 0.01 and 0.005% from the dried leaves, respectively. Of these compounds, the new saponins, **1**—**8**, were named ciwujianosides B (**1**), C_1 (**2**), C_2 (**3**), C_3 (**4**),

C_4 (**5**), D_1 (**6**), D_2 (**7**) and E (**8**), respectively.

Based on analysis of the ^{1}H- and ^{13}C-nuclear magnetic resonance (NMR) spectra and the result of acid hydrolysis as well as comparison of the optical rotation, **10** was proved to be identical with 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl oleanolic acid, which has already been isolated from leaves of this plant.[2]

The saponin **9** afforded oleanolic acid (**12**), glucose, arabinose and rhamnose on acid hydrolysis. In the ^{13}C-NMR spectrum of **9**, the signals due to the aglycone moiety were in good agreement with those of the 28-glycosyl ester of 3-*O*-glycosyl oleanolic acid (bisdesmoside of **12**)[3] and those due to the sugar moiety showed the presence of five monosaccharide units. On selective cleavage of the ester–glycoside linkage with anhydrous LiI and 2,6-lutidine in anhydrous methanol,[4] **9** afforded **10** and a methyl glycoside which was identified as an anomeric mixture of methyl α-rhamnopyranosyl-(1→4)-β-glucopyranosyl-(1→6)-(α and β)-glucopyranoside (**14**) by comparison of its ^{13}C-NMR data with those of an authentic sample.[3] The above evidence led to the formulation of **9** as 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl oleanolic acid 28-*O*-α-rhamnopyranosyl-(1→4)-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester, which has already been obtained from *Hedera helix* and named hederasaponin B.[5]

A new saponin, **5** afforded **12**, glucose, rhamnose and arabinose on acid hydrolysis. The ^{13}C-NMR and ^{1}H-NMR spectra indicated that **5** has an acetyl group (^{13}C-NMR: δ 170.6 and 20.6 and ^{1}H-NMR: δ 1.94, (3H, s)). It is well-known that an acyl linkage on a sugar moiety of bisdesmosidic saponins is highly unstable, being saponified even under mildly alkaline conditions without any cleavage of the relatively hindered glycosyl ester linkage. On mild alkaline hydrolysis,[9] **5** gave a deacetyl product which was identified as **9**. In the ^{13}C-NMR spectra (Table II), on going from **5** to **9**, the signals due to C-6 and C-5 of the central glucose moiety were displaced downfield by 3.3 ppm and upfield by 2.3 ppm, respectively, while other signals remained almost unshifted. This indicated that an acetyl group was located at the 6-hydroxyl group of the central glucosyl unit of the C-28-sugar moiety. From these results, the structure of **5** was established as 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl oleanolic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl(1→6)-β-glucopyranosyl ester.

Acid hydrolysis of **4** gave **12**, arabinose, glucose and rhamnose. Based on analysis of the ^{1}H- and ^{13}C-NMR spectra (Table I), **4** was formulated as a bisdesmoside of **12**. On alkaline hydrolysis, **4** gave a prosapogenin (**13**) which was formulated as 3-*O*-α-arabinopyranosyl oleanolic acid by comparison of the ^{13}C-NMR spectrum of **13** with those of **12** and *d*-menthyl α-L-arabinoside.[6] Comparison of the ^{13}C-NMR spectrum of **4** with those of **13** and **9** led to the structure of **4** as 3-*O*-α-arabinopyranosyl oleanolic acid 28-*O*-α-rhamnopyranosyl-(1→4)-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester.

Acid hydrolysis of **6** afforded **12**, arabinose, glucose and rhamnose. In the ^{13}C-NMR spectrum of **6** (Table I), the signals due to the aglycone moiety were consistent with the formulation as a bisdesmoside of **12**. Alkaline saponification of **6** yielded **13** as a prosapogenin. A ^{1}H-NMR signal at δ 1.94, (3H, s) and ^{13}C-NMR signals at δ 170.5 and 20.6 indicated the presence of an acetyl group in **6**. The mild alkaline hydrolysis[9] of **6** afforded a deacetyl product which was identified as **4**. Comparison of the ^{13}C-NMR spectrum of **6** with that of **4** indicated that an acetyl group was located at the 6-hydroxyl group of the central glucose unit of the C-28-sugar moiety, as in the case of **5**. The mass spectrum (MS) of the trimethylsilyl (TMSi) ether of **6** showed fragment ions at *m/z* 349 [arabinosyl $TMSi_3$]$^+$, 363 [rhamnosyl $TMSi_3$]$^+$ and 711 [(rhamnosyl-glucosyl)$TMSi_5$ Ac]$^+$, supporting this formulation. It follows that the structure of **6** was established as 3-*O*-α-arabinopyranosyl oleanolic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester.

