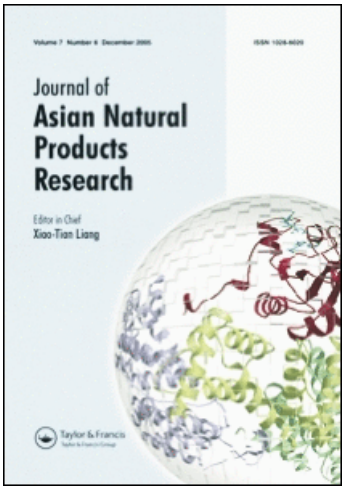

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Two new prenylated coumarins from *Spiranthes sinensis* (Pers.) Ames

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Two new prenylated coumarins, sinensins A and B, have been isolated from the roots of *Spiranthes sinensis* (Pers.) Ames. Their structures were elucidated as 5-γ,γ-dimethylallyl-8-[2-(2,6-dihydroxyphenyl)-3-dimethyl-but-2-enyol]-umbelliferon (**1**) and 4,6-di(γ,γ-dimethylallyl)-8-lavandulyl-umbelliferon (**2**) on the basis of spectroscopic analysis.

Keywords: *Spiranthes sinensis* (Pers.) Ames; prenylated coumarins; sinensin A; sinensin B; umbelliferon

1. Introduction

Spiranthes sinensis (Pers.) Ames, a famous traditional Chinese medicine, is widely used for the treatment of cancer,[1] bacterial and inflammatory diseases.[2] Previous investigations on *S. sinensis* have yielded homocyclotriucallane and dihydrophenanthrenes.[3–5] In our recent study, two new prenylated coumarins (figure 1), namely 5-γ,γ-dimethylallyl-8-[2-(2,6-dihydroxyphenyl)-3-dimethyl-but-2-enyol]-umbelliferon (**1**) and 4,6-di(γ,γ-dimethylallyl)-8-lavandulyl-umbelliferon (**2**), were isolated from 95% aqueous ethanolic extract of this plant by high-speed counter-current chromatography (HSCCC). The aim of this paper is to report the isolation and structure elucidation of two new compounds from the roots of *S. sinensis*.

2. Results and discussion

Compound **1**, a white amorphous powder, was assigned the molecular formula $C_{25}H_{24}O_6$ from $[M]^+$ peak at *m/z* 420.1545 in the HREI-MS, which was compatible with the results of ESI-MS (*m/z* 419.1 $[M - H]^-$; 421.2 $[M + H]^+$) and NMR analysis. The UV spectrum exhibited absorption maxima at 241, 267 and 337 nm. The IR spectrum of **1** showed the presence of hydroxyl (3461 cm^{-1}), carbonyl (1735 cm^{-1}), and aromatic ring (1613, 1508 and 1488 cm^{-1}). In the ^{13}C NMR and DEPT spectra of **1**, 25 carbon signals including four methyls, one methylene, seven methines and 13 quaternary carbons were observed. The umbelliferon skeleton of **1** was deduced from ^{13}C NMR signals at δ 160.9 (C-2), 113.6 (C-3), 144.1 (C-4), 143.6 (C-5), 112.2 (C-6), 162.5 (C-7), 114.6 (C-8), 123.5 (C-9), 149.6 (C-10), and ^1H NMR signal at δ 12.18 was characteristic of 7-OH. The proton signals at δ 3.11 (2H, d, $J = 8.0$ Hz, H-1‴), 5.14 (1H, m, H-2‴), 1.61 (3H, s, H-4‴), 1.67 (3H, s, H-5‴) and the corresponding carbon signals at δ 24.1 (C-1‴), 122.8 (C-2‴), 132.4 (C-3‴), 18.7 (C-4‴), 24.5 (C-5‴) were assigned to one γ,γ-dimethylallyl group, which was linked to C-5 based on the correlations of one set of doublet proton signals at δ 3.11 (2H, d, $J = 8.0$ Hz, H-1‴) with C-6 at δ 112.2 and C-9 at δ 123.5 in HMBC experiment. Through the ^{13}C NMR signals at δ 112.6 (C-1″), 157.7 (C-2″, 6″), 106.8 (C-3″, 5″), 131.1 (C-4″), and ^1H NMR signals at δ 6.37 (2H, d, $J = 8.0$ Hz, H-3″, 5″) and 6.97 (1H, t, $J = 8.0$ Hz, H-4″), one 2,6-dihydroxybenzyl was unambiguously existed in **1**, which was linked to C-2′ by the $^1H-^{13}C$ long-range correlations between one set of doublet proton signals at δ 6.37 (2H, d, $J = 8.0$ Hz, H-3″, 5″) with the carbon signal at δ 133.8 (C-2′). The ^{13}C NMR signals at δ 192.6 (C-1′), 133.8 (C-2′), 136.5 (C-3′), 17.6 (C-4′), 18.9 (C-5′), and the ^1H NMR signals at δ 1.72 (3H, s, H-4′), 1.86 (3H, s, H-5′) were attributed to the 3-dimethyl-but-2-enoyl group, the attachment position of which was established unambiguously at C-8. Thus the structure of **1** was determined to be 5-γ,γ-dimethyl-allyl-8-[2-(2,6-dihydroxyphenyl)-3-dimethyl-but-2-enyol]-umbelliferon (Figure 1), named sinensin A. All ^1H NMR and ^{13}C NMR assignments (shown in Table 1) for compound **1** were performed by $^1H-^1H$ COSY, HMQC and HMBC experiments (key HMBC and NOESY correlations are shown in Figure 2).

