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TRITERPENOID SAPONINS FROM *PHYTOLACCA RIVINOIDES* AND *PHYTOLACCA BOGOTENSIS*

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Key Word Index—*Phytolacca rivinoides*; *P. bogotensis*; Phytolaccaceae; triterpenoid saponins; serjanic acid.

Abstract—Investigation of the ethanolic extracts from *Phytolacca rivinoides* and *P. bogotensis* has resulted in the isolation of five new triterpenoid glycosides of serjanic acid. Their structures have been established mainly by spectroscopic methods (FAB-MS, ^{1}H, ^{13}C NMR, COSY, NOESY, TOCSY, HETCOR and *J*-resolved ^{1}H NMR) as 3-*O*-(*O*-β-D-galactopyranosyl-(1 → 3)-*O*-β-D-glucopyranosyl)serjanic acid 28-*O*-β-D-glucopyranosyl ester, 3-*O*-(*O*-β-D-glucopyranosyl-(1 → 3)-*O*-[β-D-galactopyranosyl-(1 → 4)]-*O*-β-D-glucopyranosyl)serjanic acid 28-*O*-β-D-glucopyranosyl ester, 3-*O*-(*O*-α-L-rhamnopyranosyl-(1 → 2)-*O*-β-D-glucopyranosyl-(1 → 2)-*O*-β-glucopyranosyl)serjanic acid 28-*O*-β-D-glucopyranosyl ester, 3-*O*-(*O*-β-D-glucopyranosyl-(1 → 3)-*O*-β-D-galactopyranosyl-(1 → 3)-*O*-β-D-glucopyranosyl)serjanic acid 28-*O*-β-D-glucopyranosyl ester and 3-*O*-(*O*-β-D-galactopyranosyl-(1 → 4)-*O*-[β-D-glucopyranosyl-(1 → 3)]-*O*-β-D-glucopyranosyl)serjanic acid.

INTRODUCTION

Plants of the Phytolaccaceae are widely used in Oriental and American traditional medicine against edema, inflammation, dermatitis and rheumatism [1, 2]. The family is known to be rich in saponins with biological activities, e.g. molluscicidal and antiinflammatory activity [3, 4].

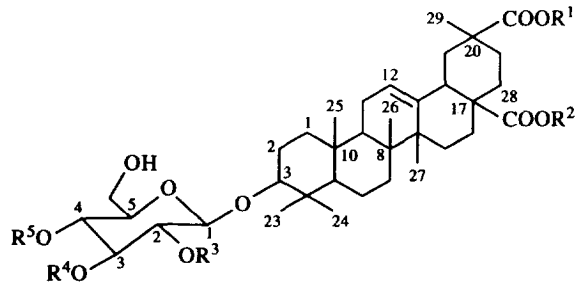
Phytolacca rivinoides and *P. bogotensis* have previously been reported to contain the triterpenoids serjanic acid and 3-acetyloleanolic acid [5, 6]. The saponin constituents from these species have apparently not been investigated previously. The only known glycosides of serjanic acid from Phytolaccaceae were isolated from *P. thyrsiflora* [7] and *P. octandra*, where the saponin was shown to have fungistatic activity [2]. The present study describes the isolation and structure elucidation of five new glycosides of serjanic acid. Compounds **1**–**3** were isolated from *P. bogotensis* and **2**, **4** and **5** from *P. rivinoides*.

RESULTS AND DISCUSSION

The ethanol extracts of *P. rivinoides* and *P. bogotensis* were partitioned to give saponin containing aqueous fractions. Biogel P2 chromatography of the aqueous fraction from *P. bogotensis* followed by repeated silica gel chromatography gave **1**–**3**. Reversed phase chromatography of the aqueous fraction of *P. rivinoides* left two fractions, which were further purified on Sephadex LH-20. Column chromatography of one fraction on silica gel gave **4**, and a compound identical to **2**, the other fraction afforded **5**.

All compounds (**1**–**5**) exhibited 31 ^{13}C NMR signals assigned to the aglycone moiety based on serjanic acid [8] (Table 1). Compounds **1**–**4** all had similar aglycone signals [deviation less than 0.2 ppm, except for C-3 (0.7 ppm)], with C-28 appearing at δ176.1, (serjanic acid and **5**, δ179.9), suggesting the C-28 acid to be esterified in these compounds. This was supported in **1**–**4** by the presence of anomeric carbon resonances at δ95.9, characteristic of an esterified sugar [9]. The downfield shift of the signal assignable to C-3 (89.1) and a number of anomeric carbons in the range δ102.1–106.6, indicated a sugar chain attached to O-3 in all compounds. Thus, based on C-13 data, saponins **1**–**4** appear to be bidesmosidic glycosides, and **5** appears to be a monodesmosidic derivative of serjanic acid.