Comparison of the ^{13}C-NMR spectra of **1**, **2**, **3**, **7** and **8** demonstrated that these

saponins are composed of the same sapogenin (Table I). On acid hydrolysis, **1**, **2**, **3** and **7** afforded arabinose, glucose and rhamnose and **8** yielded arabinose and rhamnose, while a genuine aglycone of these saponins was not obtained owing to the acid-catalyzed modification. A genuine aglycone (**15**) could be obtained from **3** by means of hydrolysis with crude hesperidinase.[7]

The infrared (IR) spectrum of **15** showed hydroxyl, carboxyl and double bond absorption bands. The ^{13}C-NMR spectrum suggested that **15** is a noroleanane type triterpene. Carbon signals observed at δ 144.1 (C=), 123.0 (=CH), 149.0 (C=) and 107.0 ($=CH_2$) and proton signals at δ 4.74 (2H, s, $C=CH_2$) and 5.48 (1H, t, H−C=C), showed the presence of one trisubstituted double bond and one *exo*-methylene group in **15** (Table I). In 1986, Ahmad *et al.* isolated saponin-1 and guaianin (**16**) from stem bark of *Guaiacum officinale*.[8] Although they could not obtain the genuine aglycone from these saponins they assigned the structures of 3-*O*-glycosides of 30-norolean-12,20(29)-dien-28-oic acid to saponin-1 and **16** (see Chart 1). Taking the glycosylation shifts on C-2 and -3 signals into account, the comparison of the ^{13}C-NMR signals of **15** with those of saponin-1 and **16** led to the identification of **15** as the common genuine aglycone of saponin-1 and **16**, *i.e.*, 30-norolean-12,20(29)-dien-28-oic acid.

The saponin **11** was proved to be identical with saponin-1, 3-*O*-α-arabinopyranoside of **15** by comparing its $[\alpha]_D$, ^{1}H- and ^{13}C-NMR spectra with the reported data.[8]

In the ^{13}C-NMR spectrum of **8**, the aglycone signals (Table I) and signals due to sugar moieties (Table II) were almost superimposable on those of **15** and **10**, respectively. It follows that **8** can be formulated as 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl-30-norolean-12, 20(29)-dien-28-oic acid.

The ^{13}C-NMR spectrum of **1** indicated that **1** is a bisdesmoside of **15** (Table I). The selective cleavage of the ester–glycoside linkage (*vide supra*) of **1** gave **8** as a prosapogenin and **14**. Further, the carbon signals due to the sugar moiety of **1** were found to be almost superimposable on those of **9**. These observations led to the formulation of **1** as 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-rhamnopyranosyl-(1→4)-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester.

In the ^{13}C-NMR spectrum of **2**, comparison of the sugar carbon signals with those of **4** (Table II) and of those due to the aglycone moiety with those of **15** (Table I) led to the formulation of **2** as 3-*O*-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-arabinopyranosyl-(1→4)-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester. On selective cleavage of the ester–glycoside linkage (*vide supra*), **2** afforded **11** as a prosapogenin and **14**, supporting this formulation. The MS of the acetate of **2**, which showed fragment ions at *m/z*: 259 [(arabinose)Ac_3]$^+$, 273 [(rhamnose)Ac_3]$^+$, 561 [(rhamnose-glucose)Ac_6]$^+$ and 849 [(rhamnose-glucose-glucose)Ac_9]$^+$, was also consistent with this structure.

In the ^{13}C-NMR spectrum of **3** (Table I), the signals due to the aglycone moiety were in good agreement with those of **1**. A proton signal at δ 1.94 (3H, s) and carbon signals at δ 170.4 and 20.6 indicated the presence of an acetyl group in **3**. On selective cleavage of the ester–glucoside linkage (*vide supra*), **3** afforded **8** as a prosapogenin and **14**. On mild alkaline hydrolysis,[9] **3** gave a deacetyl product that was identified as **1**. The MS of the TMSi ether of **3** exhibited fragment ions at *m/z* 711 [(rhamnosyl-glucosyl)AcTMSi_5]$^+$, and 621 [(rhamnosyl-arabinosyl)TMSi_5]$^+$ and 363 [(rhamnosyl)TMSi_3]$^+$. In a comparison of the ^{13}C-NMR spectrum of **3** with that of **1**, acylation shifts were observed at C-5 and C-6 of the central glucose moiety as in going from **9** to **5**. On the basis of these results coupled with a comparison of the carbon signals with those of **5**, the structure of **3** can be assigned as 3-*O*-α-rhamnopyranosyl-(1→2)-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester.