Compound **2** was obtained as a light yellow powder. The molecular formula of $C_{29}H_{38}O_3$ was determined by HREI-MS (*m/z* 434.2846, $[M]^+$), which was compatible with the results of ESI-MS (*m/z* 433.1 $[M - H]^-$; 867.2 $[2M - H]^-$; 457.1 $[M + Na]^+$) and NMR analysis. UV spectrum showed absorption maxima at 238, 275 and 342 nm. The IR spectrum of **2** indicated the presence of

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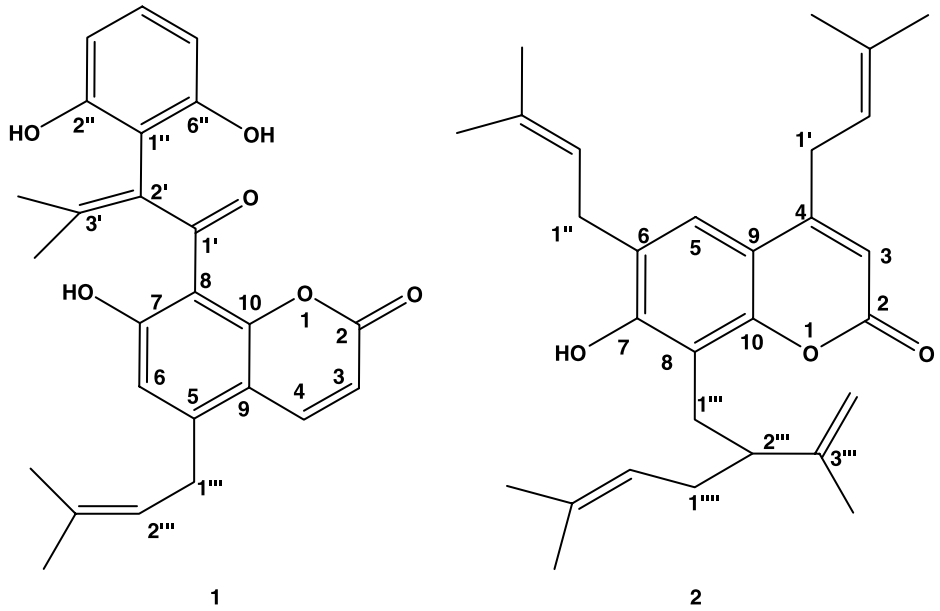

Figure 1. Structures of compounds **1** and **2**.

