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Oleanane saponins from *Gymnema sylvestre*

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Abstract

Six oleanane-type saponins, along with two known triterpene saponins, were isolated from the leaves of *Gymnema sylvestre*. The structures of the oleanane triterpene glycosides were characterized as longispinogenin 3-*O*-β-D-glucuronopyranoside, 21β-benzoylsitakisogenin 3-*O*-β-D-glucuronopyranoside, 3-*O*-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl oleanolic acid 28-*O*-β-D-glucopyranosyl ester, oleanolic acid 3-*O*-β-D-xylopyranosyl(1 → 6)-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranoside, 3-*O*-β-D-xylopyranosyl(1 → 6)-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl oleanolic acid 28-*O*-β-D-glucopyranosyl ester and 3-*O*-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl oleanolic acid 28-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl ester on the basis of hydrolysis and spectral evidence, including 1D- and 2D-NMR (TOCSY, ROESY, HMQC and HMBC) and FABMS analyses. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: *Gymnema sylvestre*; Asclepiadaceae; Oleanane saponins

1. Introduction

The plant *Gymnema sylvestre* R. Br. (Asclepiadaceae) is a vine which grows in the southern part of China, including the Guangdong, Guangxi and Fujian provinces. This plant species is also distributed in India where it is used as a stomachic, diuretic, and anti-diabetic remedy (Sastri, 1956; Gharpurey, 1926). The total saponin fraction of the leaves, commonly known as 'gymnemic acid', has an anti-sweetening effect (Suttisri, Lee & Kinghorn, 1995), and was shown to be able to inhibit glucose absorption in the small intestine and to suppress elevated glucose levels in blood following the administration of sucrose in rats (Shimizu et al., 1997; Yashioka, 1986). In addition, it was suggested that blood glucose homeostasis was achieved through an increase in serum insulin levels provided by a repair or regeneration of the pancreatic tissues (Shanmugasundaram, Gopinath, Shanmugasundaram & Rajendran, 1990). Since the gymnemic acid fraction is allegedly beneficial for treating diabetes and obesity, preparations containing Gymnema plants of Indian origin are now commercially available in Japan, Germany, and the USA as health foods.

As part of a continuing study of the saponin constituents of medicinal plants (Ye et al., 1995, 1996; Ye, He, Zhao & Che, 1998), we have now examined the saponin fraction of *G. sylvestre* leaves grown in the Guangxi region of China. Chemical purification of the total saponin fraction led to the isolation of eight saponins, six of which were determined to be unreported structures (Chandel & Rastogi, 1980; Shashi, Sudip & Gurudas, 1988; Shashi & Ashoke, 1991; Hostettmann & Marston, 1995). This paper reports the isolation and characterization of these saponins. The structural elucidation was accomplished by extensive spectroscopic studies, including the use of 2D-NMR spectroscopic methods. Our results showed that the aglycone structures of the major saponin constituents

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of *G. sylvestre* leaves obtained from China are different from those reported of the Indian species.

2. Results and discussion

The EtOH extract of the dried leaves of *G. sylvestre* was partitioned between *n*-BuOH and H_2O, and the *n*-BuOH solution was subjected to silica gel and C_{18} columns to afford the new saponins **1**–**6**, together with two known compounds, which were identified as oleanolic acid 28-*O*-β-D-glucopyranosyl ester and oleanolic acid 3-*O*-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranoside by direct comparison of their spectral data and physical properties with literature values (Kizu & Tomimori, 1982; Lin & Shoji, 1979).

The molecular formulae for all compounds were determined by FABMS in combination with ^{13}C DEPT data. The configurations at the anomeric positions of the glucopyranosyl, glucuronopyranosyl and xylopyranosyl residues were determined to have a β configuration on the basis of ^{1}H- and ^{13}C-NMR spectral data (Tables 1 and 2). Furthermore, a D absolute configuration of these sugar residues was assumed, consistent with the stereochemistry of naturally occurring monosaccharides.