Alkaline saponification of **7** afforded **11** as a prosapogenin. The presence of an acetyl

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

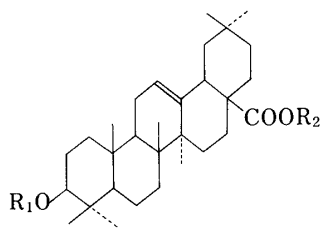
	12	**9**	**10**	**6**	**13**	**4**	**5**	**15**	**3**	**8**	**1**	**2**	**11**	**7**
C-1	38.9	39.0	38.8	38.8	38.8	38.9	38.9	38.9	38.9	38.8	38.9	38.9	38.8	38.9
C-2	28.2	26.4	26.5	26.6	26.7	26.6	26.4	28.3	26.4	26.6	26.4	26.5	26.7	26.7
C-3	78.0	88.7	88.8	88.6	88.7	88.7	88.8	78.1	88.8	88.8	88.8	88.7	88.7	88.8
C-4	39.4[a]	39.4[a]	39.4[a]	39.5[a]	39.6[a]	39.5[a]	39.4[a]	39.3[a]	39.5[a]	39.4[a]	39.4[a]	39.5[a]	39.6[a]	39.6[a]
C-5	55.8	55.9	55.9	55.9	55.9	55.9	55.9	55.8	55.9	55.9	55.9	55.9	55.9	55.9
C-6	18.8	18.5	18.5	18.4	18.5	18.5	18.5	18.4	18.4	18.5	18.4	18.5	18.5	18.5
C-7	33.3	33.1	33.2	33.1	33.3	33.1	33.1	33.2	33.0	33.2	33.1	33.0	33.2	33.1
C-8	39.8[a]	39.8[a]	39.7[a]	39.8[a]	39.8[a]	39.9[a]	39.9[a]	39.7[a]	39.9[a]	39.7[a]	39.9[a]	39.9[a]	39.8[a]	39.9[a]
C-9	48.1	48.1	48.0	48.0	48.1	48.0	48.0	48.0[c]	48.0	48.0[c]	48.0	48.0	48.0	48.1
C-10	37.4	37.0	37.0	37.0	37.0	37.0	37.0	37.3	36.9	37.0	37.0	36.9	37.0	37.0
C-11	23.8	23.2[b]	23.7	23.4[b]	23.3[b]	23.7	23.7	23.7	23.6[b]	23.7[b]	23.5	23.6[b]	23.8	23.7
C-12	122.5	122.8	122.5	122.9	122.5	122.6	122.9	123.0	122.5	122.9	122.6	123.3	123.0	123.4
C-13	144.8	144.0	144.7	144.0	144.0	144.1	144.0	144.1	143.3	144.3	143.4	143.2	144.2	143.4
C-14	42.0	42.1	42.1	42.1	42.0	42.1	42.1	42.0	41.6	42.0	41.7	41.6	42.0	42.1
C-15	28.3	28.1	28.3	28.2	28.3	28.2	28.1	28.0	28.1	28.1	28.1	28.2	28.3	28.3
C-16	23.8	23.6[b]	23.7	23.7[b]	23.8[b]	23.7	23.7	23.7	23.8[b]	23.5[b]	23.5	23.5[b]	23.8	23.7
C-17	46.7	47.0	46.6	47.0	46.7	47.0	47.0	47.0	47.5	47.0	47.3[b]	47.3[c]	47.1	47.3[b]
C-18	42.0	41.7	41.9	41.7	42.2	41.9	41.6	47.9[c]	47.5	47.9[c]	47.5[b]	47.5[c]	48.0	47.5[b]
C-19	46.7	46.7	46.2	46.2	46.5	46.2	46.2	42.0	42.0	42.1	42.1	42.0	42.1	41.7
C-20	31.0	30.7	30.7	30.7	30.6	30.6	30.7	149.0	148.1	149.1	148.3	148.3	149.1	148.4
C-21	34.3	34.0	34.2	34.0	34.3	34.0	34.2	30.4	30.1	30.4	30.1	30.4	30.4	30.1
C-22	33.3	32.6	33.2	32.9	33.3	32.8	32.6	38.3	37.8	38.5	37.8	37.8	38.4	37.6
C-23	28.7	28.1	28.0	28.2	28.3	28.2	28.2	28.1	28.8	28.1	28.1	28.2	28.3	28.3
C-24	16.5	16.9	17.0	16.9	17.0	16.9	16.9	16.5	16.9	17.0	17.0	16.9	17.0	16.9
C-25	15.5	15.6	15.5	15.6	15.5	15.6	15.6	15.5	15.6	15.5	15.6	15.6	15.5	15.6
C-26	17.5	17.4	17.3	17.4	17.4	17.5	17.4	17.3	17.4	17.3	17.5	17.4	17.4	17.5
C-27	26.2	26.0	26.1	26.1	26.2	26.1	26.0	26.1	26.0	26.1	26.0	26.0	26.2	26.0
C-28	180.2	176.4	180.1	176.4	180.2	176.5	176.4	179.3	175.6	179.4	175.7	175.7	179.4	175.7
C-29	33.3	33.1	33.2	33.1	33.3	33.1	33.1	107.4	107.4	107.1	107.5	107.4	107.1	107.5
C-30	23.8	23.6[b]	23.7	23.7[b]	23.8[b]	23.7	23.7							