Compound **1** had anomeric signals at δ106.6, 106.3 and 95.9, and 15 signals in the range δ62–90, suggesting sugar moieties consisting of one hexose attached at C-28 and two attached at O-3 as a disaccharide. This was supported by ^{1}H NMR and negative ion FAB-mass spectrometry. The FAB-mass spectrum of **1** exhibited a quasi-molecular anion at *m/z* 985 $[M - H]^-$ and fragment ions at *m/z* 823 and 661, corresponding to the consecutive loss of two hexose units (162 mass units). The ^{1}H NMR spectrum of **1** (Table 2) revealed the presence of three sugar moieties with the anomeric protons appearing at



	R^1	R^2	R^3	R^4	R^5
1	Me	-β-D-Glc	H	-β-D-Gal	H
2	Me	-β-D-Glc	H	-β-D-Glc	-β-D-Gal
2a	H	H	H	-β-D-Glc	-β-D-Gal
3	Me	-β-D-Glc	-β-D-Glc^{2}-^{1}α-D-Rha	H	H
3a	H	H	-β-D-Glc^{2}-^{1}α-D-Rha	H	H
4	Me	-β-D-Glc	H	-β-D-Gal^{3}-^{1}β-D-Glc	H
5	Me	H	H	-β-D-Glc	-β-D-Gal

Table 1. ^{13}C and ^{1}H NMR spectral data of compound **1** (100.6 and 399.9 MHz, respectively, in pyridine-d_5)

Position	^{13}C	^{1}H*
1	38.8	
2	26.6	2.18†
3	89.1	3.35 *dd*
4	40.1	
5	55.9	0.79†
6	18.7	
7	33.3	1.52†
8	39.6	
9	48.2	1.65 *dd*
10	37.1	
11	23.9	1.90†
12	124.2	5.59 *br s*
13	143.9	
14	42.2	
15	28.3	
16	23.7	2.10†
17	46.7	
18	42.6	3.23 *dd*
19	43.3	
20	44.1	
21	30.7	
22	34.1	1.95†
23	28.5	1.24 *s*
24	17.0	1.00 *s*
25	15.7	0.82 *s*
26	17.6	1.12 *s*
27	26.2	1.31 *s*
28	176.1	
29	28.5	1.29 *s*
30	177.0	
31	51.8	3.60 *s*

*Assigned from ^{1}H NMR and HETCOR spectrum.

†Multiplicity not discernible.

δ6.32 (*d*, J = 8.1 Hz), 5.25 (*d*, J = 7.9 Hz) and 4.91 (*d*, J = 7.8 Hz). A TOSCY spectrum enabled the grouping of the proton signals arising from the three spin systems each corresponding to a monosaccharide residue. Thus the anomeric proton at δ4.91 exhibited cross peaks with signals at δ3.97, 4.08, 4.15 and 4.25. Analogously the anomeric signal at δ6.32 was grouped with δ4.00, 4.23, 4.30 and 4.44. The anomeric proton at δ5.25 only showed connectivity with the signal at δ4.21 and the overlapping signals at δ4.56 and 4.59. A COSY spectrum revealed the connectivity of the anomeric protons with the six other protons (H-2–H-6), except in the last mentioned series of signals, where only signals arising from H-1, H-2, H-3 and H-4 appeared, due to the small coupling constant between equatorial H-4 and axial H-3 and H-5, consistent with a galactose unit. This assignment was supported by a NOESY spectrum, where NOE correlations were observed between H-1, H-3 and H-5 within each sugar unit. In the *J*-resolved ^{1}H NMR spectrum large coupling constants of the anomeric protons implied β configuration of all three hexoses. A HETCOR spectrum served to correlate the carbon signals of the three sugar residues to the proton signals, and the ^{13}C chemical shifts enabled a final identification of the sugar residues. Thus the sugar attached to C-28 was a β-D-glucopyranosyl group, exhibiting a lowfield doublet of the anomeric proton at δ6.32 (J = 8.1 Hz) [10]. The remaining signals were assignable to a glucose and a galactose moiety, and the signal of C-3 in the glucose unit (δ89.1) indicated that the interglycosidic linkage was 1 → 3, excluding the reverse assignment, with the linkage to C-3 in galactose (δ85.2) from C-1 in a terminal glucose [10, 11]. The disaccharide moiety was, therefore, identified as 3-*O*-(*O*-β-D-galactopyranosyl (1 → 3)-β-D-glucopyranoside) [7]. The attachment of C-1 in the glucose unit to O-3 in the aglycone and the interglycosidic linkage, was confirmed by a NOESY spectrum, revealing close spatial proximity of H-1 in glucose to H-3 in the aglycone and of H-3 in the glucose to H-1 in the galactose unit.