The positive FABMS of **1** showed a quasi-molecular

Table 1
^{13}C-NMR spectral data of the sugar moieties of compounds **1**–**6**[a]

Carbon		**1**	**2**	**3**	**4**	**5**	**6**
C-3							
GluA	1	107.3	107.3				
	2	75.6	75.6				
	3	78.2	78.2				
	4	73.5	73.6				
	5	77.8	77.7				
	6	173.1	173.3				
Glc	1			106.9	107.0	107.0	106.9
	2			75.1	75.0	75.0	75.2
	3			78.4	78.3	78.3	78.4
	4			71.6	71.5	71.5	71.5
	5			77.0	77.0	77.0	77.0
	6			**70.4**	**70.4**	**70.4**	**70.5**
Glc′	1			105.4	105.4	105.4	105.4
	2			75.5	75.6	75.6	75.6
	3			78.5	78.5	78.5	78.6
	4			71.7	71.6	71.6	71.7
	5			78.4	76.9	76.9	78.5
	6			62.7	**69.8**	**69.8**	62.6
Xyl	1				106.0	106.0	
	2				74.9	74.9	
	3				78.0	78.1	
	4				71.1	71.1	
	5				67.0	67.1	
C-28							
Glc′′	1			95.7		95.8	95.7
	2			74.1		74.1	73.9
	3			78.8		78.9	78.7
	4			71.0		71.1	70.9
	5			79.3		79.3	78.0
	6			62.1		62.2	**69.3**
Glc′′′	1						105.3
	2						75.2
	3						78.5
	4						71.7
	5						78.4
	6						62.7

[a] Spectra were measured in pyridine-d_5. Assignments were established by interpretation of ^{13}C DEPT, HMQC and HMBC spectra. Values given in boldface indicate the glycosidic positions of the aglycone.

Table 2
^{1}H-NMR spectral data of the saccharide moieties of compounds **3**–**6**[a,b]

Proton		**3**	**4**	**5**	**6**
C-3					
glc	1	4.86 *d* (7.8)	4.86 *d* (7.7	4.87 *d* (7.7)	4.88 *d* (7.7)
	2	4.04[c]	4.02[c]	4.03[c]	4.05[c]
	3	4.20[c]	4.16[c]	4.17[c]	4.21[c]
	4	4.23[c]	4.16[c]	4.16[c]	4.23[c]
	5	4.17[c]	4.11[c]	4.11[c]	4.17[c]
	6a	4.90 *d* (11.0)	4.90 *d* (10.6)	4.91 *d* (10.7)	4.90 *d* (10.9)
	6b	4.37 *dd*	4.34 *dd*	4.34 *dd*	4.36 *dd*
		(11.0, 4.9)	(10.6, 4.9)	(10.7, 4.8)	(10.9, 4.9)
glc′	1	5.11 *d* (7.3)	5.05 *d* (7.8)	5.05 *d* (7.7)	5.13 *d* (7.7)
	2	4.11[c]	3.98[c]	3.98[c]	4.10[c]
	3	4.22[c]	4.20[c]	4.20[c]	4.22[c]
	4	4.14[c]	4.15[c]	4.14[c]	4.14[c]
	5	3.98[c]	4.12[c]	4.12[c]	3.96[c]
	6a	4.49 *d* (11.0)	4.79 *d* (10.6)	4.79 *d* (10.6)	4.50 *d* (11.0)
	6b	4.36 *dd*	4.33 *d*	4.32 *dd*	4.37 *dd*
		(11.0, 4.6)	(10.6, 4.8)	(10.6, 4.9)	(11.0, 4.5)
xyl	1		4.98 *d* (7.6)	4.96 *d* (7.4)	
	2		4.03[c]	4.02[c]	
	3		4.14[c]	4.14[c]	
	4		4.36[c]	4.35[c]	
	5a		3.67 *d* (10.5)	3.65 *d* (10.3)	
	5b		4.32 *dd*	4.33 *dd*	
C-28			(10.5, 4.5)	(10.3, 4.8)	
glc′′	1	6.31 *d* (8.3)		6.33 *d* (8.1)	6.26 *d* (8.1)
	2	4.20[c]		4.19[c]	4.15[c]
	3	4.29[c]		4.30[c]	4.23[c]
	4	4.36[c]		4.36[c]	4.37[c]
	5	4.03[c]		4.04[c]	4.21[c]
	6a	4.46 *d* (11.5)		4.48 *d* (11.8)	4.72 *d* (10.5)
	6b	4.40 *dd*		4.41 *dd*	4.38 *dd*
		(11.5, 4.0)	(11.8, 4.2)		(10.5, 4.5)
glc′′′	1				5.04 *d* (7.6)
	2				4.00[c]
	3				4.23[c]
	4				4.14[c]
	5				3.94[c]
	6a				4.50 *d* (11.0)
	6b				4.36 *dd* (11.0,4.9)

[a] Recorded in pyridine-d_5. Assignments were established by COSY, TOCSY and HMBC spectra.
[b] *J* values (in Hz) in parentheses.
[c] Overlapping signals.

