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Phenolic and iridoid glycosides from *Strychnos axillaris*

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Abstract

Five phenolic glycosides **1**–**5** and an iridoid glucoside **6** were isolated, together with 22 known compounds, from the dried barks and woods of *Strychnos axillaris*. Their structures were determined by application of spectroscopic (NMR, MS) and chemical methodologies.

Keywords: *Strychnos axillaris*; Loganiaceae; Phenolic glycosides; Iridoid glucoside; Calleryanin; Vanilloloside; Cantleyoside

1. Introduction

The genus *Strychnos* (Loganiaceae) is well-known as a rich source of various bioactive indole alkaloids represented by strychnine (Bisset, 1980). Several β-carboline glucoalkaloids closely related to indole alkaloids have been isolated from *Strychnos mellodora* (Brandt et al., 1999). In continuation of our phytochemical studies on constituents of the genus *Strychnos* (Itoh et al., 2005, 2006), we examined *S. axillaris* Colebr. (=*S. pubescens* C.B. Clarke) collected in Thailand. This species has been traditionally used in East Asia as an arrow poison (Perry, 1980), but no phytochemical study of this plant has been reported. From our interests in glucoalkaloids in the polar fraction (Itoh et al., 2003), we investigated the *n*-BuOH-soluble fraction of this plant. In this paper, we report the isolation and structure elucidation of six new glycosides along with 22 known compounds from *S. axillaris*.

2. Results and discussion

The dried barks and woods of *S. axillaris* were extracted with MeOH under conditions of reflux. The extract was successively partitioned between H_2O and $CHCl_3$ and between H_2O and *n*-BuOH. The *n*-BuOH-soluble fraction was separated by a combination of chromatographic procedures to afford six new compounds **1**–**6** together with 22 known compounds: tachioside (Zhong et al., 1999), isotachioside (Zhong et al., 1999), 3,4,5-trimethoxyphenol 1-*O*-β-D-apiofuranosyl-(1→6)-β-D-glucopyranoside (Duynstee et al., 1999), calleryanin (**7**) (Challice et al., 1980), vanilloloside (**8**) (Ida et al., 1994), 3,5-dimethoxy-4-hydroxybenzyl alcohol 4-*O*-β-D-glucopyranoside (Kitajima et al., 1998), caffeoylcalleryanin (**9**) (Challice and Williams, 1968), (7*S*, 8*R*)-balanophonin 4-*O*-β-D-glucopyranoside (Warashina et al., 2005), (+)-syringaresinol *O*-β-D-glucoside (Lami et al., 1991), scorzonoside (Tolstikhina and Semenov, 1998), liriodendrin (Kobayashi et al., 1985), (−)-pinoresinol 4-*O*-β-D-glucopyranoside (Sugiyama and Kikuchi, 1991), (+)-lariciresinol 4′-*O*-β-D-glucoside (Sugiyama and Kikuchi, 1993), loganic acid (Nakamoto et al., 1988), loganin (Nakamoto et al., 1988), sweroside (Jensen et al., 1981), picconioside I (Damtoft et al., 1997), cantleyoside (**10**) (Kocsis et al., 1993), triplostoside A (Ma et al., 1992), strictosidinic acid (Arbain et al., 1993), 3,4-di-*O*-caffeoylquinic acid (Wang et al., 1992), and 3,5-di-*O*-caffeoylquinic acid (Wald et al., 1989). The structures of the new compounds were determined as follows.

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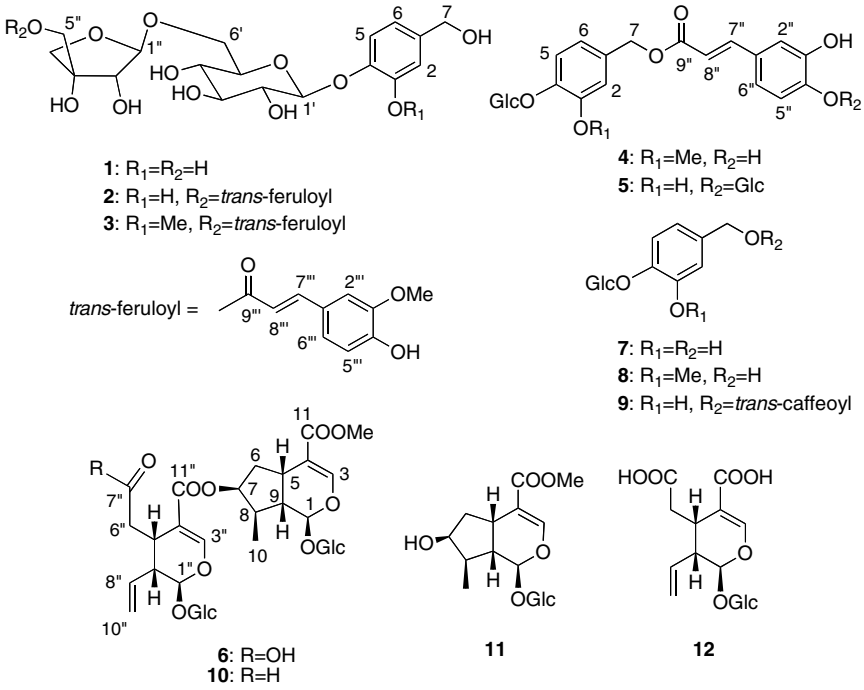