Table 2. ^{13}C MHz spectral data of the sugar moieties of saponins **1**–**5** (100.6 MHz, pyridine-d_5)

C		**1**	**2**	**3**	**4**	**5**
3-Glc	1	106.3	105.9	105.2	106.4	106.1
	2	74.5	75.8	79.0	74.6	75.8
	3	89.1	83.0	78.0	89.1	83.2
	4	70.0	77.6	73.0	69.9	77.5
	5	78.0	77.3	79.6	78.0	77.3
	6	62.8	62.4	63.6	62.8	62.4
Inner Gal	1				106.1	
	2				72.2	
	3				84.7	
	4				69.7	
	5				77.2	
	6				62.2	
Inner Glc	1			102.1		
	2			78.6		
	3			77.7		
	4			72.8[b]		
	5			79.5[a]		
	6			62.9		
Terminal Glc	1		102.6		106.6	102.8
	2		74.7[a]		76.0	74.7
	3		78.0		78.5[b]	78.0
	4		71.0		71.7	71.0
	5		78.3		78.8[a]	78.3
	6		62.1		62.8	62.1
Terminal Gal	1	106.6	105.1			105.3
	2	73.1	72.9[b]			72.9
	3	75.2	73.2[b]			73.4
	4	70.3	70.1			70.1
	5	77.5	75.2			75.2
	6	62.2	61.8			61.8
Terminal Rha	1			102.1		
	2			72.5[b]		
	3			72.1		
	4			74.5		
	5			69.6		
	6			19.2		
C-28 Glc	1	95.9	95.9	95.9	95.9	
	2	74.2	74.3[a]	74.3	74.3	
	3	79.0	79.0	79.0[a]	79.1[b]	
	4	71.1	71.1	71.1	71.2	
	5	79.4	79.5	79.4	79.5[a]	
	6	62.1	62.0	62.1	62.1	

[a,b] Signals in the vertical columns may be interchanged owing to similar correlation in HETCOR-spectrum.

The FAB-mass spectrum of **2** gave a quasimolecular anion at *m/z* 1147 $[M - H]^-$ indicating the presence of four hexose units, which was supported by the fragment ions at *m/z* 985 $[M - H - 162]^-$, 823 $[M - H - 324]$ and 661 $[M - H - 486]^-$. The ^{13}C NMR spectrum of **2** suggested a bidesmosidic structure with an estrified β-D-glucopyranose at C-28, leaving 18 ^{13}C signals to the 3-*O*-saccharide chain. This was confirmed by basic hydrolysis giving **2a** with quasimolecular ion $[M - H]^-$ at *m/z* 971 and ^{13}C NMR signals identical to those of **2**, except for the signals of C-28 and C-30 which appeared at δ180.9. From the HETCOR spectrum, the 18 carbon signals were correlated with the proton signals. This gave the unambiguous assignment of the NMR data for the anomeric positions δ5.58 (*d*, *J* = 7.9 Hz), 5.43 (*d*, *J* = 7.5 Hz) and 4.82 (*d*, *J* = 7.8 Hz) in the three sugar moieties (see Tables 2 and 3). COSY enabled the positive assignment of H-2 from the H-1 → H-2 coupling within each sugar residue, and the remaining proton signals in each spin system were located by TOCSY. NOESY supported this assignment by correlating the three anomeric protons with the H-3 and H-5 proton within each unit. The sugar moiety with the anomeric proton at δ4.82 (*d*, *J* = 7,8 Hz) was

Table 3. ^1H NMR spectral data of the sugar moieties of saponins **1**–**4** (400 MHz, pyridine-d_5)