ion $[M + Na]^+$ at m/z 657, consistent with a molecular formula of $C_{36}H_{58}O_9$. Acid hydrolysis of **1** afforded the known longispinogenin, which was identified by comparison with literature values (Yoshikawa, Taninaka, Kan & Arihara, 1994), and glucuronic acid, which was confirmed by high performance thin layer chromatography (HPTLC). A comparison of the ^{13}C-NMR spectral data of **1** (Table 3) with those of the sapogenin longispinogenin (Yoshikawa et al., 1994) indicated a glycosylation shift at the C-3 position (+10.7 ppm), leading to the conclusion that the glucuronic acid residue was attached to the C-3 position of **1**.

The FABMS of **2** displayed a $[M + Na]^+$ ion peak at m/z 777, consistent with a molecular formula of $C_{43}H_{62}O_{11}$. The ^{1}H- and ^{13}C-NMR spectra suggested the presence of a sugar residue and a benzoyloxy group, in addition to the triterpene skeleton possessing an olefinic bond, a hydroxymethyl, three oxymethine and seven methyl groups. Acid hydrolysis of **2** afforded the sapogenin sitakisogenin, which was identified by comparison of its spectral data with literature values (Yoshikawa et al., 1994). Glucuronic acid and benzoic acid were also obtained from the hydrolysate; both were identified by HPTLC comparison with reference compounds. On alkaline treatment, **2** produced a prosapogenin, which could be further hydrolyzed by 10% HCl to afforded sitakisogenin. The locations of the glucuronic acid and the benzoyloxy group on the triterpene skeleton were then deduced from the results of 2D-NMR spectroscopic experiments. Thus, in the HMBC spectrum of **2**, correlation signals were observed between H-1 (δ 5.10) of glucuronic acid and C-3 (δ 89.0) of the aglycone and between H-21 (δ 5.70) of the aglycone and the carboxylic carbon (C-7′, δ 166.3) of the benzoyloxy group. This evidence indicated that the glucuronic acid and benzoyloxy groups were attached to the C-3 and C-21 positions of the aglycone, respectively. Hence, **2** was determined to be a new saponin with the structure 21β-benzoyl-sitakisogenin 3-*O*-β-D-glucuronopyranoside.

The FABMS of **3** showed a $[M+H]^+$ peak at m/z 943, consistent with a molecular formula of $C_{48}H_{78}O_{18}$. Acid hydrolysis of **3** afforded oleanolic acid (Takemoto et al., 1984) and glucose. The ^{1}H- and ^{13}C-NMR spectra of **3** (Tables 1–3) showed the presence of three sugar residues, clearly indicated by three anomeric carbon signals at δ 106.9, 105.4 and 95.7, and three anomeric proton signals at δ 4.86 (*d*, J = 7.8 Hz), 5.11 (*d*, J = 7.3 Hz) and 6.31 (*d*, J = 8.30 Hz). Analysis of the ^{13}C-NMR data of the saccharide portion (Table 1) revealed that one of the three CH_2OH groups of the glucose residues experienced a significant downfield shift (δ 70.4), suggesting that one of the glucose residues was glycosylated at the C-6 position. Furthermore, the HMBC spectrum of **3** exhibited a cross-peak between H-6 (δ 4.37 and 4.90) of glucose and C-1′ (δ 105.4) of another glucose residue, as well as a cross-peak between H-1′ (δ 6.31) of glucose and C-28 (δ 176.4) of the aglycone. Alkaline hydrolysis of **3** yielded a prosapogenin and glucose. The prosapogenin was identified as oleanolic acid 3-*O*-β-D-glucopyranosyl (1 → 6)-β-D-glucopyranoside (chikusetsusaponin II) by comparison of the NMR data with those reported in the literature (Lin & Shoji, 1979). These findings led to the assignment of **3** as 3-*O*-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl oleanolic acid 28-*O*-β-D-glucopyranosyl ester.

