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Acylated phenolic glycosides from *Solenostemma argel*

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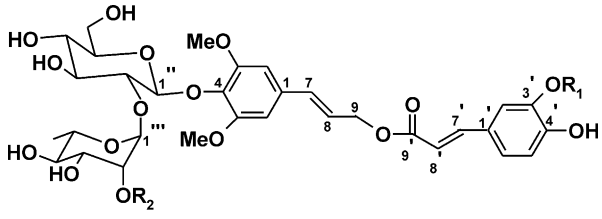
Abstract

From the aerial parts of *Solenostemma argel*, four new acylated phenolic glycosides sinapyl alcohol 9-*O*-feruloyl-4-*O*-α-rhamnopyranosyl-(1→2)-β-glucopyranoside, solargin I (**1**), sinapyl alcohol 9-*O*-caffeoyl-4-*O*-α-rhamnopyranosyl-(1→2)-β-glucopyranoside, solargin II (**2**), sinapyl alcohol 9-*O*-feruloyl-4-*O*-α-rhamnopyranosyl-(1→2)-α-rhamnopyranosyl-(1→2)-β-glucopyranoside, solargin III (**3**) and sinapyl alcohol 9-*O*-caffeoyl-4-*O*-α-rhamnopyranosyl-(1→2)-α-rhamnopyranosyl-(1→2)-β-glucopyranoside, solargin IV (**4**) have been isolated. The structures of the isolated compounds were verified by means of MS and NMR spectral analyses.

Keywords: *Solenostemma argel*; Asclepiadaceae; Phenolic glycosides; Solargins I–IV

1. Introduction

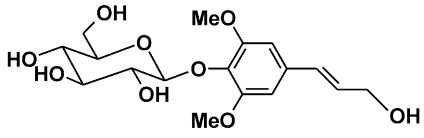
Solenostemma argel Hayne (Asclepiadaceae) is a wild perennial plant growing in the eastern desert and along the Nile in south Egypt (Tackholm, 1974). In folk medicine, it has many uses as purgative, antipyretic, expectorant and antispasmodic (Hocking, 1955). Flavonoids (El-Fishawy, 1976), monoterpene and pregnanes (Kamel et al., 2000; Hassan et al., 2001) in addition to steroids (Hamed, 2001) have been already isolated from the aerial pats of this plant. The present study deals with the isolation and assignment of new acylated phenolic glycosides from the polar fractions of the aerial parts of the plant.



Compd	R_1	R_2
(1)	Me	H
(2)	H	H
(3)	Me	Rha
(4)	H	Rha

2. Results and discussion

The methanolic extract of the aerial parts of *S. argel* was defatted with Et_2O and the aqueous layer was subjected to column chromatography on Diaion HP-20. The 40% methanol eluate was chromatographed on silica gel, followed by MPLC and HPLC on RP-18 and polyamine respectively to afford 4 glycosides (**1**–**4**).



Syringin

The molecular formula of compound **1** was deduced as $C_{33}H_{42}O_{16}$ from HR FAB-MS spectrometry (see experimental section). The ^{13}C NMR spectrum (Tables 1

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Table 1
^{13}C NMR spectral data of the aglycone and acyl moieties of compounds **1**–**4** (100 MHz, C_5D_5N)

C	1	2	3	4	Syringin[a]
1	135.8	135.7	135.6	135.3	135.6
2	105.5	105.5	105.6	105.6	105.4
3	154.0	154.0	154.0	154.1	153.9
4	132.7	132.6	132.7	132.7	134.0
5	154.0	154.0	154.0	154.1	153.9
6	105.5	105.5	105.6	105.6	105.4
7	134.0	134.1	134.1	134.2	131.1
8	123.6	123.6	123.7	123.9	129.5
9	64.9	64.9	65.1	65.1	62.8
OMe	56.6	56.7	56.7	56.7	56.6
	56.6	56.7	56.7	56.7	56.6
Feruloyl					
1′	126.4		126.9		
2′	111.6		111.9		
3′	151.2		151.2		
4′	149.8		149.9		
5′	116.8		116.7		
6′	123.6		122.7		
7′	145.9		145.2		
8′	115.0		116.0		
9′	167.2		167.0		
OMe	55.9		55.9		
Caffeoyl					
1′		126.8		126.8	
2′		115.7		115.8	
3′		150.4		150.3	
4′		148.0		148.3	
5′		116.0		116.0	
6′		123.8		123.9	
7′		145.0		145.3	
8′		115.2		115.2	
9′		166.5		166.8	

[a] Data in C_5D_5N (Martin, 1998).