a—*c*) Assignments in any column may be reversed.

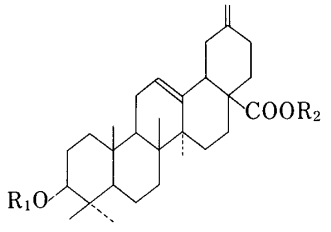
TABLE II. ^{13}C-NMR Chemical Shifts of Sugar Moieties in C_5D_5N

		9	10	6	13	4	5	3	8	1	2	11	7
3-*O*-Sugar moieties													
ara inner	1	104.7	104.8	107.4	107.5	107.5	104.6	104.7	104.8	104.9	107.4	107.3	107.5
	2	75.9	75.9	72.8	72.9	72.8	75.9	75.9	75.9	75.9	72.9	72.8	72.9
	3	73.9	74.0[a]	74.6	74.7	74.6	73.7	73.7	73.7[a]	73.8[b]	74.6	74.5	74.6
	4	68.5	69.9	69.4	69.5	69.5[a]	68.5	68.4	68.6	68.6	69.5[a]	69.4	69.5
	5	64.5	64.6	66.7	66.7	66.7	64.4	64.5	64.6	64.5	66.7	66.6	66.7
rha terminal	1	101.7	101.6				101.7	101.7	101.5	101.7			
	2	72.5	72.5				72.3	72.1	72.4	72.4			
	3	72.7	72.5				72.5	72.4	72.6	72.6			
	4	73.9	73.7[a]				73.7	73.5	74.0[a]	73.8[b]			
	5	69.7[a]	68.6				69.3[a]	69.6	69.8	69.9[a]			
	6	18.5	18.5				18.5	18.4	18.6	18.6			
28-*O*-Sugar moieties													
glc inner	1	95.6		95.5		95.6	95.7	95.5		95.8	95.7		95.7
	2	73.9		73.7		73.9	73.7	73.5		73.9[b]	73.8		73.7
	3	78.2[b]		78.6		78.7[b]	78.6[b]	78.5[a]		78.3[c]	78.2[b]		78.2
	4	70.5		70.8[a]		70.9	70.8	70.4		70.3	70.8		70.9
	5	76.4		78.0		78.0	78.0	77.9		78.1[c]	77.8		77.9
	6	69.9[a]		69.4		69.3[a]	69.8[a]	69.6		69.9[a]	69.5[a]		69.5
glc outer	1	104.7		104.7		104.8	104.6	104.7		104.8	104.8		104.7
	2	75.2		75.0		75.3	75.0	74.8		75.3	75.2		75.0
	3	76.4		76.3		76.5	76.3	76.1		76.5	76.4		76.3
	4	78.6[b]		79.1		78.3[b]	79.1[b]	79.0[a]		78.7[c]	78.6[b]		79.1
	5	77.0		73.7		77.1	73.7	73.5		77.2	77.1		73.7
	6	61.3		63.7		61.3	63.6	63.7		61.4	61.6		63.7
rha terminal	1	102.6		102.8		102.7	102.8	102.9		102.7	102.6		102.8
	2	72.5		72.3		72.3	72.3	72.1		72.6	72.5		72.3
	3	72.3		72.6		72.6	72.3	72.4		72.8	72.7		72.6
	4	73.9		73.7		72.9	72.5	73.5		73.9[b]	73.9		73.7
	5	70.3[a]		70.6[a]		70.3	70.6	70.4		70.3[a]	70.2[a]		70.6
	6	18.5		18.4		18.5	18.5	18.4		18.5	18.5		18.5
$CH_3\underline{C}O$				170.5			170.6	170.4					170.6
$\underline{C}H_3CO$				20.6			20.6	20.6					20.6

a—*c*) Assignments in any column may be reversed.