Compound **1** was obtained as an amorphous powder, $[\alpha]_D$ −81. Its HR-SIMS showed a pseudomolecular ion $[M–H]^-$ at *m/z* 433.1354, indicating a molecular formula of $C_{18}H_{26}O_{12}$. It also had UV maxima at 202 and 278 nm and IR bands at 3386 and 1508 cm^{-1}. Its ^1H NMR spectrum (Table 1) exhibited an AMX spin system at δ 6.79 (*dd*, $J = 8.5$, 2.0 Hz), 6.87 (*d*, $J = 2.0$ Hz), and 7.15 (*d*, $J = 8.5$ Hz), a signal for an oxygenated benzylic methylene group at δ 4.48 (*s*), as well as resonances for a β-glucopyranosyl unit at δ 3.35–4.70, implying the presence of a calleryanin (**7**) moiety (Challice et al., 1980). This was supported by the NOESY correlations between H-2 (δ 6.87), H-6 (δ 6.79) and H_2-7 (δ 4.48) and between H-5 (δ 7.15) and H-1′ of glucose unit (δ 4.70). The ^1H and ^{13}C NMR spectra of **1** showed additional signals for a β-apiofuranosyl unit at δ_H 3.59 (2 H, *s*), 3.76 (1H, *d*, $J = 9.5$ Hz), 3.92 (1H, *d*, $J = 2.5$ Hz), 3.97 (1H, *d*, $J = 9.5$ Hz), 4.99 (1H, *d*, $J = 2.5$ Hz) and δ_C 65.6, 75.0, 78.1, 80.5, 111.1, besides the resonances for a calleryanin moiety. Acid hydrolysis of **1** liberated D-glucose and D-apiose, which were identified by GLC analysis of their thiazolidine derivatives (Hara et al., 1986). On the basis of the HMBC correlation between the H-1″ of the apiose moiety and C-6′ of the glucose moiety, it was concluded that an apiose was linked to C-6′ of the calleryanin moiety, which was supported by the downfield shift of C-6′ and the upfield shift of C-5′ by glycosidation when compared with **7**. Accordingly, compound **1** was characterized as 6′-*O*-β-D-apiofuranosylcalleryanin.

The second new glycoside **2** was also obtained as an amorphous powder. The ^1H NMR spectroscopic features of **2** were closely similar to those of **1** except for the signals for another AMX spin system [δ 6.79 (*d*, $J = 8.5$ Hz), 7.05 (*dd*, $J = 8.5$, 2.0 Hz), 7.16 (*d*, $J = 2.0$ Hz)], *trans*-olefinic protons [δ 6.38, 7.65 (each *d*, $J = 16.0$ Hz)], and a methoxy group [δ 3.86 (*s*)]. These resonances were assignable to a *trans*-feruloyl group by its NOESY correlation between a methoxy singlet and an aromatic proton at δ 7.16 (*d*, $J = 2.0$ Hz) and HMBC interactions between aromatic protons at δ_H 7.05 and 7.16 and an olefinic carbon at δ_C 147.4. The ester linkage of the *trans*-feruloyl group to the hydroxyl group at C-5″ of apiose moiety was demonstrated by the downfield shift of C-5″ relative to those in **1** and the HMBC cross peaks between H_2-5″ [δ_H 4.27, 4.30 (each *d*, $J = 11.5$ Hz)] and C-9‴ of *trans*-feruloyl group (δ_C 169.0). Thus, compound **2** was deduced to be 5″-*O*-*trans*-feruloyl-6′-*O*-β-D-apiofuranosylcalleryanin.