M		**1**	**2**	**3**	**4**
3-Glc	1	4.91	4.82	4.99	4.89
	2	4.08	4.08	4.49	4.02
	3	4.25	4.58	3.97	4.19
	4	4.15	4.15	4.12	4.09
	5	3.97	3.88	4.60	3.93
	6	4.45	nd	nd	nd
Inner Gal	1				5.27
	2				4.70
	3				4.30
	4				4.71
	5				4.11
	6				nd
Inner Glc	1			5.95	
	2			4.37	
	3			3.88	
	4			4.74[b]	
	5			4.29[a]	
	6			nd	
Terninal Glc	1		5.43		5.41
	2		4.20[a]		4.04
	3		4.23		4.28
	4		4.25		4.24
	5		3.95		4.00[a]
	6		nd		nd
Terminal Gal	1	5.25	5.58		
	2	4.59	4.63[b]		
	3	4.21	4.59[b]		
	4	4.56	4.56		
	5	4.19	4.18		
	6	4.44	nd		
Terminal Rha	1			6.49	
	2			4.81[b]	
	3			4.14	
	4			4.39	
	5			5.11	
	6			1.85	
C-28 Glc	1	6.32	6.30	6.36	6.32
	2	4.23	4.22[a]	4.23	4.20
	3	4.30	4.31	4.31[a]	4.28
	4	4.44	4.45	4.45	4.41
	5	4.00	4.00	4.01	4.00[a]
	6	4.42	nd	nd	nd

[a,b] Signals interchangeable.
nd = Not discernable.

assigned as a β-D-glucopyranosyl group from the *J*-resolved ^{1}H NMR spectrum and the corresponding ^{13}C chemical shifts; δ105.9 (C-1), 75.8 (C-2), 83.0 (C-3) and 77.3 (C-5). NOE correlation from the anomeric proton in this residue to that of H -3 in the aglycone, indicated that this glucose was attached to the aglycone at C-1. Furthermore, a NOE between H-3 (δ4.58) in the glucose unit and the anomeric proton at δ5.43 indicated a 1 → 3 interglycosidic linkage between these two sugar moieties, which explains the rather lowfield chemical shift of the C-3 position. The ^{13}C NMR chemical shifts, which correspond to the spin system with the anomeric proton at δ5.43, giving carbon resonances at δ102.6 (C-1), 74.7 (C-2), 78.0 (C-3) and 78.3 (C-5), defined the sugar moiety as a terminal glucose unit [12]. The remaining sugar residue, with an anomeric proton at δ5.58, showed only signals arising from H-1, H-2, H-3 and H-4 in the TOCSY spectrum, due to the small coupling constant between equatorial H-4 and axial H-3 and H-5, consistent with a galactose unit. The corresponding carbon signals at δ105.1 (C-1), the overlapping signals at δ72.9–73.2 (C-2 and C-3) and C-5 at 75.2 (Table 2) confirmed the presence of a terminal galactose unit. Consequently this sugar moiety must be attached to C-4 in the 3-*O*-glucopyranosyl residue resulting in the lowfield chemical shift (δ77.6), which was confirmed by an NOE correlation between H-4 in the 3-*O* glucose unit and the anomeric proton in the terminal galactopyranosyl residue. Thus the 3-*O* saccharide chain was a branched trisaccharide with terminal β-D-galactopyranosyl and β-D-glucopyranosyl units attached to C-3 and C-4, respectively, in the 3-*O*-β-D-glucopyranosyl residue. Accordingly, **2** is 3-*O*-(*O*-β-D-glucopyranosyl-(1 → 3)-*O*[β-D-galactopyranosyl-(1 → 4)]-*O*-β-D-glucopyranosyl)serjanic acid 28-*O*-β-D-glucopyranosyl ester.

The ^{13}C NMR spectrum of **3** indicated that the sugar moiety at O-3 was a trisaccharide, consisting of three hexoses with anomeric resonances at δ105.2, 102.1 and 102.1. A signal at δ19.2 revealed the presence of rhamnose. This was confirmed by the fragmentation pattern of **3** in the negative FAB-mass spectrum. The quasimolecular ion $[M - H]^-$ at *m/z* 1131, corresponded to the molecular formula $C_{55}H_{88}O_{24}$, while ions at *m/z* 969 $[M - H - 162]^-$, 823 $[M - H - 308]^-$ and 661 $[M - H - 470]^-$ corresponded to loss of two normal and one deoxy sugar. The quasimolecular ion of **3a** at *m/z* 955 confirmed the presence of the rhamnose residue in the 3-*O* trisaccharide chain. The anomeric protons of **3** at δ6.49 (*d*, *J* = 2.2 Hz), 5.95 (*d*, *J* = 7.5 Hz), 4.99 (*d*, *J* = 8.3 Hz) and 6.36 (*d*, *J* = 8.0 Hz) indicated the α-configuration of rhamnose, β-configuration of the two hexoses in the trisaccharide, and confirmed the presence of a β-D-glucopyranose residue at C-28.