	R_1	R_2
4	-ara	-glc^{6}-glc^{4}-rha
5	-ara^{2}-rha	-glc^{6}-glc^{4}-rha ^{6}Ac
6	-ara	-glc^{6}-glc^{4}-rha ^{6}Ac
9	-ara^{2}-rha	-glc^{6}-glc^{4}-rha
10	-ara^{2}-rha	H
12	H	H
13	-ara	H



	R_1	R_2
1	-ara^{2}-rha	-glc^{6}-glc^{4}-rha
2	-ara	-glc^{6}-glc^{4}-rha
3	-ara^{2}-rha	-glc^{6}-glc^{4}-rha ^{6}Ac
7	-ara	-glc^{6}-glc^{4}-rha ^{6}Ac
8	-ara^{2}-rha	H
11	-ara	H
15	H	H
16	-ara^{3}-glc^{3}-rha	H

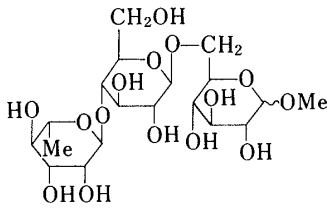


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Chart 1

group in **7** was demonstrated by the ^{13}C- and ^{1}H-NMR spectra. The mild alkaline saponification[9] of **7** gave a deacetyl product that was identical with **2**. It was found that the sugar carbon signals of **7** were almost superimposable on those of **6** (Table II). From these results, the structure of **7** was established as 3-*O*-α-arabinopyranosyl-30-norolean-12,20(29)-dien-28-oic acid 28-*O*-α-rhamnopyranosyl-(1→4)-6-*O*-acetyl-β-glucopyranosyl-(1→6)-β-glucopyranosyl ester.

Previously, Russian chemists reported the isolation and structure determination of four saponins of **12** from leaves of this plant.[2] However, in the present study, these saponins other than **10** have not been identified as yet.

Experimental

General Procedures——Optical rotations were measured with a Union PM-101 automatic digital polarimeter. IR spectra were taken on a Shimadzu IR-408 spectrometer. NMR spectra were recorded on a JEOL FX-100 spectrometer in C_5D_5N solution using tetramethylsilane (TMS) as an internal standard. For gas liquid chromatography (GLC), a Shimadzu GC-6A was used. MS were taken on a JEOL JMS-01-SG-2 spectrometer by the direct inlet method; ionization voltage 75 eV. For column chromatography, Kieselgel 60 (70—230 mesh, Merck), LiChroprep RP-8 (40—63 μm, Merck), Hydroxylapatite (Asahi Optical Ind. Co., Ltd.) and Diaion HP-20 (Mitsubishi Chem. Ind. Co., Ltd.) were used. All solvent systems for chromatography were homogeneous.

Acetylation and Trimethylsilylation for MS: see previous paper.[3]

Acid Hydrolysis of Saponins and Identification of the Resulting Monosaccharides: see the previous paper.[3]

Identification of the known saponins and prosapogenins was substantiated by comparison of the ^{13}C-NMR spectrum, sugar composition by acid hydrolysis and optical rotation with those of a respective authentic sample.

Extraction and Separation of Saponin——The dried leaves of *Acanthopanax senticosus* (1.7 kg), collected in Jilin, China, were extracted with MeOH at room temperature and then the residue was extracted with water to give MeOH extract (98 g) and water extract (226 g). The MeOH extract was chromatographed on a column of highly porous polymer (Diaion HP-20) and eluted with H_2O, 60% MeOH and MeOH, successively. The fraction eluted with MeOH was subjected to chromatography on silica gel. Elution with $CHCl_3$–MeOH–H_2O (30 : 10 : 1) provided five fractions (fr. A, B, C, D, E and F, in increasing order of *Rf* on silica gel TLC, developed with $CHCl_3$–MeOH–H_2O, 6 : 4 : 1). Fr. F was further purified by rechromatography on silica gel under the same conditions as above to yield **11**. Fr B, C, D and E were each subjected to chromatography on a reverse-phase column (LiChroprep RP-8) and then purified by high-performance liquid chromatography (HPLC) (column, TSK-GEL ODS-120T, 21 mm × 30 cm; solvent, 70%

