A New Biologically Active Acylated Triterpene Saponin from *Silene fortunei*

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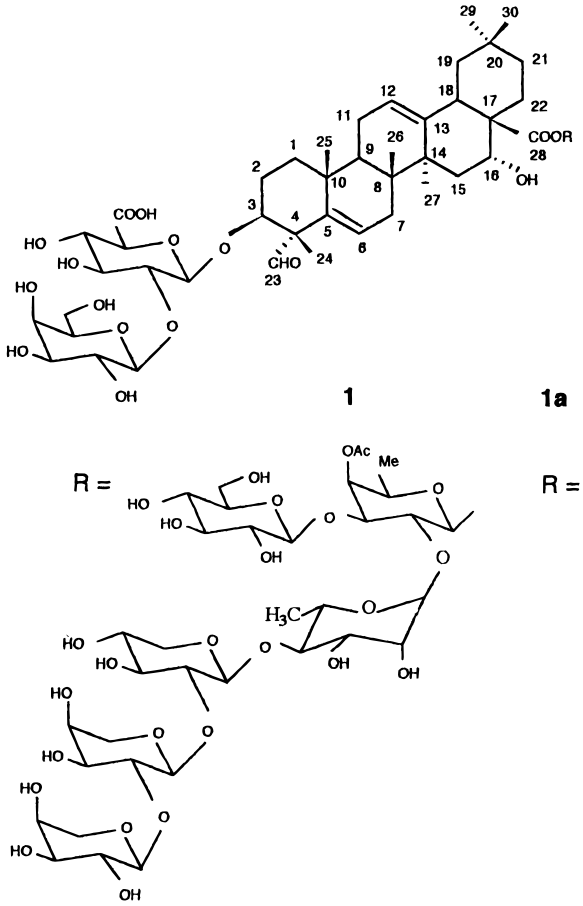
A new acylated triterpene−saponin (**1**), together with a mixture of the known jenisseensosides C and D, has been isolated from the roots of *Silene fortunei*. The structure of the new compound was established by chemical means and spectroscopic methods as 3-*O*-[*β*-D-galactopyranosyl-(1→2)-*β*-D-glucuronopyranosyl]-28-*O*-[[α-L-arabinopyranosyl-(1→2)-α-L-arabinopyranosyl-(1→3)-*β*-D-xylopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→2)]-[*β*-D-glucopyranosyl-(1→3)]-4-*O*-acetyl-*β*-D-fucopyranosyl]quillaic acid. This saponin showed a significant enhancement of granulocyte phagocytosis *in vitro*.

The root of *Silene fortunei* Wis[1] is well-known in traditional medicine in Hubei Province of the People's Republic of China as a substitute for the drug "Yin-Chai-Hu" (roots of *Stellaria dichotoma* var. *lanceolata* Bge.) and is utilized to treat various types of fever.[2−4] Furthermore, the root has been reported to treat contusions, sprains, and joint and muscle pain.[3] The whole plant has been reported to treat dysentery and urinary tract infections.[3] There are no reports on either phytochemical or pharmacological work on this plant.

In the course of our continuing search for new biologically active saponins from Caryophyllaceae,[5−7] we report herein the isolation and structure elucidation of a new acylated triterpene octaglycoside (**1**) together with the known jenisseensosides C and D from an ethanol extract of *S. fortunei* roots. The immunological properties of **1** have been investigated.

The ethanol extract of the dried roots of the plant was separately subjected to partitions between water and petroleum ether, water and ethyl acetate, and water and *n*-butanol. The *n*-butanol-soluble portion was subjected to silica gel column chromatography to afford a saponin fraction that was further separated by repeated medium-pressure liquid chromatography over reversed-phase and normal silica gel yielding the pure compound **1**, together with a mixture of the known jenisseensosides C and D.[7] The structure of **1** was elucidated mainly by 500 MHz NMR analysis including 1D and 2D NMR (^{1}H−^{1}H COSY, DQF-COSY, HMQC, HMBC) spectroscopy.