After subtraction of the ^{13}C signals of the C-28 glucopyranose (Table 2), the remaining 15 signals from the trisaccharide were consistent with the presence of one α-L-rhamnosyl unit and two β-D-glucopyranosyl units. The assignment was supported by TOCSY, COSY, *J*-resolved ^{1}H NMR and HETCOR spectroscopy. The spin system in the TOCSY spectrum arising from the rhamnose unit was identified by the signal at δ1.85, originating from the methyl group of rhamnose, and it was used as a starting point in the assignment of the H-5–H-1 COSY signals. The remaining spin systems were assigned to the C-28 glucopyranose unit and the two glucopyranose units in the trisaccharide chain, by use of TOCSY and the corresponding ^{13}C chemical shifts.

The glucose unit with an anomeric proton at δ4.99 showed NOE correlation to the 3-H proton of the aglycone, suggesting the attachment of this saccharide residue to the C-3 position in the aglycone. The NOE between H-2 of the 3-*O*-glucopyranose unit and the anomeric proton at δ5.95 indicated a 1 → 2 interglycosidic linkage to the other glucopyranose residue. NOE between H-2 in this glucopyranose and H-1 in the rhamnose residue, revealed the saccharide chain to be unbranched, and confirmed that the rhamnose was terminal. The assignment of the 3-*O* trisaccharide chain was in agreement with reported data [13].

The ^{13}C NMR spectrum of **4** showed 18 carbohydrate signals at δ60–106.6, beside the signals assignable to the C-28 glucopyranosyl residue. The FAB-mass spectrum confirmed the presence of four hexose units by a quasimolecular ion at *m/z* 1147 $[M - H]^-$ supported by the fragment ions at *m/z* 985 $[M - H - 162]^-$, 823 $[M - H - 324]^-$, 661 $[M - H - 486]^-$ indicating the consecutive loss of three hexose units. The 18 carbon signals were correlated with the proton signals by HETCOR techniques. COSY and TOCSY enabled the grouping of the ^{1}H NMR signals to each sugar residue. From a NOESY experiment it was possible to reveal the interglycosidic linkage of the sugars. Thus the residue with an anomeric proton at δ4.89 showed close spatial proximity between H-1 and H-3 of the aglycone, and between H-3 and H-1 of the sugar moiety with anomeric proton resonance at δ5.27. The 3-*O*-sugar moiety was established as a β-D-glucopyranose from the ^{13}C NMR signals and the *J*-resolved ^{1}H NMR spectrum, where the C-3 signal appearing at δ89.1 confirmed the 1 → 3 interglycosidic linkage [12]. The TOCSY spin system corresponding to the anomeric proton at δ5.27, correlated only to H-2 and H-3, due to a small coupling constant between H-3 and H-4 indicating a galactose residue. Carbon signals assignable to this residue reveal a downfield shift for C-3 (δ84.7) by 10 ppm, suggesting a galactopyranosyl moiety glucosylated at the C-3 position. This was confirmed by NOESY, revealing a correlation between H-3 in the galactopyranosyl moiety and the anomeric proton at δ5.41. Consequently the trisaccharide chain at 3-*O* is unbranched and the terminal sugar residue is β-D-glucopyranose (Table 2). Thus the structure of **4** is 3-*O*-(*O*-β-D-glucopyranosyl-(1 → 3)-*O*-β-D-galactopyranosyl-(1 → 3)-*O*-β-D-glucopyranosyl)serjanic acid 28-*O*-β-D-glucopyranosyl ester.