The FABMS of **4** displayed a quasi-molecular ion peak $[M + Na]^+$ at m/z 935, consistent with a mol-

Table 3
^{13}C-NMR spectral data of the aglycone moieties of compounds **1**–**6**[a]

Carbon	1	2	3	4	5	6
1	38.8	38.8	38.8	38.7	38.7	38.7
2	26.6	26.6	26.6	26.7	26.7	26.7
3	**89.0**	**89.0**	**88.9**	**89.0**	**89.0**	**89.0**
4	39.5	39.6	39.4	39.5	39.5	39.5
5	55.7	55.7	55.7	55.8	55.8	55.8
6	18.4	18.4	18.4	18.3	18.5	18.5
7	32.9	33.0	33.0	33.1	33.1	33.1
8	40.1	40.1	39.8	39.9	39.9	39.9
9	47.1	47.1	47.9	48.0	48.0	48.0
10	36.7	36.7	36.9	37.0	37.0	37.0
11	23.8	23.9	23.7	23.7	23.8	23.7
12	122.6	123.1	122.9	122.8	123.0	122.9
13	143.9	142.6	144.0	144.4	144.0	144.1
14	43.8	43.7	42.0	42.1	42.1	42.1
15	36.7	36.8	28.2	28.2	28.2	28.2
16	66.6	66.4	23.3	23.4	23.4	23.4
17	41.1	43.8	46.9	46.5	47.0	47.0
18	44.4	44.2	41.6	41.9	41.7	41.7
19	47.1	47.2	46.2	46.1	46.2	46.3
20	31.1	36.0	30.7	30.9	30.8	30.8
21	34.3	75.6	33.9	34.4	34.0	34.0
22	26.2	33.3	32.5	33.1	32.5	32.5
23	28.2	28.2	28.8	28.2	28.2	28.3
24	16.9	16.9	17.0	17.0	17.0	17.0
25	15.7	15.7	15.5	15.8	15.6	15.6
26	17.0	17.0	17.4	17.3	17.5	17.5
27	27.2	27.0	26.0	26.1	26.4	26.4
28	68.9	66.8	**176.4**	180.2	**176.5**	**176.5**
29	33.4	29.2	33.1	33.2	33.2	33.2
30	24.1	18.8	33.6	23.7	23.7	23.7
Acyl part						
1		131.6				
2		129.9				
3		128.9				
4		133.2				
5		128.9				
6		129.9				
7		166.3				

[a] Spectra were measured in pyridine-d_5. Assignments were established by interpretation of ^{13}CDEPT, HMQC and HMBC spectra. Values given in boldface indicate the glycosidic positions of the aglycone.

ecular formula of $C_{47}H_{76}O_{17}$. The ^{13}C and DEPT NMR spectra (Tables 1–3) displayed 47 signals, of which 17 could be readily assigned to the saccharide portion and the remaining 30 to a triterpene skeleton. Acid hydrolysis of **4** afforded oleanolic acid (Takemoto et al., 1984), glucose, and xylose. Further comparison of the ^{13}C-NMR data of **4** with those of oleanolic acid indicated a glycosylation shift at the C-3 position, leading to the conclusion that **4** was a monodesmosidic glycoside with a sugar chain attached to the C-3 position of oleanolic acid. The oligosaccharide structure was subsequently determined by 2D-NMR studies. Thus, in the HMBC spectrum of **4**, correlation peaks were observed between H-1 (δ 4.86) of the inner glucose and C-3 (δ 89.0) of the aglycone, between H-1′ (δ 5.05) of the central glucose and C-6 (δ 70.4) of the inner glucose, as well as between H-1 (δ 4.98) of xylose and C-6′ (δ 69.8) of the central glucose residue. Hence, the structure of **4** was established to be oleanolic acid 3-*O*-β-D-xylopyranosyl(1 → 6)-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranoside.

The FABMS of **5** showed a $[M + Na]^+$ ion peak at *m/z* 1097, consistent with a molecular formula of $C_{53}H_{86}O_{22}$. Acid hydrolysis of **5** afforded oleanolic acid (Takemoto et al., 1984), glucose, and xylose. Alkaline hydrolysis afforded **4**, which was confirmed by a direct comparison of TLC and NMR data. Glucose was detected in the hydrolysate. Analysis of the NMR data of **5** (Tables 1–3), and a comparison with those of **4**, showed that the former compound possessed an additional glucopyranosyl residue, leading to the assignment of a glucose ester at the C-28 position of **4**, which was supported by the results of HMBC and ROESY experiments (see Fig. 1). The HMBC spectrum of 5 clearly showed a cross-peak between H-1″ (δ 6.33) of glucose and C-28 (δ 176.5) of the aglycone. Other correlation peaks were observed between H-1 (δ 4.87) of the inner glucose and C-3 (δ 89.0) of the aglycone, between H-1′ (δ 5.05) of the central glucose and C-6 (δ 70.4) of the inner glucose, and between H-1 (δ 4.96) of xylose and C-6′ (δ 69.8) of the central glucose. These data led to the assignment of **5** as 3-*O*-β-D-xylopyranosyl(1 → 6)-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl oleanolic acid 28-*O*-β-D-glucopyranosyl ester.