Compound **1** was obtained as a white amorphous powder. Its FABMS (negative-ion mode) exhibited a quasimolecular ion peak at *m*/*z* 1715 [M − H]$^-$, indicating a molecular weight of 1716, compatible with the molecular formula $C_{77}H_{120}O_{42}$. Other fragment ion peaks at *m*/*z* 1451 [(M − H) − 2 × 132]$^-$, 1131 [(M − H) − 3 × 132 − 146 − 42]$^-$, and 823 [(M − H) − 160 − 146 − 146 − 42 − 3 × 132]$^-$ indicated the respective loss of two pentosyl moieties and then the loss of one pentosyl, one desoxyhexosyl, one acetyl group, and finally the elimination of one desoxyhexoxyl and one hexosyl moiety. Another fragment ion peak at *m*/*z* 485 corresponded to the pseudomolecular ion of the aglycon.



Acid hydrolysis of **1** with 2 N TFA at 100 °C afforded galactose, glucose, fucose, rhamnose, xylose, and arabinose (in a molar ratio of 1:1:1:1:1:2 as estimated by GLC analysis after conversion into their alditol acetates), glucuronic acid (co-TLC with an authentic sample), and an aglycon, which was identified as quillaic acid, from the ^{1}H NMR, ^{13}C NMR, DEPT, ^{1}H−^{1}H COSY, HMQC, and HMBC spectra of **1**. Most of the signals were assigned through $^2J_{H-C}$ and $^3J_{H-C}$ couplings of the seven methyls and were in good agreement with literature data.[7]

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The alkaline hydrolysis of **1** with 5% KOH (1 h at 100 °C) gave the prosapogenin **1a.** The FABMS (negative-ion mode) (*m/z* 823 $[M - H]^-$), ^{1}H NMR, and ^{13}C NMR data of **1a** were in good agreement with those of the prosapogenin of the jenisseensosides, which has been previously identified as 3-*O*-[*β*-D-galactopyranosyl-(1→2)-*β*-D-glucuronopyranosyl]quillaic acid.[6]

The above data indicated that **1** must be an acylated bidesmosidic saponin in which two sugars are bound by a glycosidic linkage to the aglycon at C-3 while the six remaining sugars must be bound to the genin by a glycosidic ester linkage at C-28.