The spectroscopic properties of compound **3** were quite similar to those of **2** except for additional signals for a methoxy group at δ_H 3.84 and δ_C 56.7 (Table 1). The location of the methoxy group was determined to be at C-3 by comparison of the ^1H and ^{13}C NMR spectroscopic data for the benzyl alcohol moiety with those of vanilloloside (**8**) (Ida et al., 1994). Further evidence was obtained from the NOESY correlation between H-2 [δ_H 6.99 (*d*, $J = 2.0$ Hz)] and the methoxy singlet. Accordingly, compound **3** was established as 5″-*O*-*trans*-feruloyl-6′-*O*-β-D-apiofuranosylvanilloloside.

Compound **4**, $C_{23}H_{26}O_{11}$, was also isolated as an amorphous powder. Its ^1H and ^{13}C NMR spectroscopic data (Table 2) showed that **4** consisted of a vanilloloside (**8**) moiety esterified with a *trans*-caffeoyl group. Acylation of the hydroxyl group at C-7 was determined by the HMBC correlation between H_2-7 and C-9″ of the caffeoyl group, and by comparison of the ^{13}C NMR spectroscopic data of **4** with those of **8** and caffeoylcalleryanin (**9**) (Challice and Williams, 1968). Accordingly, the structure of **4** was determined to be 7-*O*-*trans*-caffeoylvanilloloside.

Table 1
^{1}H and ^{13}C NMR spectral data of compounds 1–3 in CD3OD

	1				**2**				**3**			
	δ_C	δ_H			δ_C	δ_H			δ_C	δ_H		
1	138.5				138.6				137.8			
2	116.0	6.87	d	(2.0)	116.1	6.84	d	(2.0)	112.7	6.99	d	(2.0)
3	148.4				148.4				150.8[a]			
4	146.0				146.1				147.3			
5	118.9	7.15	d	(8.5)	118.9	7.15	d	(8.5)	118.1	7.12	d	(8.0)
6	119.7	6.79	dd	(8.5, 2.0)	119.7	6.76	dd	(8.5, 2.0)	120.9	6.86	dd	(8.0, 2.0)
7	64.7	4.48	s		64.9	4.42	s		65.0	4.49	s	
3-OMe									56.7	3.84	s	
glc												
1′	104.5	4.70	d	(8.0)	104.5	4.67	d	(8.0)	103.0	4.81	d	(7.5)
2′	74.9	3.48	brt	(8.5)	74.9	3.48	dd	(9.0, 8.0)	75.0	3.49	dd	(9.0, 7.5)
3′	77.6	3.44	t	(9.0)	77.6	3.42	t	(9.0)	77.9	3.43	t	(9.0)
4′	71.6	3.35	brt	(9.0)	71.7	3.34	brt	(9.0)	71.8	3.33	brt	(9.5)
5′	77.2	3.55	ddd	(9.5, 6.5, 2.0)	77.1	3.57	ddd	(9.0, 6.5, 2.0)	77.1	3.56	ddd	(9.5, 6.5, 2.0)
6′	68.8	3.63	dd	(11.0, 6.5)	68.7	3.63	dd	(11.0, 6.5)	68.8	3.61	dd	(11.0, 6.5)
6′		4.02	dd	(11.0, 2.0)		4.05	dd	(11.0, 2.0)		4.00–4.04	m	
api												
1″	111.1	4.99	d	(2.5)	110.8	5.02	d	(2.0)	110.8	4.99	d	(2.0)
2″	78.1	3.92	d	(2.5)	78.6	3.98	d	(2.0)	78.6	3.94	d	(2.0)
3″	80.5				79.0				79.0			
4″	75.0	3.76	d	(9.5)	75.1	3.84	d	(9.5)	75.1	3.82	d	(9.5)
4″		3.97	d	(9.5)		4.05	d	(9.5)		4.02	d	(9.5)
5″	65.6	3.59	s		67.4	4.27	d	(11.5)	67.4	4.24	d	(11.0)
5″						4.30	d	(11.5)		4.28	d	(11.0)
acyl												
1‴					127.6				127.7			
2‴					111.8	7.16	d	(2.0)	111.8	7.17	d	(2.0)
3‴					149.5				149.5			
4‴					150.9				150.9[a]			
5‴					116.6	6.79	d	(8.5)	116.6	6.80	d	(8.5)
6‴					124.3	7.05	dd	(8.5, 2.0)	124.3	7.06	dd	(8.5, 2.0)
7‴					147.4	7.65	d	(16.0)	147.3	7.64	d	(16.0)
8‴					115.1	6.38	d	(16.0)	115.1	6.38	d	(16.0)
9‴					169.0				168.9			
3‴-OMe					56.5	3.86	s		56.5	3.87	s	

[a] Assignments with the same superscript may be interchanged.