The FABMS of **6** displayed a quasi-molecular ion peak $[M + Na]^+$ at *m/z* 1127, consistent with a molecular formula of $C_{54}H_{88}O_{23}$. On alkaline hydrolysis, **6** afforded glucose and a prosapogenin, identified as oleanolic acid 3-*O*-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranoside (chikusetsusaponin II) by comparison of its NMR data with those reported in the literature (Lin & Shoji, 1979). The ^1H- and ^{13}C-NMR spectra (Tables 1–3) of **6** revealed the presence of four sugar residues. Downfield shifts of the carbon signals for C-3 (δ 89.0) and C-28 (δ 176.5) indicated that both positions were glycosylated. A comparison of the NMR spectral data between **3** and **6** suggested that the latter contained one additional glucose residue on the sugar moiety located at the C-28 position. The results of HMBC and ROESY experiments (see Fig. 2) also supported this assumption. Thus, the HMBC spectrum of **6** displayed correlation signals between H-1 (δ 4.88) of glucose and C-3 (δ 89.0) of the aglycone, as well as between H-1′ (δ 5.13) of glucose and C-6 (δ 70.5) of another glucose. Moreover, correlations were also demonstrated between H-1″ (δ 6.26) of glucose and C-28 (δ 176.5) of the aglycone, and between H-1‴ (δ 5.04) of glucose and C-6′ (δ 69.3) of another glucose. Further information was derived from the results of a ROESY experiment, in which correlation peaks between H-1′ (δ 5.13) of glucose and H-6 (δ 4.90 and 4.36) of another glucose, as well as between H-1‴ (δ 5.04) of glucose and H-6″ (δ 4.72 and 4.38) of another

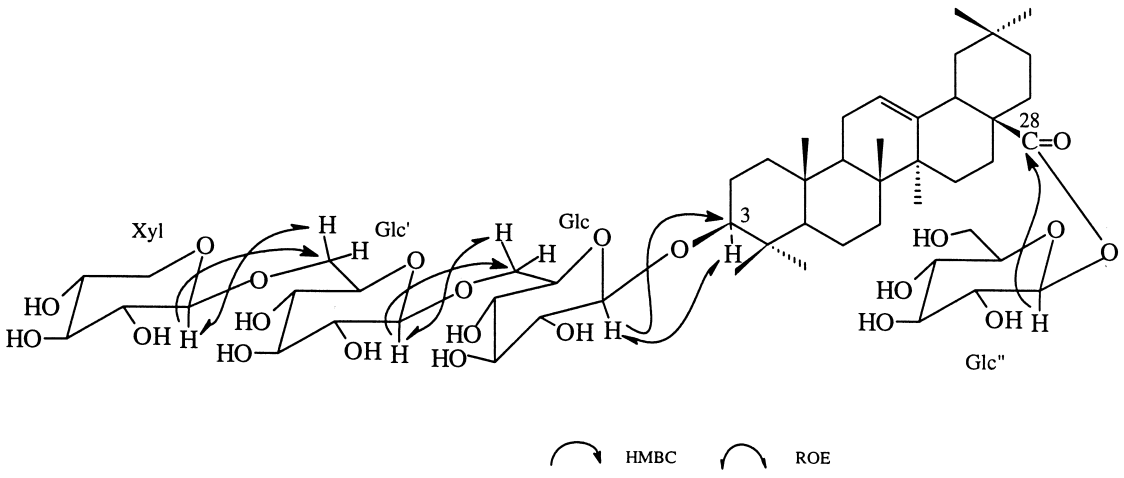
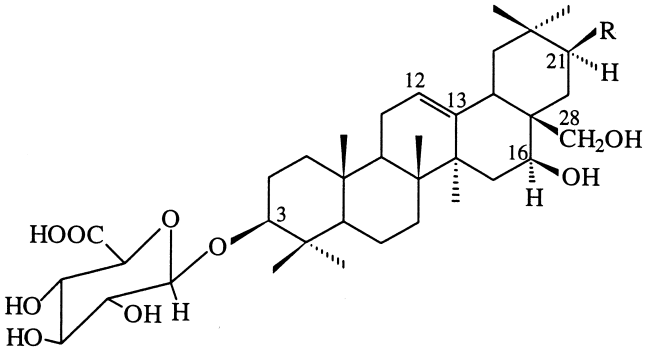


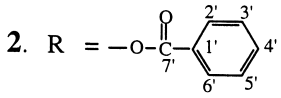
Fig. 1. Key correlations of HMBC and ROE of **5**.

glucose were clearly observed. All available evidence led to the conclusion that **6** was a new saponin with a structure of 3-*O*-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl oleanolic acid 28-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranosyl ester.