MeOH; flow rate, 6 ml/min; detection, RI) to give **1** and **9** from fr. B, **7** and **6** from fr. D and **10** and **8** from fr. E. Fr. C was subjected to chromatography on a hydroxylapatite column (solvent, 85% CH_3CN aq.) to provide **2**—**5**. The water extract was also separated through the same procedure as used for the MeOH extract to give **1**—**10**. Total yields of **1**, **2**, **3**, **4**, **5**, **6**, **7**, **8**, **9**, **10** and **11** were 0.05, 0.03, 0.13, 0.03, 0.04, 0.02, 0.07, 0.01, 0.08, 0.01 and 0.005%, respectively.

10: A white powder, $[\alpha]_D^{23}$ +10.9° (c=0.55, MeOH). ^{1}H-NMR δ: 0.85 (3H, s), 1.01 (9H, s), 1.06 (3H, s), 1.19(3H, s), 1.32 (3H, s), 1.64 (1H, d, J=6 Hz, Me of rhamnoside), 4.91 (1H, d, J=6 Hz, anomeric proton of arabinoside), 4.50 (1H, br s, 12-H), 6.18 (1H, s, anomeric proton of rhamnoside).

9: A white powder, $[\alpha]_D^{18}$ −21.4° (c=1.26, MeOH). *Anal.* Calcd for $C_{59}H_{96}O_{25}\cdot 4H_2O$: C, 55.47; H, 8.21. Found: C, 55.32; H, 7.97. ^{1}H-NMR δ: 0.92 (9H, s), 1.09 (6H, s), 1.19 (3H, s), 1.28 (3H, s), 1.64, 1.72 (each 3H, d, J=6 Hz, Me of rhamnosides), 4.92 (1H, d, J=7Hz, anomeric proton), 5.01 (1H, d, J=6 Hz, anomeric proton), 5.44 (1H, br s, 12-H), 5.85, 6.18 (each 1H, s, anomeric protons of rhamnosides), 6.24 (1H, d, J=7 Hz, anomeric proton). On mineral acid hydrolysis, **9** yielded **12**, glucose, arabinose and rhamnose. Compound **9** was identified as hederasaponin B by comparison of $[\alpha]_D$, and the ^{1}H- and ^{13}C-NMR spectra.

Selective Cleavage of the Ester–Glycoside Linkage of 9[4]——A solutiof of **9** (150 mg), anhydrous LiI (60 mg) and 2,6-lutidine (4 ml) in anhydrous MeOH (2 ml) was refluxed for 16 h. The reaction mixture was deionized with Amberlite MB-3 resin and concentrated to dryness. The residue was chromatographed on silica gel (solvent: $CHCl_3$–MeOH, 6:1) to give **10** (23 mg, *vide infra*) and a methyl oligoglycoside (**14**) (15 mg); the latter was identified by comparison of the ^{13}C-NMR spectrum with that of an authentic sample.[3]

4: A white powder, $[\alpha]_D^{18}$ −7.68° (c=0.52, MeOH). *Anal.* Calcd for $C_{53}H_{86}O_{21}\cdot 6H_2O$: C, 54.59; H, 7.81. Found: C, 54.61; H, 7.78. ^{1}H- NMR δ: 0.96 (9H, s), 0.98 (3H, s), 1.12 (3H, s), 1.24 (6H, s), 1.72 (1H, d, J=6 Hz, Me of rhamnoside), 4.96 (1H, d, J=7 Hz, anomeric proton), 5.02 (1H, d, J=7 Hz, anomeric proton), 5.42 (1H, s, 12-H), 5.84 (1H, s, anomeric proton of rhamnoside), 6.28 (1H, d, J=7 Hz, anomeric proton). On mineral acid hydrolysis, **4** yielded **12**, glucose, arabinose and rhamnose.

5: A white powder, $[\alpha]_D^{18}$ −23.67° (c=0.51, MeOH). *Anal.* Calcd for $C_{61}H_{98}O_{26}\cdot 5H_2O$: C, 54.83; H, 7.91. Found: C, 54.73; H, 7.63. ^{1}H-NMR δ: 0.89 (6H, s), 0.92 (3H, s), 1.08 (6H, s), 1.16 (3H, s), 1.25 (3H, s), 1.63, 1.70 (each 3H, d, J=6 Hz, Me of rhamnosides), 1.94 (3H, s, CH_3CO), 4.92 (1H, d, J=7 Hz, anomeric proton), 5.01 (1H, d, J=7 Hz, anomeric proton), 5.42 (1H, s, 12-H), 5.53, 5.82 (each 1H, s, anomeric protons of rhamnosides), 6.22 (1H, d, J=7 Hz, anomeric proton). On mineral acid hydrolysis, **5** yielded **12**, glucose, arabinose and rhamnose.