Compound **5** had 49 ^{13}C signals similar to **2** (max. deviation + 0.3 ppm), except for the signals assignable to C-13 (δ144.8), C-18 (43.1), C-19 (43.7), C-20 (44.5), C-21 (31.2), C-22 (34.8) and C-28 (179.9). As seen in Table 2, the 3-*O* saccharide chain of **5** was the same as found in **2**. The absence of the C-28 sugar moiety as shown in the

^{13}C NMR spectrum, and the low field shift of C-28 indicated that **5** was the monodesmosidic analogue of **2**. This was confirmed by basic hydrolysis producing **2a**, where C-28 and C-30 were shifted to δ180.9, the remaining signals being identical with **5** (max. deviation + 0.5 ppm for C-13). Thus **5** is 3-*O*-(*O*-β-D-galactopyranosyl-(1 → 4)-*O*-[β-D-glucopyranosyl-(1 → 3)]-*O*-β-D-glucopyranosyl)serjanic acid.

In the case of **1**–**3** and **5** the close similarity of the negative Cotton effect curves with extrema near 230 and 200 nm all attest to the identical absolute configuration of the aglycones. The monosaccharides were assumed to belong in the D-series.

EXPERIMENTAL

General. The NMR spectra were recorded in pyridine-d_5 on a Varian 400 FT-NMR (1D spectra) and on an AMX-400 Bruker instrument (2D experiments) both spectrometers operating at 400 MHz (^{1}H), and at 100.6 MHz (^{13}C). Negative ion FAB-MS: Jeol JMS HX/HX110A Tandem Mass Spectrometer, (thioglycerol matrix, Xe atoms of 8 kV). Biogel P2 (Bio Rad), Merck Kiesel gel 60 (40–63 mm) and Merck RP8 (40–63 mm) were used for CC. Precoated Kieselgel 60, F_{254} sheets (thickness 0.2 mm from Merck) were used for TLC.

Extraction and isolation. Phytolacca rivinoides and *P. bogotensis* were collected in the Pichincha province, Ecuador in 1990. Voucher specimens (FGhia290 and FGhia303) are preserved in the herbarium of Department of Economic Botany, Escuela Politecnica Nacional. The dried, chopped plant material (whole specimens, except roots of *P. rivinoides*) was repeatedly extracted with EtOH at room temp. The solvent from the combined extracts was removed in *vacuo*. the EtOH extract from *P. bogotensis* (7 g) was partitioned between C_7H_{16} and 90% aq. MeOH. After evapn *in vacuo*, the 90% aq. MeOH extract (5 g) was partitioned between 50% aq. MeOH and $CHCl_3$. Extraction with EtOH of the solid 50% aq. MeOH extract (4 g), gave a water soluble residue (1.7 g). This residue was chromatographed on a Biogel P2 column (3 × 100 cm), which gave a minor fraction (380 mg) containing the saponins. The latter fraction was submitted to CC gradient elution on LiChroprep Si-60 starting with $CHCl_3$–90% aq. MeOH (4:1) to $CHCl_3$–90% aq. MeOH (1:1), flow rate 7.5 ml min^{-1}. Fractions of 7.5 ml were collected and combined after TLC analysis on silica gel plates (Merck) to yield three main fractions A (11–13), B (14–7) and C (18–21). Fraction B (47.7 mg) was chromatographed on LiChroprep Si-60 using $CHCl_3$–MeOH–H_2O (14:6:1) giving 16.5 mg **1** and other saponins in minor amounts. Fraction C was eluted with $CHCl_3$–MeOH–H_2O on LIChroprep Si-60 to give **2** (9:5:1) (22 mg) and **3** (9.1 mg).

The EtOH extract of *P. rivinoides* (10.5 g) was partitioned between EtOAc and H_2O. The aq. layer (6.4 g) was extracted with *n*-BuOH, and after evapn in vacuo, the *n*-BuOH extract (3.0 g) was partitioned between $CHCl_3$ and H_2O (2.8 g). The aq. extract (810 mg) was chromatographed on LiChroprep RP8 with gradient elution starting with 5% aq. MeCN to 95% aq. MeCN, flow rate 7.5 ml min^{-1} and UV detection at 203 nm. The two main fractions A and B were further chromatographed on a column of Sephadex LH-20 (1 × 30 cm) using MeOH as eluent. Fr. A gave a compound (10.9 mg) identical to **2** after CC on LiChroprep Si-60 using EtOH–conc NH_3 (9:1) and a fraction with saponins in minor amount, giving **5** (1.9 mg) after repeated CC on LiChroprep Si-60 using $CHCl_3$–MeOH–H_2O (9:5:1). B gave **4** (18.2 mg).