It is noteworthy that the sapogenins previously reported from *G. sylvestre* of Indian origin belong to oleanane- and dammarane-type (such as gymnemagenin, gymnestrogenin and 23-hydroxylongispinogenin) (Liu, Kiuchi & Tsuda, 1992; Yoshikawa, Murakami & Matsuda, 1997). In our hands oleanolic acid was obtained as the major sapogenin from *G. sylvestre* leaves of Guangxi (China) origin.



1. R = H



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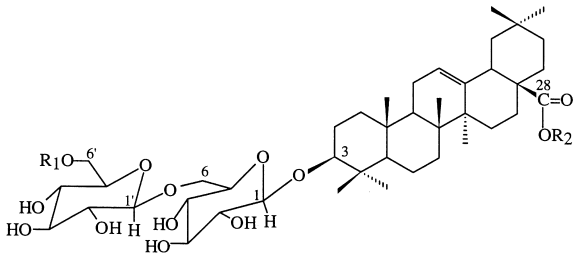
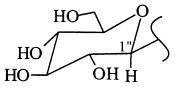
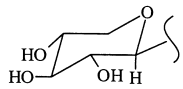
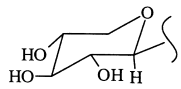
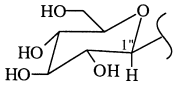
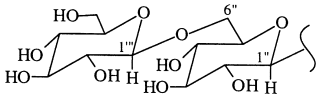


	R_1	R_2
3.	H	
4.		H
5.		
6.	H	

3. Experimental

3.1. General

The IR spectra were recorded on a Perkin–Elmer 16 PC FT-IR spectrometer. Optical rotations were

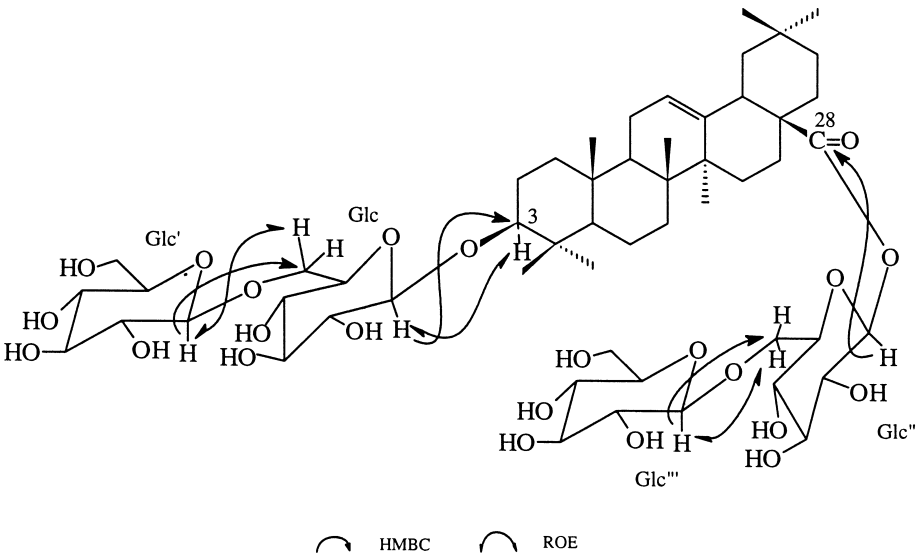


Fig. 2. Key correlations of HMBC and ROE of **6**.

measured on a Perkin–Elmer 241 polarimeter. ^{1}H (400 or 500 MHz), ^{13}C (100 or 125 MHz), and 2D-NMR spectra were recorded on JEOL JNM-EX 400 or Varian Unity INOVA-500 spectrometers. FABMS spectra were determined in the positive ion mode on a Finnigan MAT TSQ7000 spectrometer. Column chromatography was carried out on silica gel (200–400 mesh), ODS gel (10–40 μm) and Sephadex LH-20. TLC was conducted on silica gel 60 F_{254} and RP-18 F_{254} S plates (Merck).

3.2. Plant materials

The leaves of *G. sylvestre* R. Br. were collected in October in the Guangxi Autonomous Region of the China, and authenticated by Dr. Qin Min-Jang. A voucher specimen has been deposited in the Herbarium of the China Pharmaceutical University.