hydroxyl group (3432 cm^{-1}), carbonyl (1741 cm^{-1}), and aromatic ring (1624, 1538, 1478 cm^{-1}) in its structure. The ^{13}C NMR and DEPT spectra showed seven methyls, five methylenes, six methines and 11 quaternary carbons, and compound **2** had an umbelliferon skeleton, similar to compound **1**, according to the ^{13}C NMR and ^{1}H NMR spectral data. Two γ,γ-dimethylallyl groups were deduced by the carbon signals at δ 23.6 (C-1′), 122.4 (C-2′), 132.1 (C-3′), 19.1 (C-4′), 25.1 (C-5′), 22.2 (C-1″), 121.8 (C-2″), 131.8 (C-3″), 18.8 (C-4″), 24.7 (C-5″), and proton signals at δ 3.08 (2H, d, $J = 8.0$ Hz, H-1′), 5.11 (1H, m, H-2′), 1.54 (3H, s, H-4′), 1.46 (3H, s, H-5′), 3.11 (2H, d, $J = 8.0$ Hz, H-1″), 5.17 (1H, m, H-2″), 1.59 (3H, s, H-4″), 1.51 (3H, s, H-5″). A lavandulyl group was deduced by the ^{1}H NMR signals at δ 2.49 (2H, m, H-1‴), 2.41 (1H, m, H-2‴), 4.64 (1H, s, H-4‴), 4.57 (1H, s, H-4‴), 1.42 (3H, s, H-5‴), 1.88 (2H, m, H-1⁗), 5.08 (1H, brs, H-2⁗), 1.36 (3H, s, H-4⁗), 1.42 (3H, s, H-5⁗), and ^{13}C NMR signals at δ 25.6 (C-1‴), 42.2 (C-2‴), 144.7 (C-3‴), 110.4 (C-4‴), 21.2 (C-5‴), 30.2 (C-1⁗), 125.8 (C-2⁗), 130.4 (C-3⁗), 20.1 (C-4⁗), 27.6 (C-5⁗). These data suggested that compound **2** has an umbelliferon skeleton with two γ,γ-dimethylallyl and one lavandulyl groups. The attachment positions of these groups were established unambiguously at C-4 (γ,γ-dimethylallyl group), C-6 (γ,γ-dimethylallyl group), and C-8 (lavandulyl group), by the $^{1}H-^{13}C$ long-range correlations between H-1′ (δ 3.08) with C-3 (δ 113.6) and C-9 (δ 113.1), H-1″ (δ 3.11) with C-5 (δ 126.4) and C-7 (δ 153.2), and H-1‴ (δ 2.49) with C-7 (δ 153.2) and C-10 (δ 147.9), respectively, which were confirmed by the DEPT, $^{1}H-^{1}H$ COSY and HMQC experiments. All ^{1}H NMR and ^{13}C NMR assignments (shown in Table 2) for compound **2** were performed by $^{1}H-^{1}H$ COSY, HMQC and HMBC

Table 1. ^{1}H NMR (500 MHz) and ^{13}C NMR (125 MHz) spectral data of compound **1** in DMSO-d_6.

Position	δ_C	δ_H (J in Hz)	Position	δ_C	δ_H (J in Hz)
2	160.9	–	3′	136.5	–
3	113.6	5.72 *d* (2.0)	4′, 5′	17.6, 18.9	1.72 *s*, 1.86 *s*
4	144.1	6.84 *d* (2.0)	1″	112.6	–
5	143.6	–	2″, 6″	157.7	–
6	112.2	6.12 *s*	3″, 5″	106.8	6.37 *d* (8.0)
7	162.5	12.18 *brs*	4″	131.1	6.97 *t* (8.0)
8	114.6	–	1‴	24.1	3.11 *d* (2H, 8.0)
9	123.5	–	2‴	122.8	5.14 *m*
10	149.6	–	3‴	132.4	–
1′	192.6	–	4‴	18.7	1.61 *s*
2′	133.8	–	5‴	24.5	1.67 *s*