Deacetylation of 5[9]——A solution of **5** (35 mg) in 0.05 N KOH aq. (1 ml) was allowed to stand at 4 °C for 24 h. The reaction mixture was neutralized with 1% HCl and was deionized with Amberlite MB-3 resin. Then the mixture was extracted with BuOH, and the BuOH layer was concentrated to dryness to give **9** (26 mg).

11: A white powder, $[\alpha]_D^{23}$ +60.78° (c=0.51, C_5H_5N). ^{1}H-NMR: 0.83, 0.91, 0.94, 1.20, 1.26 (each 3H, s), 4.72 (1H, d, J=7 Hz, anomeric proton of arabinoside), 4.75 (2H, s, 29-H_2), 5.43 (1H, s, 12-H). On mineral acid hydrolysis, **11** afforded arabinose.

8: A white powder, $[\alpha]_D^{23}$ +55.81° (c=0.43, MeOH). *Anal.* Calcd for $C_{40}H_{62}O_{11}\cdot 2H_2O$: C, 60.81; H, 8.80. Found: C, 60.68; H, 8.82. ^{1}H-NMR δ: 0.84, 0.97, 1.07, 1.19, 1.28 (each 3H, s), 1.62 (3H, d, J=6 Hz, Me of rhamnoside), 4.76 (2H, s, 29-H_2), 4.92 (1H, d, J=6 Hz, anomeric proton of arabinoside), 5.49 (1H, s, 12-H), 6.13 (1H, s, anomeric proton of rhamnoside). On mineral acid hydrolysis, **8** yielded a rhamnose and an arabinose.

1: A white powder, $[\alpha]_D^{17}$ +1.32° (c=0.76, MeOH). *Anal.* Calcd for $C_{58}H_{92}O_{25}\cdot 3H_2O$: C, 56.08; H, 7.81. Found: C, 56.29; H, 7.91. ^{1}H-NMR δ: 0.90 (3H, s), 1.10 (6H, s), 1.24 (6H, s), 1.66, 1.73 (each 1H, d, J=6 Hz, Me of rhamnosides), 4.95 (1H, d, J=7 Hz, anomeric proton), 5.02 (1H, d, J=7 Hz, anomeric proton), 5.47 (1H, s, 12-H), 5.57, 5.88 (each 1H, s, anomeric proton of rhamnosides), 6.21 (1H, d, J=7 Hz, anomeric proton). On mineral acid hydrolysis, **1** afforded glucose, arabinose and rhamnose. On selective cleavage of the ester glycosyl linkage (*vide supra*), **1** (130 mg) yielded **8** (15 mg) and **14** (12 mg), which were identified by comparison of the ^{1}H-NMR and ^{13}C-NMR spectra with those of an authentic sample.[3]

2: A white powder, $[\alpha]_D^{18}$ +14.6° (c=1.03, MeOH). *Anal.* Calcd for $C_{52}H_{82}O_{21}$: C, 59.92; H, 7.93. Found: C, 59.69; H, 8.19. ^{1}H-NMR δ: 0.89, 0.97, 1.08, 1.24, 1.28 (each 3H, s), 1.73 (3H, d, J=6.8 Hz, Me of rhamnoside), 4.92 (1H, d, J=7 Hz, anomeric proton), 4.98 (1H, d, J=7 Hz, anomeric proton), 5.86 (1H, s, anomeric proton of rhamnoside), 6.24 (1H, d, J=8 Hz, anomeric proton), 5.47 (1H, s, 12-H). On mineral acid hydrolysis, **2** yielded glucose, rhamnose and arabinose.

3: A white powder, $[\alpha]_D^{18}$ −6.93° (c=1.01, MeOH). *Anal.* Calcd for $C_{60}H_{94}O_{26}$: C, 57.63; H, 7.82. Found: C, 57.59; H, 7.65. ^{1}H-NMRδ: 0.89 (6H, s), 0.97 (3H, s), 1.08 (3H, s), 1.24 (3H, s), 1.64, 1.70 (each 3H, d, J=6 Hz, Me of rhamnosides), 1.94 (3H, s, CH_3CO), 4.92 (1H, d, J=7 Hz, anomeric proton), 4.98 (1H, d, J=6 Hz, anomeric proton), 5.52, 5.83 (each 1H, s, anomeric proton of rhamnosides), 6.19 (1H, d, J=7 Hz, anomeric proton). On mineral acid hydrolysis, **3** yielded glucose, arabinose and rhamnose.