Compound **5**, $C_{28}H_{34}O_{16}$, was obtained as an amorphous powder and its ^{1}H and ^{13}C NMR spectroscopic features were closely similar to those of **9**, except for additional signals arising from a β-glucopyranosyl unit. The attachment of the second glucopyranosyl unit at C-4″ of caffeoylcalleryanin moiety was confirmed by the HMBC correlation between H-1‴ of glucose unit and C-4″ and by the NOESY interaction between H-5″ and H-1‴. Accordingly, compound **5** was assigned to 4″-*O*-β-D-glucopyranosyl-caffeoylcalleryanin.

Compound **6**, named axillaroside, was assigned the molecular formula $C_{33}H_{46}O_{20}$ from its HR-SIMS. Its ^{1}H NMR spectrum showed signals for two olefinic protons at δ 7.24 (*d*, $J = 1.5$ Hz) and 7.49 (*d*, $J = 2.0$ Hz), two acetal proton resonances at δ 5.28 (*d*, $J = 5.0$ Hz) and 5.50 (*d*, $J = 4.5$ Hz), two sets of signals for β-glucopyranosyl units at δ 3.20–4.68, a doublet for a secondary methyl group at δ 1.07 ($J = 6.5$ Hz), and resonances for a terminal vinyl group at δ 5.23 (*dd*, $J = 10.0$, 1.5 Hz), 5.27 (*dd*, $J = 17.0$, 1.0 Hz), and 5.66 (*br dt*, $J = 17.0$, 10.0 Hz), indicating the presence of an iridoid glucoside unit and a secoiridoid glucoside unit in the molecule. These spectroscopic features were similar to those of cantleyoside (**10**), which was also isolated in this study, except for the absence of the signal for an aldehyde proton in its ^{1}H NMR spectrum (Kocsis et al., 1993). Its ^{13}C NMR spectrum exhibited three carbonyl carbon resonances at δ 168.0, 169.5, and 176.1. These observations suggested that glucoside **6** was composed of loganin (**11**) unit and secologanoside (**12**) units. The presence of a secologanoside unit was further supported by a fragment ion peak at *m*/*z* 389.

The site of ester linkage was confirmed by HMBC experiments. The signal at δ_c 169.5 was assigned to C-11 of the loganin moiety by HMBC correlations from H-3, H-5 and the methoxyl resonance at δ 3.69, and the signal at δ_c 176.1 was assigned to C-7″ of the secologanoside moiety by HMBC correlations from H-5″ and H_2-6″. The remaining carbonyl carbon resonance at δ_c 168.0, which was assigned to C-11″ of the secologanoside moiety by HMBC interactions with H-3″ and H-5″, was correlated with H-7 [δ 5.20 (*br t*, $J = 5.5$ Hz)] of the loganin unit, indicating that the C-11 carboxyl group of the secologanoside moiety was linked to the

Table 2
^{1}H and ^{13}C NMR spectral data of compounds **4** and **5** in CD_3OD

	4				**5**			
	δ_C	δ_H			δ_C	δ_H		
1	132.7				133.3			
2	114.0	7.06	d	(2.0)	117.2	6.91	d	(2.0)
3	150.9				148.4[a]			
4	148.0				146.7			
5	117.9	7.16	d	(8.0)	118.7	7.18	d	(8.0)
6	122.5	6.96	dd	(8.0, 2.0)	121.0	6.84	dd	(8.0, 2.0)
7	67.1	5.15	s		67.0	5.11	s	
3-OMe	56.8	3.87	s					
glc								
1′	102.8	4.91	d	(7.0)	104.2	4.77	d	(7.5)
2′	75.0	3.50	brt	(8.0)	74.8[b]	3.51	dd	(9.0, 7.5)
3′	77.9	3.46	brt	(9.0)	77.5[c]	3.38–3.50	m	
4′	71.4	3.36–3.46	m		71.3	3.38–3.50	m	
5′	78.3	3.36–3.46	m		78.3[d]	3.38–3.50	m	
6′	62.5	3.69	dd	(12.0, 5.0)	62.4	3.71	dd	(12.0, 5.0)
6′		3.87	dd	(12.0, 1.5)		3.90	dd	(12.0, 2.0)
acyl								
1″	127.7				131.3			
2″	115.2	7.03	d	(2.0)	116.0	7.11	d	(2.0)
3″	146.9				148.6[a]			
4″	149.7				148.9			
5″	116.5	6.77	d	(8.0)	118.1	7.19	d	(8.5)
6″	123.1	6.94	dd	(8.0, 2.0)	122.3	7.04	dd	(8.5, 2.0)
7″	147.2	7.56	d	(16.0)	146.3	7.59	d	(16.0)
8″	115.0	6.28	d	(16.0)	117.2	6.38	d	(16.0)
9″	169.1				168.6			
glc								
1‴					103.5	4.85	d	(7.5)
2‴					74.9[b]	3.51	dd	(9.0, 7.5)
3‴					77.6[c]	3.38–3.50	m	
4‴					71.3	3.38–3.50	m	
5‴					78.4[d]	3.38–3.50	m	
6‴					62.4	3.71	dd	(12.0, 5.0)
6‴						3.90	dd	(12.0, 2.0)