We could confirm that saponin **1** contains eight sugar residues as shown by HMQC experiments, which revealed eight correlations between carbons from anomeric carbon signals in the *δ* 105−93 range and anomeric proton signals resonating between *δ* 4.0 and 5.5. The anomeric ^{13}C NMR signals at *δ* 104.7, 104.5, 104.3, 103.2, 101.4, 100.5, 100.2, and 93.1 give correlations with anomeric protons at *δ* 4.24 (d, $J = 7.3$ Hz), 4.47 (d, $J = 7.5$ Hz), 4.34 (d, $J = 7.3$ Hz), 4.34 (d, $J = 7.4$ Hz), 4.11 (d, $J = 7.5$ Hz), 4.78 (br s), 5.11 (s), and 5.38 (d, $J = 7.6$ Hz), respectively. Evaluation of spin−spin couplings and chemical shifts allowed the identification one *β*-galactopyranose (Gal), one *β*-xylopyranose (Xyl), one α-arabinopyranose (Ara), one *β*-glucopyranose (Glc), one *β*-glucuronopyranose (Glc-A), one α-arabinopyranose (Ara), one α-rhamnopyranose (Rha), and one *β*-fucopyranose (Fuc) unit, respectively. The common D configuration for Glc, Gal, Glc-A, Xyl, and Fuc and the L configuration for Rha and Ara were assumed according to those most often encountered among the plant glycosides. In view of the *J* values of the two arabinopyranosyl units (δ_{H-1} 4.34, d, $J = 7.3$ Hz, δ_{H-1} 4.78, br s), it could be noted that one arabinose ring predominantly adopts the $^4C_1$ conformation ($J = 7-8$ Hz), whereas the second arabinose ring predominantly adopts the $^1C_4$ conformation ($J = 2-3$ Hz).[8] The high conformational mobility of arabinopyranosides between $^4C_1$ and $^1C_4$ is well known.[8] After subtraction of the anomeric signals of the sugars linked at the C-3 position from the total HMQC spectrum, the signals of six sugars linked to the aglycon by an ester linkage remained. Furthermore, a correlation in the HMQC spectrum between a ^{13}C NMR signal at δ_C 20.8 and a ^{1}H NMR resonance at δ_H 2.1(s) and a correlation in the HMBC spectrum between signals at δ_H 2.1 and δ_C 170.5 confirmed the presence of one acetyl group in the molecule. The sequence of the sugars and the acetyl position were obtained by analysis of ^{1}H−^{1}H COSY, HMBC, and HMQC experiments. A correlation in the HMQC spectrum at δ_C/δ_H 93.1/5.38 (d, $J = 8.0$ Hz) showed that the fucose residue was attached to the carboxylic group of the aglycon by an ester linkage. This conclusion was confirmed by the HMBC experiments, which showed a correlation between signals at $\delta_{H(Fuc-1)}$ 5.38 and $\delta_{C(Agly-C28)}$ 175.1, confirming that the Fuc was bound to the carboxyl unit of the aglycon. Another correlation in the HMQC spectrum between the ^{13}C NMR signal at δ_C 72.4 and the deshielded ^{1}H NMR resonance at δ_H 5.12 (d, $J = 3.0$ Hz) corresponded to one secondary alcoholic function after acetylation. Accordingly, from the cross-peak in the ^{1}H−^{1}H COSY spectrum of **1**, it was easy to assign the proton chemical shifts in fucose as δ_{H-1} 5.38 (d, $J = 8$ Hz), δ_{H-2} 3.68, δ_{H-3} 4.05, δ_{H-4} 5.12 (d, $J = 3.0$ Hz), δ_{H-5} 3.82, and δ_{H-6} 0.95 (d, $J = 6.3$ Hz). Furthermore, the long-range correlations in the HMBC spectrum between the deshielded ^{1}H NMR signal of Fuc at $\delta_{H(Fuc-4)}$ 5.12 and a ^{13}C NMR signal of a carboxyl group at δ_C 170.5, between the deshielded ^{1}H NMR signal at $\delta_{H(Fuc-4)}$ 5.12 and the ^{13}C NMR signals at $\delta_{C(Fuc-2)}$ 73.2 (^3J), $\delta_{C(Fuc-3)}$ 79.8, and $\delta_{C(Fuc-5)}$ 68.8, and between the ^{1}H NMR signals at $\delta_{H(Fuc-6)}$ 0.95 and the ^{13}C NMR signals at $\delta_{C(Fuc-5)}$ 68.8 (^2J), and $\delta_{C(Fuc-4)}$ 72.4 (^3J) thereby confirmed that the acetyl group was located at Fuc-C-4 OH. Other correlations between the ^{1}H NMR signal at $\delta_{H(Rha-1)}$ 5.11 (s) and the ^{13}C NMR signals at $\delta_{C(Fuc-2)}$ 73.2 indicated that the rhamnose was linked to the fucose by a 1→2 linkage. This attachment was confirmed by observation of a reverse correlation between the ^{1}H NMR resonance at $\delta_{H(Fuc-2)}$) 3.68 and the ^{13}C NMR signal at $\delta_{C(Rha-1)}$ 100.5. Assignments of the ^{1}H NMR and ^{13}C NMR signals from HMQC and HMBC spectra showed that the four remaining sugars were a terminal Glc (T-Glc), a terminal Ara (T-Ara), a disubstituted Xyl-1,3, and a disubstituted Ara-1,2. All the carbon signals due to these sugar moieties were in good agreement with the published data for similarly linked sugar moieties.[9−11] The longe-range correlation in the HMBC spectrum between the anomeric ^{1}H NMR signal of Glc at $\delta_{H(Glc-1)}$ 4.34 and the ^{13}C NMR signals at $\delta_{C(Fuc-3)}$ 79.8 and a reverse correlation between the proton resonance at $\delta_{H(Fuc-3)}$ 4.05 and the carbon signal at $\delta_{C(Glc-1)}$ 103.2 proved the T-Glc to be attached at Fuc-C3 OH. For the sequencing of the three remaining pentosyl moieties (T-Ara, Xyl-1,3 and Ara-1,2), the observation of cross-peaks in the HMBC spectrum between the anomeric proton signals and carbon signals in adjacent systems were all present together with reverse correlations between proton-ring signals and anomeric carbon signals: $\delta_{H(Xyl-1)}$ 4.47 → $\delta_{C(Rha-4)}$ 81.6, $\delta_{H(inner\ Ara-1)}$ 4.78 → $\delta_{C(Xyl-3)}$ 83.7, $\delta_{H(T-Ara-1)}$ 4.34 → $\delta_{C(inner\ Ara-2)}$ 78.7), ($\delta_{H(Rha-4)}$ 3.4 → $\delta_{C(Xyl-1)}$ 104.5, $\delta_{H(Xyl-3)}$ 3,38 → $\delta_{C(inner\ Ara-1)}$ 100.5, $\delta_{H(inner\ Ara-2)}$ 3.68 → $\delta_{C(T\ Ara-1)}$ 104.3. Therefore, the sequence of this trisaccharide and its attachment position at the rhamnoyl unit were formulated as α-L-arabinopyranosyl-(1→2)-α-L-arabinopyranosyl-(1→3)-*β*-D-xylopyranosyl-(1→4)-α-L-rhamnopyranosyl.