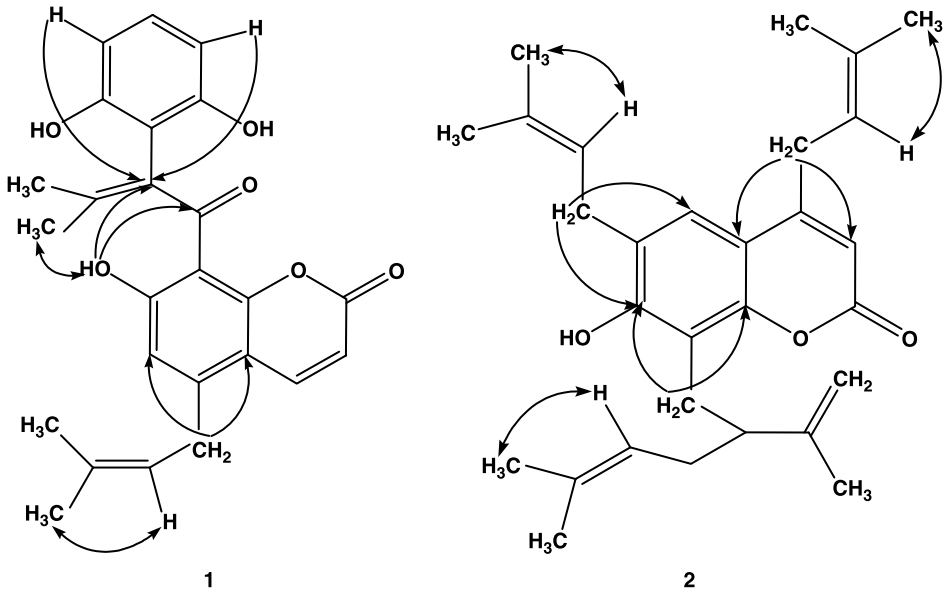

Figure 2. Key HMBC and NOESY correlations of compounds **1** and **2**.

experiments (key HMBC and NOESY correlations are shown in Figure 2). Thus the structure of **2** was determined to be 4,6-di(γ,γ-dimethylallyl)-8-lavandulyl-umbelliferon (Figure 1), named sinensin B.

3. Experimental

3.1 General experimental procedures

UV spectra were obtained with a Shimadzu UV 210A UV–Vis recording spectrophotometer. IR spectra were recorded on a Hitachi 275-50 spectrophotometer. The analytical HPLC was performed on 1200 HPLC series (Agilent, USA) with G-1311A quaternary pump, G1329A autosampler, G1314B UV–Vis detector using a Lichrospher C_{18} (150 × 4.6 mm) column, and the preparative chromatography system was performed by a TBE-300A HSCCC (Shenzhen, Tauto Biotech, China) composed of an *S* constant-flow pump (Beijing Boyikang Lab Implement, Beijing, China), a model 8823B UV detector, and a model N2000 workstation (Zhejiang University, Hangzhou, China). Column chromatography was carried out on D_{101} macroporous resin (Chemical Plant of Nankai University, Tianjin, China). NMR spectra were run on a Bruker AVANCE 500 NMR spectrometer (500 MHz for ^{1}H, and 125 MHz for ^{13}C) with TMS as internal standard. ESI-MS spectra were performed on API 3200 mass spectrometer (USA) and HREI-MS spectra were obtained on a Finnigan MAT 711 mass spectrometer.

Table 2. ^{1}H NMR (500 MHz) and ^{13}C NMR (125 MHz) spectral data of compound **2** in DMSO-d_6.