3-*O*-(*O*-β-D-*galactopyranosyl*-(1 → 3)-O-β-D-*glucopyranosyl*)*Serjanic acid* 28-*O*-β-D-*glucopyranosyl ester* (**1**). Amorphous powder. FAB-MS, negative mode, thioglycerol matrix, *m/z* 985, 823, 661. CD (EtOH; *c* 0.0117) $\Delta\varepsilon_{201}$ 8.99; $\Delta\varepsilon_{228}$ − 1.46. ^{13}C NMR and ^{1}H NMR see Tables 1–3.

3-O-(O-β-D-*glucopyranosyl*-(1 → 3)-O-[β-D-*galactopyranosyl*-(1 → 4)]-O-β-D-*glucopyranosyl*)*Serjanic acid* 28-O-β-D-glucopyranosyl ester (**2**). Amorphous powder. FAB-MS, negative mode, thioglycerol matrix *m/z* 1147, 283, 985, 823, 661. CD (EtOH; *c* 0.0134) $\Delta\varepsilon_{203}$ 7.59; $\Delta\varepsilon_{229}$ − 1.17. Chemical shifts of the aglycon corresponded to those described for **1**. For signals of the sugar moieties, see Tables **2** and **3**.

3-O-(O-β-D-*galactopyranosyl*-(1 → 4)-O-[β-D-*glucopyranosyl*-(1 → 3)]-*O*-(β-D-*glucopyranosyl*)*Spergulagenic acid* (**2a**). From basic hydrolysis of **2**. Amorphous powder. FAB-MS, negative mode, thioglycerol matrix *m/z* 971, 809. ^{13}C NMR signals for the compound were identical to those of **2** except for the signals of C-28 and C-30 which appeared at δ180.9. The NMR signals of the sugar moieties corresponded to those of the 3-*O*-saccharide chain in **2** and **5**.

3-O-(O-α-L-*rhamnopyranosyl*-(1 → 2)-O-β-D-*glucopyranosyl*-(1 → 2)-O-β-D-*glucopyranosyl*)*Serjanic acid* 28-O-β-D-*glucopyranosyl ester* (**3**). Amorphous powder. FAB-MS, negative mode, thioglycerol matrix *m/z* 1131, 969, 823, 661. CD (EtOH; *c* 0.0161) $\Delta\varepsilon_{202}$ 9.77; $\Delta\varepsilon_{229}$ − 1.44. ^{1}H and ^{13}C NMR data for the aglycon correspond to those described for **1**. For signals of the sugar moieties, see Tables **2** and **3**.

3-O-(O-α-L-*rhamnopyranosyl*-(1 → 2)-O-β-D-*glucopyranosyl*-(1 → 2)-O-β-D-*glucopyranosyl*)*Spergulagenic acid* (**3a**). From basic hydrolysis of **3**. Amorphous powder. FAB-MS, negative mode, thioglycerol matrix *m/z* 955.

3-O-(O-β-D-*glucopyranosyl*-(1 → 3)-O-β-D-*galactopyranosyl*-(1 → 3)-O-β-D-*glucopyranosyl*)*Serjanic acid* 28-β-D-glucopyranosyl ester (**4**). Amorphous powder. FAB-MS, negative mode, thioglycerol matrix, *m/z* 1147, 985, 823, 661. ^{1}H and ^{13}C NMR data of the aglycone correspond to those described for **1**. For signals of the sugar moieties, see Table 2 and 3.

3-O-(O-β-D-*galactopyranosyl*-(1 → 4)-O-[β-D-*glucopyranosyl*-(1 → 3)]-*O*-β-D-*glucopyanosyl*)*Serjanic acid* (**5**). Amorphous powder. FAB-MS, negative mode, thioglycerol matrix *m/z* 985, 832, 661. CD (EtOH; *c* 0.0158) $\Delta\varepsilon_{195}$ 5.76; $\Delta\varepsilon_{219}$ − 1.12. ^{1}H and ^{13}C NMR data of the aglycon are identical to those described for **1**, except for C-28 which appears at δ180.9. ^{13}C sugar signals, see Table 2, ^{1}H NMR signals of the sugar moieties correspond to those listed for **2** in Table 3.

Basic hydrolysis of compounds **2** *and* **3**. The saponins **2** (6 mg) and **3** (4 mg) in 1 ml 1 M NaOH were heated at 95° in a stoppered vial for 5 hr. The reaction mixture was adjusted to pH 6 and the solvent evapd to dryness. The solid residue was extracted with MeOH to give the saponins **2a** and **3a**.

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