Table 1. ^{13}C NMR Data of the Aglycons of Saponins **1** and of Compound **1a** (*δ* ppm, DMSO-d_6 as Solvent)[a]

position	DEPT	**1**	**1a**
1	CH_2	37.7	37.7
2	CH_2	24.0	24.1
3	CH	81.1	82.1
4	C	53.9	54.0
5	CH	45.9	46.0
6	CH_2	19.8	19.8
7	CH_2	31.8	31.8
8	C	38.9	38.9
9	CH	47.2	47.4
10	C	35.4	35.5
11	CH_2	22.9	22.9
12	CH	121.3	121.2
13	C	143.1	143.3
14	C	40.6	40.6
15	CH_2	34.6	34.8
16	CH	72.5	72.5
17	C	48.1	48.0
18	CH	41.1	41.1
19	CH_2	46.5	46.5
20	C	30.1	30.1
21	CH_2	34.9	34.9
22	CH_2	30.6	30.6
23	CHO	209.3	209.0
24	Me	10.1	10.3
25	Me	15.5	15.5
26	Me	16.8	16.8
27	Me	26.3	26.3
28	C	175.1	175.1
29	Me	32.8	32.8
30	Me	24.3	24.3

[a] Chemical shifts are referenced to dimethyl sulfoxide at *δ* 39.5 ppm. Multiplicities were assigned from DEPT spectra.

Table 2. ^{13}C and ^{1}H NMR Spectral Data of Sugar Moieties for Compounds **1** and **1a** (in DMSO-d_6, ppm)[a]