Position	δ_C	δ_H (J in Hz)	Position	δ_C	δ_H (J in Hz)
2	161.5	–	2″	121.8	5.17 *m*
3	113.6	5.93 *s*	3″	131.8	–
4	156.1	–	4″	18.8	1.59 *s*
5	126.4	6.25 *s*	5″	24.7	1.51 *s*
6	125.6	–	1‴	25.6	2.49 *m*
7	153.2	11.56 *brs*	2‴	42.2	2.41 *m*
8	118.8	–	3‴	144.7	–
9	113.1	–	4‴	110.4	4.64 *s* 4.57 *s*
10	147.9	–	5‴	21.2	1.42 *s*
1′	23.6	3.08 *d* (2H, 8.0)	1⁗	30.2	1.88 *m*
2′	122.4	5.11 *m*	2⁗	125.8	5.08 *brs*
3′	132.1	–	3⁗	134.0	–
4′	19.1	1.54 *s*	4⁗	20.1	1.36 *s*
5′	25.1	1.46 *s*	5⁗	27.6	1.42 *s*
1″	22.2	3.11 *d* (2H, 8.0)			

3.2 Plant material

The roots of *Spiranthes sinensis* (Pers.) Ames were purchased in August 2006 from a local drug store, Dalian, Liaoning, China. A voucher specimen (DLMU, PLC0601), identified by Professor Dan Yuan, College of Traditional Chinese Materia Medica, Shenyang Pharmaceutical University, is deposited in the Herbarium of the School of Pharmacy, Dalian Medicinal University, Dalian, China.

3.3 Extraction and isolation

Before extraction, the roots of *S. sinensis* were ground into powder, and 5.0 kg powders were weighed and refluxed with 95% aqueous ethanol two times. After evaporation of EtOH *in vacuo*, the residue (466 g) was diluted in water and subjected to a glass column (6.0 × 100 cm, containing 4.0 kg D_{101} macroporous resin). Water (8000 ml), 40% aqueous ethanol (8000 ml), 60% aqueous ethanol (10,000 ml), and 90% aqueous ethanol (6000 ml) were used to elute the column in series, 90% ethanol eluent was collected and evaporated to dryness under reduced pressure at 50°C, and 6.4 g yellow powder was obtained, which was directly subjected to HSCCC separation. Compounds **1** (25 mg) and **2** (38 mg) were isolated from the crude sample (400 mg) only in one run under optimised HSCCC conditions using *n*-hexane/ethyl acetate/methanol/water (8:11:11:8, v/v) as the separation solvent system.

3.3.1 Sinensin A (*1*)

White amorphous powder; UV (MeOH) λ_{max} (nm): 241, 267, 337; IR (KBr) ν_{max} (cm^{-1}): 3461 (OH), 1735 (C=O), 1613, 1508, 1488, 1192, 935; ^1H NMR and ^{13}C NMR spectral data: see Table 1; ESI-MS *m/z*: 419.1 $[M - H]^-$, 421.2 $[M + H]^+$; HREI-MS *m/z* 420.1545 $[M]^+$ (calcd for $C_{25}H_{24}O_6$, 420.1573).

3.3.2 Sinensin B (*2*)

Light yellow powder; UV (MeOH) λ_{max} (nm): 238, 275, 342; IR (KBr) ν_{max} (cm^{-1}): 3432 (OH), 1741 (C=O), 1624, 1538, 1478, 1025, 846; ^1H NMR and ^{13}C NMR spectral data: see Table 1; ESI-MS *m/z*: 433.1 $[M - H]^-$, 867.2 $[2M - H]^-$, 457.1 $[M + Na]^+$; HREI-MS *m/z* 434.2846 $[M]^+$ (calcd for $C_{29}H_{38}O_3$, 434.2821).

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References

[1] W.L. Li. *J. Math. Med.* **18**, 255 (2005).
[2] E.N. Matu and J. Van Staden. *J. Ethnopharmacol* **87** 35 (2003).
[3] K. Tezuka, K. Nagashima, and H. Hirano. *Chem. Pharm. Bull.* **37**, 1667 (1989).
[4] Y.L. Liu, W.Y. Wang, Y.H. Kuo, and Y.H. Liu. *Chem. Pharm. Bull.* **49**, 1098 (2001).
[5] Y.L. Liu, R.L. Huang, M.J. Don, and Y.H. Kuo. *J. Nat. Prod.* **632**, 1608 (2000).