Table 2
^{13}C NMR spectral data of the sugar moieties of compounds **1**–**4** (100 MHz, C_5D_5N)

C	1	2	3	4	Syringin[a]
Glc					
1″	102.3	102.3	102.3	102.4	105.0
2″	79.2	79.2	79.2	79.7	76.0
3″	78.2	78.2	78.9	78.9	78.3
4″	71.6	71.6	71.2	71.4	71.7
5″	79.1	79.2	79.2	79.2	78.6
6″	62.4	62.5	62.5	62.5	62.7
Rha					
1‴	102.2	102.1	101.7	101.8	
2‴	72.6	72.6	78.2	78.2	
3‴	72.8	72.8	71.7	71.7	
4‴	74.1	74.1	74.1	74.2	
5‴	69.7	69.7	69.7	69.7	
6‴	18.4	18.5	18.5	18.9	
Term. Rha					
1⁗			102.2	102.2	
2⁗			72.6	72.8	
3⁗			72.8	72.6	
4⁗			74.2	74.2	
5⁗			69.7	69.9	
6⁗			18.5	18.5	

[a] Data in C_5D_5N (Martin, 1998).

and 2) of **1** displayed the presence of one 2-substituted β-glucopyranosyl unit (Tipson and Horton, 1983), one unsubstituted α-rhamnopyranosyl unit (Bradbury and Jenkins, 1984) together with 10 carbon signals for a feruloyl moiety (Miyase et al., 1992) and 11 carbon signals for the aglycone. From DEPT ^{13}C NMR experiment, the carbon signals of the aglycone were assigned as two methoxyl groups (δ_C 56.6), one methylene (δ_C 64.9), four methines (δ_C 105.5 for two carbons, 134.0 and 124.6) and four quaternary carbons (δ_C 154.0 for two carbons, 135.8 and 132.7). The ^{13}C NMR spectral data of the aglycone moiety as well as the glucopyranosyl residue of **1** (Tables 1 and 2) were almost similar to those reported for sinapyl alcohol 4-*O*-β-glucopyranoside (syringin) (Martin, 1998). However, the downfield shift of C-7 (+3 ppm) together with the upfield shift of C-8 (−5.9 ppm) of **1** aglycone suggested the attachment of the feruloyl moiety to C-9 of the aglycone (Zechmeister, 1979) that was supported by the downfield shift of C-9 (+2.1 ppm) in the ^{13}C NMR spectrum (Miyase et al., 1992). At the same time, the downfield shift of C-2″ of the glucopyranosyl unit of **1** (+3.2 ppm) together with the upfield shift of the anomeric carbon signal (−2.7 ppm) revealed the attachment of a terminal rhamnopyranosy unit to C-2″ of the glucosyl unit (Bradbury and Jenkins, 1984). The β configuration of the glucopyranosyl unit was deduced from the doublet signal of its anomeric proton at δ_H 5.70 (*J* 7.3 Hz in the ^1H NMR spectrum) while the α configuration of the rhamnopyranosyl unit was established from the upfield shift of its C-5 (δ_C 69.7) in the ^{13}C NMR spectrum (Kasai et al., 1979). In the ^1H NMR spectrum of **1**, the coupling constant (15.8 Hz) of the two doublets at δ_H 6.69 and 7.95 (each 1H) indicated the *trans* configuration of the feruloyl moiety (Fiasson et al., 1997). Moreover, the *trans* configuration at C-7 and 8 of the aglycone was established from the coupling constants of the doublet signal at δ_H 6.72 (15.6 Hz, H-7) and the double triplet signal at δ_H 6.43 (15.6, 5.9 Hz, H-8). The HMQC spectral data of **1** revealed the correlations between each carbon and its directly attached protons while H–H COSY interpreted the proton-proton couplings. The HMBC spectral analysis of **1** (Fig. 1) confirmed the above mentioned data from the significant correlation peaks between H-9 of the aglycone with C-9′ of the feruloyl moiety as well as H-1 of the glucopyranosyl unit with C-4 of the aglycone and H-1 of the rhamnopyranosyl unit with C-2″ of the glucopyranosyl unit together with other correlations between H-8′ with C-1′ of the feruloyl moiety and H-8 with C-1 of the aglycone. The negative FAB-MS spectrum of **1** exhib-