[a–d] Assignments with the same superscript may be interchanged.

C-7 hydroxy group of loganin component. Accordingly, axillaroside was formulated as shown in formula **6**.

3. Concluding remarks

In the present study, five new phenolic glycosides **1**–**5** with a benzyl alcohol moiety and a new iridoid glucoside **6** were isolated along with a glucoalkaloid strictosidic acid. Iridoid glycosides and their related indole alkaloids have previously been reported from the genus *Strychnos*. However, this is the first instance of the isolation of benzyl alcohol glycosides such as calleryanin and vanilloloside from the genus.

4. Experimental

4.1. General

UV spectra were recorded on a Shimadzu UV-2500PC spectrophotometer and IR spectra on a Shimadzu FTIR-8200 spectrophotometer. ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra were recorded on a Varian VXR-500 spectrometer with TMS as an internal standard. MS and HRMS were obtained with a Hitachi M-4100 mass spectrometer. Glycerol or 3-nitrobenzyl alcohol was used as the matrix for SIMS and HRSIMS. MPLC was carried out with Wakogel FC-40 or Wakosil 40C18. TLC was performed on precoated Kieselgel $60F_{254}$ plates (Merck).

4.2. Plant material

The bark and wood of *Strychnos axillaris* Colebr. were collected at Sakae Rat, Nakhon Ratchasima, Thailand in March 1986 and identified by Dr. T. Smitinand, The Forest Herbarium, Royal Forest Department, Bangkok, Thailand.

4.3. Extraction and isolation

Dried and powdered mixture (2000 g) of bark and wood of *S. axillaris* was extracted with MeOH under conditions of reflux for 1 h, with the extracts concentrated *in vacuo* and a part (39 g) of the resulting residue (130 g) resuspended

in H_2O and extracted successively with $CHCl_3$ and *n*-BuOH. The residue (23.6 g) from the *n*-BuOH layer was fractionated using reversed-phase MPLC. Elution with H_2O-MeOH mixtures of the indicated MeOH content (%v/v) gave nine frs, 1 (5%, 286 mg), 2 (10%, 160 mg), 3 (15%, 414 mg), 4 (20%, 197 mg), 5 (20%, 797 mg), 6 (25–30%, 1.11 g), 7 (30–35%, 457 mg), 8 (35%, 4.59 g), and 9 (40–45%, 1.54 g). Fr. 1 was purified by preparative HPLC (μBondasphere 5μ C18-100 Å, MeOH–H_2O, 1:4, MeOH–MeCN–H_2O, 1:2:97, v/v/v) and preparative TLC (EtOAc–C_6H_6–EtOH, 4:1:3, $CHCl_3$–MeOH, 7:3, $CHCl_3$–MeOH–H_2O, 70:30:3, v/v/v) to afford **7** (68.7 mg), **8** (20.7 mg), loganic acid (18.8 mg), tachioside (3.4 mg), and isotachioside (3.3 mg), respectively. In the same way, frs. 2–9 were purified by a combination of reversed-phase MPLC with MeOH–H_2O, normal-phase MPLC with $CHCl_3$–MeOH, preparative HPLC (μBondasphere 5μ C18-100 Å, MeOH–H_2O, 2:8–11:9 (by vol.), MeOH–H_2O–AcOH, 40:60:1, MeCN–H_2O, 3:17, MeCN–H_2O–AcOH, 20:85:1, 15:85:1, v/v/v) and preparative TLC ($CHCl_3$–MeOH, 7:3; $CHCl_3$–MeOH–H_2O, 70:30:3, 14:6:1, v/v/v). Fr. 2 yielded **1** (15.2 mg), **8** (19.6 mg), loganic acid (20.9 mg), and 3,5-dimethoxy-4-hydroxybenzyl alcohol 4-*O*-β-D-glucopyranoside (7.6 mg); fr. 3: loganic acid (196 mg); fr. 4: loganin (4.2 mg), sweroside (24.6 mg), and 3,4,5-trimethoxyphenol 1-*O*-β-D-apiofuranosyl-(1 → 6)-β-D-glucopyranoside (20.1 mg); fr. 5: sweroside (46.8 mg), 3,4-di-*O*-caffeoylquinic acid (17.0 mg), liriodendrin (158 mg), and 3,5-di-*O*-caffeoylquinic acid (15.7 mg); fr. 6: 3,4-di-*O*-caffeoylquinic acid (40.2 mg), scorzonoside (9.7 mg), liriodendrin (588 mg), and (+)-lariciresinol 4-*O*-β-D-glucoside (21.1 mg); fr. 7: **2** (2.9 mg), **3** (2.8 mg), **5** (57.8 mg), (7*S*, 8*R*)-balanophonin 4-*O*-β-D-glucopyranoside (3.7 mg), **10** (4.9 mg), (+)-syringaresinol *O*-β-D-glucoside (10.8 mg), 3,4-di-*O*-caffeoylquinic acid (28.4 mg), scorzonoside (8.8 mg), (−)-pinoresinol 4-*O*-β-D-glucopyranoside (5.7 mg), and strictosidinic acid (8.7 mg); fr. 8: **4** (13.4 mg), **5** (7.5 mg), **6** (5.6 mg), **9** (535 mg), **10** (768 mg), and 3,4-di-*O*-caffeoylquinic acid (78.4 mg); fr. 9: **10** (25.7 mg), picconioside I (11.9 mg), and triplostoside A (160 mg).