position		**1**, δ_C	**1**, δ_H	**1a**, δ_C	**1a**, δ_H	position		**1** δ_C	**1** δ_H
3-*O*-						28-*O*-			
Glc-A	1	101.4	4.11 (d, 7.5)	101.4	4.12,(d, 7.5)	Fuc	1	93.1	5.38 (d, 8.0)
	2	81.5	3.15 (m)	81.5	3.18 (m)		2	73.2	3.68 (m)
	3	75.1	3.35 (m)	75.2	3.40 (m)		3	79.8	4.05 (m)
	4	71.8	3.09 (m)	72.3	3.10 (m)		4	72.4	5.12 (d, 3.0)
	5	76.5	3.29 (m)	76.4	3.31 (m)		5	68.8	3.82 (m)
	6	171.9		172.2			6	16.1	0.95 (d, 6.3)
						Ac	CH_3	20.8	2.1 (s)
							COO	170.5	
Gal	1	104.7	4.24 (d, 7.3)	104.9	4.25 (d, 7.5)	Rha	1	100.2	5.11 (s)
	2	72.8	3.30 (m)	72.3	3.31 (m)		2	69.8	3.80 (m)
	3	73.1	3.15 (m)	72.9	3.18 (m)		3	70.1	3.64 (m)
	4	67.8	3.65 (m)	67.7	3.67 (m)		4	81.6	3.42 (m)
	5	73.8	3.37 (m)	74.0	3.40 (m)		5	67.2	3.58 (m)
	6	59.8	3.45 (m)	59.8	3.40 (m)		6	17.9	1.17 (d, 6.0)
			3.58 (m)		3.60 (m)				
						Glc	1	103.2	4.34 (d, 7.4)
							2	73.6	2.95 (m)
							3	76.7	3.12 (m)
							4	69.8	3.05 (m)
							5	76.9	3.07(m)
							6	61.2	3.65 (m)
									3.4 (m)
						Xyl	1	104.5	4.47 (d,7.5)
							2	73.8	3.20 (m)
							3	83.7	3.38 (m)
							4	67.6	3.35 (m)
							5	65.5	3.12 (m)
									3.85 (m)
						Ara	1	100.5	4.78 (br. s)
							2	78.7	3.66 (m)
							3	70.7	3.68 (m)
							4	65.7	3.78 (m)
							5	63.1	3.40 (m)
									3.82 (m)
						Ara	1	104.3	4.34, (d, 7.3)
							2	71.1	3.39 (m)
							3	72.3	3.40 (m)
							4	67.3	3.60 (m)
							5	65.5	3.44 (m)
									3.75 (m)

[a] Measured at 500 MHz for ^{1}H and 125 MHz for ^{13}C with reference to δ 39.5 in DMSO-d_6. Assignments were made on the basis of $^{1}H-^{1}H$ DQFCOSY, HMQC, HMBC, and DEPT experiments. β-D-Galactopyranose (Gal), β-D-xyloppyranose (Xyl), β-D-glucopyranose (Glc), β-D-glucuronopyranose (Glc-A), α-L-arabinopyranose (Ara), α-L-rhamnopyranose (Rha), and β-D-fucopyranose (Fuc).

On the basis of the above results and the assumption that Fuc, Gal, Glc, Glc-A, and Xyl are members of the commonly found D series and Ara, Rha of the L series, the structure of the saponin **1** was represented as 3-*O*-[β-D-galactopyranosyl-(1→2)-β-D-glucuronopyranosyl]-28-*O*-[[α-L-arabinopyranosyl-(1→2)-α-L-arabinopyranosyl-(1→3)-β-D-xylopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→2)]-[β-D-glucopyranosyl-(1→3)]-4-*O*-acetyl-β-D-fucopyranosyl]quillaic acid. According to previous reports on triterpene saponins,[12−14] **1** is a new natural compound.

Also obtained as an inseparable mixture were jenisseensosides C and D (3-*O*-[β-D-galactopyranosyl-(1→2)-β-D-glucuronopyranosyl]-28-*O*-[[α-L-rhamnopyranosyl-(1→2)]-[4-*O*-*trans*-*p*-methoxycinnamoyl]-β-D-fucopyranosyl]-quillaic acid and its cis isomer). All the spectral data were in full agreement with those of the previously isolated compounds from *Silene jenisseensis*.[7]

Since triterpene saponins from plants in the Caryophyllaceae have been reported to exert immunostimulating activities,[12,13] the saponin **1** was tested in an *in vitro* granulocyte phagocytosis assay according to a new flow cytometric technique[15−17] and in a T-cell activation assay.[18] In the *in vitro* phagocytosis assay, **1** did not show any cytotoxicity for the granulocytes up to 100 μg/mL. At a concentration range of 10−100 μg/mL, **1**, which should not contain lipopolysaccharides according to the purification procedure, showed a significant enhancement of granulocyte phagocytosis (24−51%). The results obtained with **1** and with the mixture of jenisseensosides C and D[7] confirmed the applicability of this method for measuring the phagocytosis-enhancing potential of immunostimulating saponins from plants. In the T-cell activation assay, **1** did not show any significant activating effect of the lymphocytes at the tested concentrations (100 pg/mL−10 μg/mL).