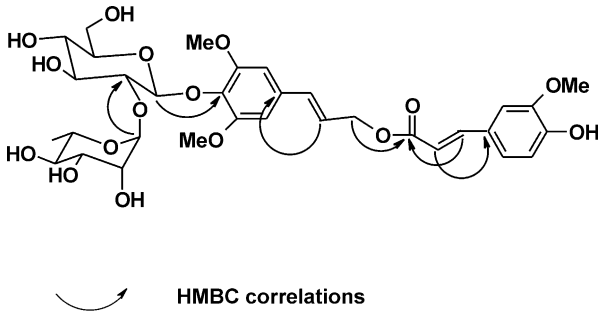

Fig. 1. Significant HMBC correlations of **1**.

ited M^+ at *m/z* 693 $[M–H]^-$ as well as significant peaks at *m/z* 547 [M–H–rhamnose]$^-$ and *m/z* 385 [M–H–(glucose + rhamnose)]$^-$. Therefore, the structure of compound **1** was assigned as sinapyl alcohol 9-*O*-feruloyl-4-*O*-α-rhamnopyranosyl-(1→2)-β-glucopyranoside and named solargin I.

The molecular formula of compound **2** was deduced as $C_{32}H_{40}O_{16}$ from HR FAB-MS spectrometry (see Experimental). The ^{13}C and DEPT NMR spectra of **2** (Tables 1 and 2) were superimposable with those of **1** in the aglycone and sugar moieties while the signals of the acyl moiety were quite different from those of **1**. The absence of the methoxyl signal at δ_C 55.9 in **2** together with downfield shift of C-2′ (+ 4.1 ppm) and upfield shift of C-4′ (−1.8 ppm) revealed that this acyl moiety is caffeoyl (Bloor, 1998). The negative FAB-MS spectrum of **2** exhibited M^+ at *m/z* 679 $[M–H]^-$ as well as significant peaks at *m/z* 533 [M–H–rhamnose]$^-$ and *m/z* 371 [M–H–(glucose + rhamnose)]$^-$. Therefore, the structure of compound **2** was assigned as sinapyl alcohol 9-*O*-caffeoyl-4-*O*-α-rhamnopyranosyl-(1→2)-β-glucopyranoside and named solargin II.

The molecular formula of compound **3** was determined as $C_{39}H_{52}O_{20}$ from HR FAB-MS spectrometry (see Experimental). Inspection of ^{13}C NMR spectral data of **3** (Tables 1 and 2) revealed their similarity to those of **1** both in the aglycone, acyl and sugar moieties. However the downfield shift of C-2‴ of the rhamnopyranosyl unit to δ_C 78.2 (+ 5.6 ppm) revealed its substitution at this position with an additional rhamnopyranosyl unit from the signals at δ_C 102.2, 72.6, 72.8, 74.2, 69.7 and 18.5 (Bradbury and Jenkins, 1984). In the ^1H NMR spectrum of **3**, the doublet signal at δ_H 5.89 with *J* constant 7.6 Hz of the anomeric proton of the glucosyl residue indicated its β configuration. Moreover, the broad doublet signal at δ_H 6.32 for two protons was assigned for two anomeric protons of the α-rhamnopyranosyl units. FAB-MS spectrum of **3** exhibited significant peaks at *m/z* 839 $[M–H]^-$, 693 [M–H–rhamnose]$^-$, 547 [M–H–(2 rhamnose)]$^-$ and 385 [M–H–(2 rhamnose + glucose)]$^-$. Consequently, the structure of compound **3** was assigned as sinapyl alcohol 9-*O*-feruloyl-4-*O*-α-rhamnopyranosyl-(1→2)-α-rhamnopyranosyl-(1→2)-β-glucopyranoside and named solargin III.

The molecular formula of compound **4** was deduced as $C_{38}H_{50}O_{20}$ from HR FAB-MS spectrometry (see experimental section). ^{13}C and DEPT NMR spectra of **4** (Tables 1 and 2) were coincident with those of **3** in the aglycone and sugar moieties while the signals of the acyl moiety were quite different than those of **3** and superimposable with those of **2**. Consequently, the acyl moiety of **4** was determined as caffeoyl. The negative FAB-MS spectrum of **4** exhibited M^+ at *m/z* 825 $[M–H]^-$ as well as significant peaks at *m/z* 825 $[M–H]^-$, 679 [M–H–rhamnose]$^-$, 533 [M–H– (2 rhamnose)]$^-$ and 371 [M–H–(2 rhamnose + glucose)]$^-$. Therefore, the structure of compound **3** was assigned as sinapyl alcohol 9-*O*-caffeoyl-4-*O*-α-rhamnopyranosyl-(1→2)-α-rhamnopyranosyl-(1→2)-β-glucopyranoside and named solargin IV.