4.4. 6′-*O*-β-D-Apiofuranosylcalleryanin (*1*)

White powder; $[\alpha]_D^{23}$ − 81 (*c* 1.5, MeOH); UV λ_{max}^{MeOH} nm (log ε): 202 (3.82), 278 (3.39); IR ν_{max}^{KBr} cm^{-1} : 3386, 1508; For ^1H and ^{13}C NMR spectroscopic data, see: Table 1; NOESY: H-7 and H-3, 5; H-6 and H-1′; HMBC: H-3 to C-1, 2, 5, 7; H-5 to C-1, 3, 7; H-6 to C-1, 2, 4; H-7 to C-3, 4, 5; H-1′ to C-1; H-1″ to C-6′; negative ion HRSIMS *m*/*z* 433.1354 (calcd for $C_{18}H_{25}O_{12}$, 433.1347).

4.5. 5″-*O*-trans-Feruloyl-6′-*O*-β-D-apiofuranosylcalleryanin (*2*)

White powder; $[\alpha]_D^{25}$ − 44 (*c* 0.3, MeOH); UV λ_{max}^{MeOH} nm (log ε): 219sh (4.26), 231sh (4.14), 286 (3.98), 298sh (3.99), 327 (4.14); IR $\nu_{max}^{KBr}$$cm^{-1}$: 3426, 1717, 1597, 1516; For ^1H and ^{13}C NMR spectroscopic data, see: Table 1; NOESY: H-7 and H-3, 5; H-6 and H-1′; H-6′ and H-1″; H-2‴, 6‴ and H-7‴; OMe and H-2‴; HMBC: H-3 to C-1, 5; H-5 to C-1; H-6 to C-1, 2; H-7 to C-3, 4, 5; H- 1′ to C-1; H-6′ to C-1″; H- 5″ to CO; H-2‴ to C-3‴, 4‴, 7‴; H-5‴ to C-3‴, 4‴; H-6‴ to C-2‴, 4‴; H-7‴ to C-2‴, 6‴, CO; H-8‴ to C-1‴, CO; OMe to C-3‴; negative ion SIMS *m*/*z* 609 [M−H]$^-$, 139; negative ion HRSIMS *m*/*z* 609.1823 (calcd for $C_{28}H_{33}O_{15}$, 609.1820).