3.3. Extraction and isolation

Dried leaves of *G. sylvestre* (3.0 kg) were extracted with EtOH. The extract was concentrated, defatted with cyclohexane, and partitioned with *n*-BuOH. The *n*-BuOH layer was dried under reduced pressure and repeatedly separated by silica gel column chromatography using $CHCl_3$–CH_3OH as eluant to yield compounds **1** (80 mg), **2** (60 mg), oleanolic acid 28-*O*-β-D-glucopyranosyl ester (180 mg), oleanolic acid 3-*O*-β-D-glucopyranosyl(1 → 6)-β-D-glucopyranoside (115 mg), and a mixture of glycosides. The mixture containing **3**–**6** was further purified on an ODS column using MeOH-–H_2O (30:70→70:30) as eluant to afford **3** (45 mg), **4** (30 mg), **5** (250 mg) and **6** (210 mg).

3.4. Acid hydrolysis of 1–6

A solution of each compound (8–15 mg in 5 ml 10% HCl in 50% EtOH) was refluxed for 6 h. The reaction mixture was diluted with H_2O and neutralized with Ag_2CO_3. The neutralized solution was extracted with $CHCl_3$ and purified on a Sephadex LH-20 column using MeOH as eluant to yield the aglycone. The aglycone was identified by comparison with an authentic sample or literature values. The neutral hydrolysate revealed the presence of glucose, xylose, glucoronic acid, and/or benzoic acid by HPTLC when compared with authentic samples.

3.5. Alkaline hydrolysis of 2, 3, 5 and 6

The saponin (10 mg) was dissolved into 5% NaOH in 50% EtOH (10 ml) and warmed at 50°C for 20 h. The reaction mixture was neutralized by HCl and extracted with *n*-BuOH. The *n*-BuOH layer was evaporated to give a residue, which was subjected to chromatographic separation on an ODS column eluted by MeOH–H_2O to afforded the prosapogenin. The prosapogenin was identified by NMR spectral data and/or by direct comparison with authentic samples. The water layer was analyzed by HPTLC (*n*-BuOH–HOAC–H_2O, 4:1:1) to reveal the presence of glucose or xylose.

3.6. Compound 1

Amorphous powder, mp 198–202°, $[\alpha]_{20}^{D}$ +16.0° (*c* 0.10, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3414 (OH), 1724 (COOH), 1636 (C=C), 1458, 1380, 1054. ^{1}H-NMR spectral data (500 MHz, pyridine-d_5): δ 0.86 (3H, *s*, Me), 0.95 (3H, *s*, Me), 1.01 (9H, *s*, 3 × Me), 1.32 (3H, *s*, Me), 1.39 (3H, *s*, Me), 3.39 (1H, *dd*, *J* = 4.3 and 11.8 Hz, H-3α), 3.68 (1H, *d*, *J* = 10.5 Hz, H-28a), 4.43 (1H, *d*, *J* = 10.5 Hz, H-28b), 4.68 (1H, *m*, H-16α), 5.04 (1H, *d*, *J* = 7.8 Hz, H-1 of glucuronic acid), 5.26 (1H, *br s*, H-12). ^{13}C-NMR spectral data (125 MHz, pyridine-d_5), see Tables 1 and 3. FABMS *m*/*z*: 657 [M + Na]$^+$.

3.7. Compound 2

Amorphous powder, mp 192–195°, $[\alpha]_{20}^{D}$ + 27.2° (*c* 0.15, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3444 (OH), 1724, 1700, 1635 (C=C), 1457, 1388, 1280, 1074, 720. ^{1}H-NMR spectral data (500 MHz, pyridine-d_5): δ 0.98 (3H, *s*, Me), 1.01(3H, *s*, Me), 1.02 (3H, *s*, Me), 1.07 (3H, *s*, Me), 1.30 (3H, *s*, Me), 1.34 (3H, *s*, Me), 1.36 (3H, *s*, Me), 3.40 (1H, *dd*, *J* = 4.5 and 12.0 Hz, H-3α), 3.70 (1H, *d*, *J* = 10.2 Hz, H-28a), 4.42 (1H, *d*, *J* = 10.2 Hz, H-28b), 4.70 (1H, *m*, H-16α), 5.10 (1H, *d*, *J* = 7.8 Hz, H-1 of glucuronic acid), 5.70 (1H, *dd*, *J* = 4.7 and 12.3 Hz, H-21α), 7.47 (3H, overlapped, H-3, -4 and -5), 8.25 (2H, *dd*, *J* = 1.4 and 8.4 Hz, H-2 and -6). ^{13}C-NMR spectral data (125 MHz, pyridine-d_5), see Tables 1 and 3. FABMS *m*/*z*: 777 [M + Na]$^+$.

