

Communication

Two New Constituents from *Artemisia capillaris* Thunb.

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Abstract: Two new compounds, 6'-O-caffeoyl-*p*-hydroxyacetophenone-4-*O*- β -D-glucopyranoside (**1**) and 6-amino-9-[1-(3,4-dihydroxyphenyl)ethyl]-9*H*-purine (**2**) were isolated from the aerial parts of *Artemisia capillaris* Thunb. The structures were established on the basis of spectral data.

Keywords: *Artemisia capillaris*; *p*-hydroxyacetophenone; adenine

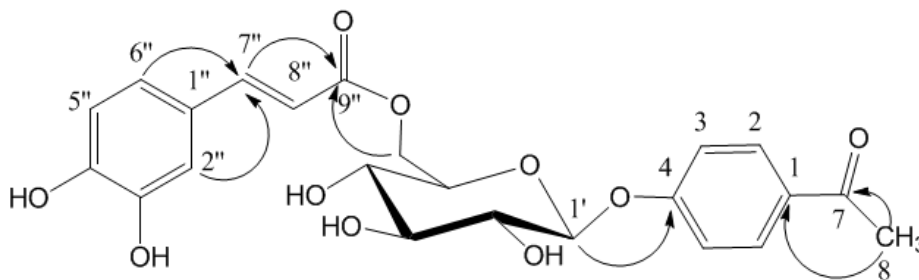
Introduction

Infusions of the buds, stems and leaves of *Artemisia capillaris* (Yin Chen Hao, Capillary or Oriental Wormwood) have been used in Chinese Traditional Medicine since antiquity as a cholagogic, antipyretic, anti-inflammatory and diuretic purposes and in the treatment of jaundice [1-6]. Coumarins, flavonol glycosides and a group of unidentified aglycones have been reported so far from the inflorescence of *Artemisia capillaris* [7,8]. Our investigation of the aerial parts of this plant has led to the isolation of two new constituents. This paper deals with the isolation and structural determination of these compounds.

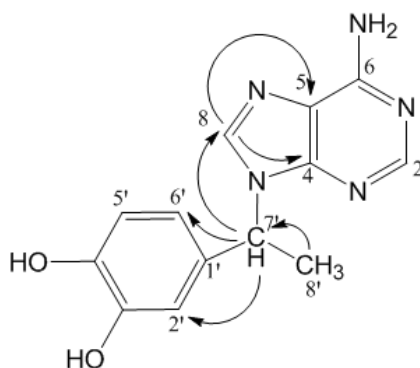
Results and Discussion

Compound **1**, a yellow powder, was formulated as $C_{23}H_{24}O_{10}$ on the basis of HR-FAB-MS data (m/z 461.1436, $[M+H]^+$, calcd. 461.1446). Its 1H -NMR spectrum indicated one methyl (δ 2.38, s), a 1,4-disubstituted benzene ring [δ 7.85 (2H, d, $J = 8.7$ Hz) and δ 7.10 (2H, d, $J = 8.7$ Hz)] and a 1,2,4-trisubstituted benzene ring [δ 6.76 (1H, d, $J = 8.1$ Hz), δ 6.96 (1H, d, $J = 8.1$ Hz) and δ 7.05 (1H, s)]. In addition, the coupling constants of the proton signals at δ 7.46 and δ 6.25 were 15.8 Hz, indicating the presence of two *trans*-olefinic protons. Thus, it was presumed that **1** contained an *O*-caffeoyl group [9,10]. The 1H -NMR and ^{13}C -NMR spectra also indicated the presence of a glucopyranosyl unit [δ_H 5.07 (anomeric proton), δ_C 99.6, 76.4, 74.0, 73.2, 70.1, 63.3]. The coupling constant of the anomeric proton signal ($J = 9.0$ Hz) indicated the β -configuration of the glucose. Long-range correlations were observed in the heteronuclear multiple-bond connectivity (HMBC) spectrum as follows: (a) δ_H 7.05 and δ_C olefinic carbon 145.8, δ_H 6.96 and δ_C 145.8, δ_H 7.46 and ester carbonyl carbon δ_C 166.4 which further evidenced the presence of *O*-caffeoyl group; (b) methyl proton at δ_H 2.30 and δ_C 196.4, δ_H 2.30 and 130.9 indicating that the methyl and 1,4-disubstituted benzene ring was connected by a carbonyl carbon [4]; (c) anomeric proton of glucose δ_H 5.07 and δ_C 160.9, H-6' of glucose and ester carbonyl carbon δ_C 166.4 showed that the *O*-caffeoyl group and 1,4-disubstituted benzene ring must be linked at the position C-6' and C-1' of the glucose, respectively. Acid hydrolysis of **1** furnished the sugar component, which was confirmed as glucose by TLC comparison with an authentic sample. Thus, compound **1** was established to be 6'-*O*-caffeoyl-*p*-hydroxyacetophenone-4-*O*- β -D-glucopyranoside (Figure 1).

Figure 1. Important HMBC correlations of **1**.