3. Experimental

3.1. General

^1H and ^{13}C NMR (TMS as internal standard): 400 MHz and 100 MHz respectively were recorded on a Jeol JNM α-400 spectrometer. FAB MS spectra were taken on a Jeol JMS-SX 102 spectrometer by direct inlet method at an ionizing voltage of 70 eV. Optical rotations were measured with a Union PM-1 digital polarimeter. MPLC: RP-18 column (20 mm i.d.×40 cm); flow rate of mobile phase 3 ml/min. HPLC: polyamine column (20 mm i.d.×25 cm, YMC) with a Toyo Soda high speed chromatograph HLC-803 D pump and a Tosoh refraction index (RI-8) detector; flow rate of mobile phase 6 ml/min, injection vol. 0.8–1.0 ml. On the polyamine column 88% MeCN was used. CC: Kieselgel 60 (70–230 mesh, Merck) and Diaion HP 20 (Mitsubishi). TLC: silica gel 60 precoated plates F-254 and HPTLC using RP-18 precoated plates, F-254 s (Merck).

3.2. Plant material

The aerial parts of *S. argel* Hayne (Asclepiadaceae) were collected in March 1998 from the eastern desert of Egypt and identified by Dr. Salah El-Naggar, Department of Botany and Plant Taxonomy, Faculty of Science, Assiut University, Assiut, Egypt. A voucher specimen is deposited at the Herbarium of Department of Pharmacognosy, Faculty of Pharmacy, Assiut University, Assiut, Egypt.

3.3. Extraction and isolation of compounds (*1*–*4*)

The air-dried powdered aerial parts of *S. argel* (1 kg) were extracted with MeOH. The dried methanolic extract was suspended in H_2O and defatted with Et_2O. The defatted aq. layer was applied to a column of Dia-

ion HP-20 and eluted with H_2O, 40% MeOH, 80% MeOH, MeOH and acetone successively. The 40% MeOH eluate was chromatographed by silica gel CC using CH_2Cl_2–MeOH–H_2O (75:25:2–65:35:5) to give 6 fractions. Fraction 4 was subjected to MPLC on RP-18 using 45% MeOH followed by HPLC on polyamine column using 88% MeCN to afford compounds **1** (65 mg), **2** (20 mg), **3** (55 mg) and **4** (18 mg).

3.3.1. Solargin I (*1*)

White amorphous powder, $[\alpha]_D^{23}$ −11.3° (MeOH; *c* 0.80), R_t 16 min (polyamine, 88% MeCN). HR FAB-MS (negative) *m/z*: 693.6652 $[M–H]^-$ $C_{33}H_{41}O_{16}$ (req. 693.6690). ^{13}C NMR (C_5D_5N, Tables 1 and 2). ^1H NMR (C_5D_5N): δ_H 7.95 (1H, *d*, *J* = 15.8 Hz, H-7′), 7.31 (1H, *d*, *J* = 1.7 Hz, H-2′), 7.23 (1H, *dd*, *J* = 1.7, 8.0 Hz, H-6′), 7.18 (1H, *d*, *J* = 8.0 Hz, H-5′), 6.80 (2H, *s*, H-2, 6), 6.72 (1H, *d*, *J* = 15.6 Hz, H-7), 6.69 (1H, *d*, *J* = 15.8 Hz, H-8′), 6.43 (1H, *dt*, *J* = 15.6, 5.9 Hz, H-8), 6.2 (1H, *bs*, H-1 Rha), 5.70 (1H, *d*, *J* = 7.3 Hz, H-1 Glc), 4.99 (2H, *bd*, *J* = 5.9 Hz, H-9), 3.80 (6H, *s*, 2 OMe at C-3,5), 3.70 (3H, *s*, OMe at C-3′), 1.61 (3H, *d*, *J* = 6.4 Hz, Me-6 Rha).