Enzymic Hydrolysis of 2 and 3——A solution of **2** or **3** (250 mg) and crude hesperidinase (400 mg, Tanabe Pharm., Co., Ltd., Osaka, Japan) in H_2O (25 ml) was incubated at 37 °C for 7 d. The reaction mixture was diluted with water and then extracted with $CHCl_3$. The $CHCl_3$ extract was evaporated to dryness and then subjected to chromatography on silica gel (C_6H_6: acetone (4:1)) to give **15** (51 mg).

15: A white powder, $[\alpha]_D^{18}$ +159.3° (c=0.54, pyridine). *Anal.* Calcd for $C_{29}H_{44}O_3$: C, 76.86; H, 9.90. Found: C,

76.80; H, 10.14. IR (KBr): 3440 (OH), 1680 (COOH), 1640, 885 (C=CH_2), 825 (H-C=C) cm^{-1}. ^{1}H-NMR δ: 0.89, 0.98, 1.01, 1.19, 1.26 (each 3H, s), 2.66 (1H, t, 18-H), 3.35 (1H, m, 3-H), 4.74 (2H, s, C=CH_2), 5.48 (1H, t, H-C=C).

Selective Cleavage of the Ester–Glycoside Linkage of 2 and 3[4]——A solution of **2** or **3** (200 mg), anhydrous LiI (80 mg) and 2,6-lutidine (4 ml) in anhydrous MeOH was treated as described above to give **11** (6 mg) from **2** and **8** (28 mg) from **3** together with **14**, which was identified by comparison of the ^{13}C-NMR spectrum with that of an authentic sample.[3]

Deacetylation of 3[9]——A solution of **3** (40 mg) in 0.05 N KOH aq. (1 ml) was allowed to stand at 4 °C for 24 h. The reaction mixture was neutralized with 1% HCl, and deionized with Amberlite MB-3 resin. Then, the mixture was extracted with BuOH and the BuOH layer was concentrated to dryness to give **1** (31 mg).

7: A white powder, $[\alpha]_D^{16}$ +17.65° (c=0.51, MeOH). *Anal.* Calcd for $C_{54}H_{84}O_{22}\cdot 2H_2O$: C, 57.6; H, 7.85. Found: C, 57.3; H, 7.97. ^{1}H-NMR δ: 0.91, 0.98, 1.08, 1.24, 1.29 (each 3H, s), 1.70 (3H, d, J=6 Hz, Me of rhamnoside), 1.92 (3H, s, CH_3CO), 4.98 (1H, d, J=7.8 Hz, anomeric proton), 5.56 (1H, s, anomeric proton of rhamnoside), 5.46 (1H, s, 12-H), 6.20 (1H, d, J=7 Hz, anomeric proton). On mineral acid hydrolysis, **7** yielded glucose, arabinose and rhamnose. The deacetylation of **7** (30 mg) by the aforementioned procedure afforded **2** (22 mg).

6: A white powder, $[\alpha]_D^{18}$ −9.8° (c=0.41, MeOH). *Anal.* Calcd for $C_{55}H_{88}O_{22}\cdot 3H_2O$: C, 57.19; H, 8.15. Found: C, 56.81; H, 8.01. ^{1}H-NMR δ: 0.92 (9H, s), 0.98 (3H, s), 1.07 (3H, s), 1.26 (6H, s), 1.69 (3H, d, J=6 Hz, Me of rhamnoside), 1.93 (3H, s, CH_3CO), 4.99 (1H, d, J=7.6 Hz, anomeric proton), 5.53 (1H, s, anomeric proton of rhamnoside), 6.23 (1H, d, J=7 Hz, anomeric proton). On mineral acid hydrolysis, **6** gave **12**, arabinose, glucose and rhamnose.

Alkaline Hydrolysis of 6——**6** (30 mg) was hydrolyzed by heating in 0.5 N KOH aq. (2 ml) at 90 °C for 1 h. The reaction mixture was neutralized with Amberlite MB-3 resin and then partitioned between H_2O and BuOH. The BuOH layer was concentrated dryness to give **13**.

Deacetylation of 6[9]——A solution of **6** (40 mg) in 0.05 N KOH aq. (2 ml) was allowed to stand at 4 °C for 24 h and then treated as described above to provide **4** (29 mg).

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