3.8. Compound 3

Amorphous powder, mp 206–209°, $[\alpha]_{20}^{D}$−6.5° (*c* 0.11, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3424 (OH), 1735 (COOR), 1636 (C=C), 1457, 1034. ^{1}H-NMR spectral data (400 MHz, pyridine-d_5): δ 0.82 (3H, *s*, Me), 0.87 (3H, *s*, Me), 0.91 (3H, *s*, Me), 0.97 (3H, *s*, Me), 1.07 (3H, *s*, Me), 1.20 (3H, *s*, Me), 1.23 (3H, *s*, Me), 3.17 (1H, *dd*, *J* = 3.5 and 10.2 Hz, H-18), 3.30 (1H, *dd*, *J* = 3.9 and 11.7 Hz, H-3α), 5.37 (1H, *br s*, H-12). ^{1}H-NMR spectral data of the saccharide residues, see Table 2. ^{13}C-NMR spectral data (100 MHz, pyridine-d_5), see Tables 1 and 3. FABMS *m*/*z*: 943 [M + H]$^+$.

3.9. Compound **4**

Amorphous powder, mp 202–204°, $[\alpha]_{20}^{D}$ −3.2° (*c* 0.15, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3410 (OH), 1710 (COOH), 1638 (C=C), 1458, 1036. ^{1}H-NMR spectral data (400 MHz, pyridine-d_5): δ 0.87 (3H, *s*, Me), 0.91 (3H, *s*, Me), 0.96 (3H, *s*, Me), 1.02 (3H, *s*, Me), 1.10 (3H, *s*, Me), 1.24 (3H, *s*, Me), 1.29 (3H, *s*, Me), 3.30 (1H, *dd*, J = 4.5 and 11.5 Hz, H-3α), 5.38 (1H, *br s*, H-12). ^{1}H -NMR spectral data of the saccharide residues, see Table 2. ^{13}C-NMR spectral data (100 MHz, pyridine-d_5), see Tables 1 and 3. FABMS m/z: 935 $[M + Na]^+$.

3.10. Compound **5**

Amorphous powder, mp 212–215°, $[\alpha]_{20}^{D}$ −9.6° (*c* 0.20, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3414 (OH), 1740 (COOR), 1636 (C=C), 1460, 1364, 1044, 896. ^{1}H-NMR spectral data (500 MHz, pyridine-d_5): δ 0.85 (3H, *s*, Me), 0.90 (3H, *s*, Me), 0.94 (3H, *s*, Me), 1.00 (3H, *s*, Me), 1.09 (3H, *s*, Me), 1.23 (3H, *s*, Me), 1.27 (3H, *s*, Me), 3.19 (1H, *dd*, J = 4.0 and 13.7 Hz, H-18), 3.32 (1H, *dd*, J = 4.4 and 11.7 Hz, H-3α), 5.40 (1H, *br s*, H-12). ^{1}H-NMR spectral data of the saccharide residues, see Table 2. ^{13}C-NMR spectral data (125 MHz, pyridine-d_5), see Tables 1 and 3. FABMS m/z: 1097 $[M + Na]^+$.

3.11. Compound **6**

Amorphous powder, mp 209–211°, $[\alpha]_{20}^{D}$ −12.1° (*c* 0.12, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3424 (OH), 1734 (COOR), 1636 (C=C), 1458, 1074. ^{1}H-NMR spectral data (500 MHz, pyridine-d_5): δ 0.87 (3H, *s*, Me), 0.90 (3H, *s*, Me), 0.92 (3H, *s*, Me), 1.00 (3H, *s*, Me), 1.09 (3H, *s*, Me), 1.22 (3H, *s*, Me), 1.26 (3H, *s*, Me), 3.20 (1H, *dd*, J = 3.5 and 13.6 Hz, H-18), 3.33 (1H, *dd*, J = 4.4 and 11.5 Hz, H-3α), 5.39 (1H, *br s*, H-12). ^{1}H-NMR data of the saccharide residues, see Table 2. ^{13}C-NMR spectral data (125 MHz, pyridine-d_5), see Tables 1 and 3. FABMS m/z: 1127 $[M + Na]^+$.

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