Experimental Section

General Experimental Procedures. The NMR spectra were obtained with a Bruker DRX 500 spectrometer (500 MHz for ^{1}H and 2D $^{1}H-^{1}H$ COSY spectra and 125 MHz for ^{13}C spectra). The carbon type (Me, CH_2, methine) was determined by DEPT experiments. The ^{1}H-detected one-bond and multiple-bond ^{13}C multiple-quantum coherence spectra (HMQC and HMBC, respectively) were measured at 500 MHz with a DRX 500 spectrometer that was equipped to allow inverse detection. The magnitude of the delay for optimizing one-bond correlations in the HMQC spectrum and suppressing them in the HMBC spectrum was 3.45 ms, and the evolution delay for long-range couplings in the latter was set to 60 ms. All 1D and 2D spectra were recorded using standard software, and data manipulation of the 2D spectra was performed on a Silicon Graphics Indigo computer. All chemical shifts (δ) are given in ppm and the samples were solubilized in DMSO-d_6. Fast-atom bombardment (FABMS) (negative-ion mode, thioglycerol

matrix) was conducted on a JEOL DX 300 with JMA-3500 system. The target was bombarded with 6 keV Xe atoms. Optical rotations were taken with a Perkin-Elmer 241 polarimeter. IR spectra (KBr disk) were recorded on a Perkin-Elmer 281 spectrophotometer. UV spectra were obtained on one line by an HP 1040M photodiode array detector atached to an HP 1050 liquid chromatograph (Hewlett-Packard). TLC and HPTLC employed precoated silica gel plates $60F_{254}$ (Merck). The following TLC solvent systems were used: for saponins (a) $CHCl_3-MeOH-AcOH-H_2O$ (15:8:3:2); for sapogenins (b) toluene$-Me_2CO$ (4:1); for monosaccharides (c) $CHCl_3-MeOH-H_2O$ (8:5:1). Spray reagents for the saponins were as follows: Komarowsky reagent, a mixture (5:1) of *p*-hydroxybenzaldehyde (2% in MeOH) and H_2SO_4 50%; for the sugars, diphenylaminephosphoric acid reagent. Isolations were carried out using a medium-pressure liquid chromatography (MPLC) system [Gilson pump M 303, head pump 25SC, manometric module M 802, Injector Rheodyne 7125, Büchi column (460 × 25 mm), Büchi precolumn (110 × 15 mm)]. GLC analysis: Perkin-Elmer 900 B, glass column (200 × 0.3 cm) packed with OV 225, carrier gas, Ar, 30 mL/min.

Plant Material. The roots of *S. fortunei* were collected in July 1990 in Hubei Province, People's Republic of China. A voucher specimen (No. 5002) is deposited in the Herbarium of the Department of Pharmacognosy, School of Pharmacy, Beijing Medical University. The plant was identified by Dr. Z. H. Cui.

Extraction and Isolation. The ethanol (95%) extract (56 g), obtained by maceration of 500 g of the dried roots of *S. fortunei*, was suspended in H_2O (1 L) and submitted to successive extractions by petroleum ether, EtOAc, and *n*-BuOH. After evaporation under reduced pressure of the solvent, 5 g of a petroleum ether extract, 6 g of a EtOAc extract, and 10 g of the *n*-BuOH extract were obtained. The *n*-BuOH extract was subjected to column chromatography over silica gel 60, eluted successively by $CHCl_3-MeOH$ (1:1 → 0−100%) and by $CHCl_3-MeOH-H_2O$ (6:4:1), yielding 2 g of a crude saponin fraction. This mixture was further purified by MPLC on reversed-phase material RP-18 (Lichroprep C_{18}, Merck, 25−40 μm) (solvent: $MeOH-H_2O$, 1:1 → 3:1) and on silica gel (Si gel 60, Merck, 15−40 μm) ($CHCl_3-MeOH-H_2O$, 8:5:1) to yield **1** (25 mg) and jenisseensosides C and D as an inseparable mixture (15 mg).