4.6. 5″-*O*-trans-Feruloyl-6′-*O*-β-D-apiofuranosylvanilloloside (*3*)

White powder; $[\alpha]_D^{26}$ − 45 (*c* 0.3, MeOH); UV λ_{max}^{MeOH}nm (log ε): 220sh (4.26), 230sh (4.19), 285 (3.92), 297sh (3.91), 327 (4.09); IR $\nu_{max}^{KBr}$$cm^{-1}$: 3421, 1716, 1595, 1516; For ^1H and ^{13}C NMR spectroscopic data, see: Table 1; NOESY: H-3 and H-7, OMe; H-5 and H-7; H-6 and H-1′; H-6′ and H-1″; H-6‴ and H-7‴; OMe and H-2‴; HMBC: H-3 to C- 5; H-5 to C-3, 7; H-6 to C-4; H-7 to C-3, 4, 5; H-1′ to C-1; H-1″ to C-6′; H- 5″ to CO; H-2‴ to C-3‴, 6‴; H-5‴ to C-1‴, 3‴; H-6‴ to C-2‴; H-7‴ to C-1‴, 2‴, 6‴, 8‴, CO; H-8‴ to C-1‴, CO; OMe to C-3‴; negative ion SIMS *m*/*z* 623 [M-H]$^-$, 153; negative ion HRSIMS *m*/*z* 623.1979 (calcd for $C_{29}H_{35}O_{15}$, 623.1977).

4.7. 7-*O*-trans-Caffeoylvanilloloside (*4*)

White powder; $[\alpha]_D^{25}$ − 39 (*c* 0.7, MeOH); UV λ_{max}^{MeOH}nm (log ε): 249sh (3.98), 285sh (4.02), 300sh (4.07), 329 (4.16); IR $\nu_{max}^{KBr}$$cm^{-1}$: 3421, 1697, 1601, 1516; For ^1H and ^{13}C NMR spectroscopic data, see: Table 2; NOESY: H-3, 5 and H-7; OMe and H-3; H-6 and H-1′; HMBC: H-3 to C- 1, 2, 7; H-5 to C-1, 3, 7; H-6 to C-1, 2; H-7 to C-3, 4, 5, 9″; H-2″ to C-4″, 7″; H-5″ to C-3″, 4″; H- 6″ to C-2″, 4″, 7″; H-7″ to C-2″, 6″, 8″, 9″; H-8″ to C-1″, 9″; OMe to C-2, H-1′ to C-1; negative ion SIMS *m*/*z* 477 [M-H]$^-$, 179; negative ion HRSIMS *m*/*z* 477.1387 (calcd for $C_{23}H_{25}O_{11}$, 477.1398).

4.8. 4″-*O*-β-D-Glucopyranosyl-caffeoylcalleryanin (*5*)

White powder; $[\alpha]_D^{24}$ − 83 (*c* 1.0, MeOH); UV λ_{max}^{MeOH}nm (log ε): 244sh (4.04), 286 (4.20), 320 (4.11); IR $\nu_{max}^{KBr}$$cm^{-1}$: 3389, 1697, 1635, 1508; For ^1H and ^{13}C NMR spectroscopic data, see: Table 2; NOESY: H-3, 5 and H-7; H-6 and H-1′; H-5″ and H-1‴; HMBC: H-3 to C- 1, 5, 7; H-5 to C-1, 3, 6, 7; H-6 to C-1, 4; H-7 to C-3, 4, 5, 9″; H-2″ to C-4″, 7″; H-5″ to C-1″, 6″; H- 6″ to C-2″, 4″, 5″, 7″; H-7″ to C-1″, 2″, 6″, 8″, 9″; H-8″ to C-1″, 9″; H-1′ to C-1; H-1‴ to C-4″; negative ion SIMS *m*/*z* 625 [M-H]$^-$, 463; negative ion HRSIMS *m*/*z* 625.1777 (calcd for $C_{28}H_{33}O_{16}$, 625.1770).

4.9. Axillaroside (*6*)

White powder; $[\alpha]_D^{25}$ − 60 (*c* 0.6, MeOH); UV λ_{max}^{MeOH}nm (log ε): 234 (4.24); IR $\nu_{max}^{KBr}$$cm^{-1}$: 3421, 1697, 1636; ^1H