Compound **2** was obtained as an amorphous powder. The molecular formula was established to be $C_{13}H_{13}N_5O_2$ based on HR-FAB-MS data (m/z 272.1136, $[M+H]^+$, calcd for $C_{13}H_{14}N_5O_2$: 272.1128). The signals in the 1H -NMR spectrum of compound **2** at δ 8.27 (s), 8.11 (s) and 7.19 (2H, s, NH_2) along with those in the ^{13}C -NMR spectrum at δ 118.9, 139.2, 149.2, 152.4 and 156.0 indicated the presence of an adenine ring. The 1H -NMR spectrum also showed a 1,2,4-trisubstituted benzene ring [δ 6.67, 1H, d, $J = 8.4$ Hz), δ 6.64 (1H, d, $J = 8.4$ Hz) and δ 6.69 (1H, s)] and a methyl which was connected to a methine [δ 1.85, d, $J = 7.2$ Hz), δ 5.64 (q, $J = 7.2$ Hz)]. In its HMBC spectrum long-range correlations were observed between δ_H 5.64 and δ_C 139.2 and δ_H 5.64 and δ_C 114.0, indicating that both the adenine ring and benzene ring were connected to the methine. In summary, based on HSQC and HMBC spectral data, compound **2** was determined to be 6-amino-9-[1-(3,4-dihydroxyphenyl)ethyl]-9*H*-purine (Figure 2).

Figure 2. Important HMBC correlations of **2**.

Experimental

General

The NMR data were obtained on a Bruker ARX-300 spectrometer (300 MHz for ^1H and 75 MHz for ^{13}C) in $\text{DMSO}-d_6$ with TMS as internal standard. The HR-FAB-MS data were obtained on the Micros Mass Autospec-UltimaE TOF mass spectrophotometer. Chromatography was performed on silica gel (200-300 mesh), Sephadex LH-20 and with a Shimadzu LC-8A HPLC instrument equipped with a reversed-phase column.

Plant material

The material investigated were aerial parts of *A. capillaris* purchased from the Cooperative of Traditional Chinese Medicine of Shenyang, P.R. China. A voucher specimen was identified by Prof. Qi-Shi Sun and deposited at the School of Traditional Chinese Medicine of Shenyang Pharmaceutical University, P.R.China.

Extraction, isolation and product characterization

The dry aerial parts (10 kg) of *A. capillaries* were extracted with boiling water three times, and then precipitated with 75% aqueous alcohol. After evaporation of the solvents under reduced pressure, the residue (1.1 kg) was suspended in H_2O and extracted sequentially with petroleum ether, ethyl acetate and *n*-butanol. The ethyl acetate extract (53 g) was subjected to silica gel CC with elution by CHCl_3 - CH_3OH in increasing polarity to obtain eight fractions (A to G). Fraction F was then purified by Sephadex LH-20 column chromatography eluted with CH_3OH , and further separated by preparative RP-HPLC eluted with 35% aqueous CH_3OH to give compounds **1** (20 mg) and **2** (8 mg). ^1H - and ^{13}C -NMR data are shown in Table 1.

Table 1. NMR data of compounds **1**, **2** (ppm from TMS, in DMSO- d_6).

Position	1		2	
	H	C	H	C
1		130.9		
2	7.84 (d, $J=8.7$ Hz)	130.2	8.11 (s)	152.4
3	7.10 (d, $J=8.7$ Hz)	116.0		
4		160.9		149.2
5	7.10 (d, $J=8.7$ Hz)	116.0		118.9
6	7.84 (d, $J=8.7$ Hz)	130.2		156.0
7		196.4		
8	2.38 (s)	26.3	8.27 (s)	139.2
1'	5.07 (d, $J=9.0$ Hz)	99.6		132.5
2'	3.30 (m)	73.2	6.69 (s)	114.0
3'	3.32 (m)	76.4		145.2
4'	3.20 (m)	70.1		149.2
5'	3.70 (m)	74.0	6.67 (d, $J=8.4$ Hz)	115.3
6'	4.41(d, $J=11$ Hz), 4.17 (dd, $J=11, 7.3$ Hz)	63.3	6.64 (d, $J=8.4$ Hz)	117.0
7'			5.64 (q, $J=7.2$ Hz)	52.0
8'			1.85 (d, $J=7.2$ Hz)	20.6
1''		125.0		
2''	7.05 (s)	114.1		
3''		145.2		
4''		148.8		
5''	6.76 (d, $J=8.1$ Hz)	115.2		
6''	6.96 (d, $J=8.1$ Hz)	121.0		
7''	7.46 (d, $J=15.8$ Hz)	145.8		
8''	6.25 (d, $J=15.8$ Hz)	113.0		
9''		166.4		

Acid Hydrolysis of **1** [12]

Compound **1** (8 mg) was refluxed with 10% HCl in 75% EtOH (3 mL) for 6 h. After cooling, the reaction mixture was extracted with EtOAc (3 ml). The water layers were concentrated and compared with *D*-glucose by TLC analysis [system 1: silica-gel, *n*-BuOH- C_5H_5N - H_2O (8.0:4.0:3.0), *R_f*: 0.24; system 2: silica-gel, EtOAc-MeOH- H_2O -AcOH (6.5:2.0:1.5:1.5), *R_f*: 0.42]. Those confirmed that the sugar moiety obtained from aqueous acid hydrolysis of compound **1** was *D*-glucose.

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Sample Availability: Available from the authors.