Acid Hydrolysis of 1. A solution of **1** (3 mg) in 2 N aqueous CF_3COOH (5 mL) was refluxed on a water bath for 3 h. After this period, the reaction mixture was diluted with H_2O (15 mL) and extracted with CH_2Cl_2 (3 × 5 mL). The combined CH_2Cl_2 extracts were washed with H_2O and dried with Na_2SO_4. Evaporation of the solvent gave quillaic acid (co-TLC with an authentic sample). After repeated evaporations of the solvent of the aqueous layer by adding MeOH to remove the acid, the sugars were analyzed by silica gel TLC in comparison with standard sugars (solvent system c). A 2 mg quantity of saponin **1** was refluxed in 2 N aqueous CF_3-COOH (2 mL) in a sealed serum vial at 100 °C for 3 h. After this period, sugars in the hydrolysate were converted into the alditol acetates and then subjected to GLC analysis according to a method previously described.[19]

Alkaline Hydrolysis of 1. Compound **1** (7 mg) was refluxed with 5% aqueous KOH (10 mL) for 1 h. The reaction mixture was adjusted to pH 6 with dilute HCl and then extracted with H_2O-saturated *n*-BuOH (3 × 10 mL). The combined *n*-BuOH extracts were washed (H_2O). Evaporation of *n*-BuOH gave the prosapogenin **1a** (4 mg).

Compound 1: white amorphous powder; $[\alpha]^{20}_D$ −6.0° (*c* 0.132, H_2O); UV ($MeCN-H_2O$) λ_{max} 210 nm; IR (KBr) ν_{max} 3500−3400 (OH), 2930 (CH), 1725, 1735 (CO ester), 1710 (CO carboxylic acid) cm^{-1}; ^1H NMR (DMSO-d_6, 500 MHz) δ 0.69, 0.85, 0.90, 0.91, 1.07, 1.32 (each 3H, s, Me of C-26, C-29, C-25, C-30, C-24 and C-27 of the aglycon), 0.95 (3H, d, J = 6.34 Hz, Me of Fuc), 1.17 (3H, d, J = 6.06 Hz, Me of Rha), 5.12 (1H, *m*, H-12 of the aglycon), 9.50 (1H, s, aldehydic proton of the aglycon), 2.1 (3H, s, proton of the acetyl group); ^{13}C NMR (DMSO-d_6, 125 MHz), see Tables 1 and 2; long-range correlations in the HMBC spectrum used for defining the aglycon of **1** δ 1.07 (Me-24) → C-3 (81.1), C-5 (45.9), C-4 (53.9); δ 0.69 (Me-26) → C-7 (31.8), C-8 (38.9), C-9 (47.2); δ 1.32 (Me-27) → C-8 (38.9), C-13 (143.1), C-14 (40.6), C-15 (34.6); δ 0.85 (Me-29) → C-19 (46.5), C-20 (30.1), C-21 (34.9), C-30 (24.3); δ 0.91 (Me-30) → C-20 (30.1), C-21 (34.9), C-29 (32.8); negative FABMS *m/z* 1715 $[M-H]^-$, 1451 $[(M-H)-2\times132]^-$, 1131 $[(M-H)-3\times132-146-42]^-$, 823 $[(M-H)-3\times132-146-42-162-146]^-$, 485 $[(M-H)-3\times132-146-42-162-146-162-176]^-$; TLC R_f 0.20 (system a); gray-violet spots by spraying with Komarowsky reagent.

Compound 1a. The spectral data were almost superimposable with those described for the prosapogenin of jenisseensosides A−D in our previous reports.[6,7]

Jenisseensosides C and D. The spectral data (MS, ^1H NMR, and ^{13}C NMR) were in full agreement with those of jenisseensosides C and D isolated from *Silene jenisseensis*.[7]

Bioassays. The granulocyte phagocytosis assay was performed.[15−17] The T-cell activation assay was performed according to a previous protocol.[18]

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