NMR (CD_3OD) δ 1.07 (1H, *d*, $J = 6.5$ Hz, H-10), 1.74 (1H, *ddd*, $J = 14.5$, 8.0, 5.0 Hz, H-6), 2.07 (1H, *td*, $J = 9.0$, 5.0 Hz, H-9), 2.10–2.18 (1H, *m*, H-8), 2.29 (1H, *ddd*, $J = 14.5$, 7.5, 1.5 Hz, H-6), 2.30 (1H, *dd*, $J = 16.5$, 9.0 Hz, H-6″), 2.81 (1H, *br dt*, $J = 9.0$, 5.0 Hz, H-9″), 2.91 (1H, *dd*, $J = 16.5$, 4.0 Hz, H-6″), 3.11 (1H, *br q*, $J = 8.0$ Hz, H-5), 3.20, 3.22 (2H, each *dd*, $J = 9.0$, 8.0 Hz, H-2′, 2‴), 3.26-3.38 (2H, *m*, H-5′, 5‴), 3.27, 3.28 (2 H, each *br t*, $J = 9.0$ Hz, H-4′, 4‴), 3.28-3.38 (1 H, *m*, H-5″), 3.36, 3.37 (2H, each *t*, $J = 9.0$ Hz, H-3′, 3‴), 3.66, 3.67 (2H, each *dd*, $J = 12.0$, 6.0 Hz, H-6′, 6‴), 3.69 (3H, *s*, OMe), 3.89, 3.90 (2H, each *dd*, $J = 12.0$, 2.0 Hz, H-6′, 6‴), 4.66 (1H, *d*, $J = 8.0$ Hz, H-1′), 4.68 (1H, *d*, $J = 8.0$ Hz, H-1‴), 5.20 (1H, *br t*, $J = 5.5$ Hz, H-7), 5.23 (1H, *dd*, $J = 10.0$, 1.5 Hz, H-10″), 5.27 (1H, *dd*, $J = 17.0$, 1.0 Hz, H-10″), 5.28 (1H, *d*, $J = 5.0$ Hz, H-1), 5.50 (1H, *d*, $J = 4.5$ Hz, H-1″), 5.66 (1H, *br dt*, $J = 17.0$, 10.0 Hz, H-8″), 7.24 (1H, *d*, $J = 1.5$ Hz, H-3), 7.49 (1H, *d*, $J = 2.0$ Hz, H-3″); ^{13}C NMR (CD_3OD) δ 13.9 (C-10), 29.1 (C-5″), 32.7 (C-5), 35.3 (C-6″), 40.4 (C-6), 41.4 (C-8), 45.4 (C-9″), 47.2 (C-9), 51.8 (OMe), 62.8 (C-6′, 6‴), 71.6, 71.7 (C-4′, 4‴), 74.7, 74.8 (C-2′, 2‴), 78.1 (C-3′, 3‴), 78.3 (C-7), 78.5 (C-5′, 5‴), 97.6 (C-1), 97.7 (C-1″), 100.1 (C-1‴), 100.3 (C-1′), 110.5 (C-4″), 113.3 (C-4), 120.6 (C-10″), 134.5 (C-8″), 152.7 (C-3), 153.7 (C-3″), 168.0 (C-11″), 169.5 (C-11), 176.1 (C-7″); HMBC: H-1 to C-3, 5; H-3 to C-1, 4, 5, 11; H-5 to C-6, 9, 11; H-6 to C-5; H-7 to C-5, 9, 11″; H-8 to C-9; H-9 to C-8, 10; H-10 to C-8, 9; H-1″ to C-3″, 5″; H-3″ to C-1″, 4″, 5″, 11″; H-5″ to C-7″, 11″; H-6″ to C-4″, 5″, 7″, 9″; H-9″ to C-4″, 5″, 8″; H-10″ to C-9″; OMe to C-11; H-1′ to C-1; H-1‴ to C-1″; negative ion SIMS *m/z* 761 $[M–H]^-$, 599, 389; negative ion HRSIMS *m/z* 761.2503 (calcd for $C_{33}H_{45}O_{20}$, 761.2506).

4.10. Acid hydrolysis of compounds **1-6**

Each compound (1 mg) was heated at 95 °C with dioxane (0.5 mL) and 5% H_2SO_4 (0.5 mL) for 1 h. After neutralization with Amberlite IRA-400 (OH^- form), each reaction mixture was concentrated and the residue was passed through a Sep-Pak C_{18} cartridge with H_2O. The eluate was concentrated and the residue was treated with L-cysteine methyl ester hydrochloride (1 mg) in pyridine (0.125 mL) at 60 °C for 1 h. The solution was then treated with *N,O*-bis(trimethylsilyl)trifluoroacetamide (0.05 mL) at 60 °C for 1 h. The supernatant was applied to GLC; GLC conditions: column, Supelco SPB™-1, 30 m × 0.25 mm; column temperature, 230 °C; N_2 flow rate, 0.8 mL/min; t_R of derivatives, D-glucose 12.9 min, L-glucose 13.5 min, D-apiose 7.2 min. D-Glucose and D-apiose were detected from **1**–**3** and D-glucose was detected from **4**–**6**.

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