3.3.2. Solargin II (*2*)

White amorphous powder, $[\alpha]_D^{23}$ −13.7° (MeOH; *c* 0.62), R_t 17 min (polyamine, 88% MeCN). HR FAB-MS (negative) *m/z*: 679.6371 $[M–H]^-$ $C_{32}H_{39}O_{16}$ (req. 679.6420). ^{13}C NMR (C_5D_5N, Tables 1 and 2). ^1H NMR (C_5D_5N): δ_H 8.17 (1H, *d*, *J* = 1.7 Hz, H-2′), 7.45 (1H, *dd*, *J* = 1.7, 8.0 Hz, H-6′), 7.17 (1H, *d*, *J* = 8.0 Hz, H-5′), 6.90 (1H, *d*, *J* = 13.0 Hz, H-7′), 6.80 (1H, *d*, *J* = 15.3 Hz, H-7), 6.70 (2H, *s*, H-2, 6), 6.40 (1H, *dt*, *J* = 15.3, 6.1 Hz, H-8), 6.30 (1H, *bs*, H-1 Rha), 6.0 (1H, *d*, *J* = 13.0 Hz, H-8′), 5.80 (1H, *d*, *J* = 7.3 Hz, H-1 Glc), 4.90 (2H, *bd*, *J* = 6.1 Hz, H-9), 3.80 (6H, *s*, 2 OMe at C-3, 5), 1.80 (3H, *d*, *J* = 6.1 Hz, Me-6 Rha).

3.3.3. Solargin III (*3*)

White amorphous powder, $[\alpha]_D^{23}$ −14.4° (MeOH; *c* 0.20), R_t 21 min (polyamine, 88% MeCN). HR FAB-MS (negative) *m/z*: 693.6652 $[M–H]^-$ $C_{33}H_{41}O_{16}$ (req. 693.6690). ^{13}C NMR (C_5D_5N, Tables 1 and 2). ^1H NMR (C_5D_5N): δ_H 7.93 (1H, *d*, *J* = 16.1 Hz, H-7′), 7.54 (1H, *d*, *J* = 8.8 Hz, H-5′), 7.28 (1H, *d*, *J* = 1.9 Hz, H-2′), 7.18 (1H, *dd*, *J* = 1.9, 8.8 Hz, H-6′), 6.81 (2H, *s*, H-2, 6), 6.79 (1H, *d*, *J* = 13.1 Hz, H-7), 6.70 (1H, *d*, *J* = 16.1 Hz, H-8′), 6.42 (1H, *dt*, *J* = 13.1, 6.1 Hz, H-8), 6.32 (2H, *bs*, 2 H-1 Rha), 5.89 (1H, *d*, *J* = 7.6 Hz, H-1 Glc), 5.00 (2H, *bd*, *J* = 6.1 Hz, H-9), 3.79 (6H, *s*, 2 OMe at C-3, 5), 3.75 (3H, *s*, OMe at C-3′), 1.63 (6H, *d*, *J* = 6.4 Hz, 2 Me-6 Rha).

3.3.4. Solargin IV (*4*)

White amorphous powder, $[\alpha]_D^{23}$ −17.2° (MeOH; *c* 0.35), R_t 22 min (polyamine, 88% MeCN). HR FAB-MS (negative) *m/z*: 825.7853 $[M–H]^-$ $C_{38}H_{49}O_{20}$ (req. 825.8740). ^{13}C NMR (C_5D_5N, Tables 1 and 2). ^1H NMR (C_5D_5N): δ_H 7.90 (1H, *d*, *J* = 1.8 Hz, H-2′), 7.47 (1H, *dd*, *J* = 1.8, 8.4 Hz, H-6′), 7.2 (1H, *d*, *J* = 8.4 Hz, H-5′), 6.88 (1H, *d*, *J* = 13.8 Hz, H-7′), 6.78 (2H, *s*, H-2, 6), 6.68 (1H, *d*, *J* = 16.0 Hz, H-7), 6.40 (1H, *dt*, *J* = 16.0, 5.6 Hz, H-8), 6.30 (2H, *bs*, 2 H-1 Rha), 6.05 (1H, *d*, *J* = 13.8 Hz, H-8‘), 5.90 (1H, *d*, *J* = 7.6 Hz, H-1 Glc), 4.90 (2H, *bd*, *J* = 5.6 Hz, H-9), 3.80 (6H, *s*, 2 OMe at C-3, 5), 1.75 (3H, *d*, *J* = 6.1 Hz, Me-6 Rha), 1.64 (3H, *d*, *J* = 6.0 Hz, Me-6 Rha).

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