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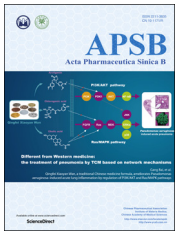

ORIGINAL ARTICLE

Two new phenylpropanoid glycosides from the aerial parts of *Lespedeza cuneata*

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KEY WORDS

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Cuneataside E;
Cuneataside F

Abstract Two new phenylpropanoid glycosides named cuneataside E (**1**) and cuneataside F (**2**), were isolated from the aerial parts of *Lespedeza cuneata* (Dum. Cours.) G. Don, whose structures were *E* and *Z* isomer, respectively. Their structures were elucidated on the basis of comprehensive spectroscopic analysis (UV, IR, HR-ESI-MS, 1D and 2D NMR). In *in vitro* bioassays at 10 μmol/L, compound **1** showed moderate hepatoprotective activity against *N*-acetyl-*p*-aminophenol (APAP)-induced toxicity in HeG2 cells.

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1. Introduction

Lespedeza cuneata (Dum. Cours.) G. Don, an annual herbaceous plant, is distributed in China, Korea, India, Australia and USA[1], which named "ye guan men" in Chinese, is a very important traditional medicine, and has been used in the treatment of diabetes[2], hematuria, insomnia and malnutrition[3]. Previous phytochemical studies have revealed flavonoids, sterols, triterpenoids[4–6] and phenylpropanoid glycosides[7] as chemical constituents of the plant, which showed antioxidant effects[8–12], anti-inflammatory effects[13] and antibacterial avtivities[14]. Among them, flavonoids were the main components of *L. cuneata*. In our continuing effort in studying constituents from this important medicinal plant, two new phenylpropanoid glycosides (Fig. 1) were isolated. Their structures were elucidated by various spectroscopic methods (UV, IR, HR-ESI-MS, 1D and 2D NMR). The isolation and structural elucidation of the new compounds were described in this paper.

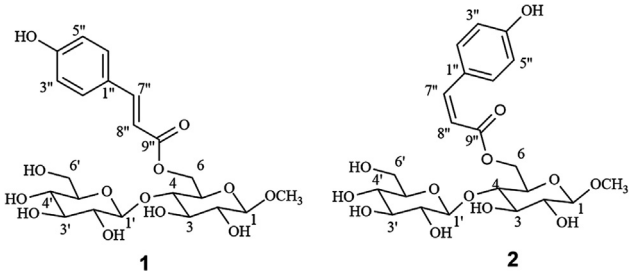

Figure 1 Structures of compounds **1** and **2**.

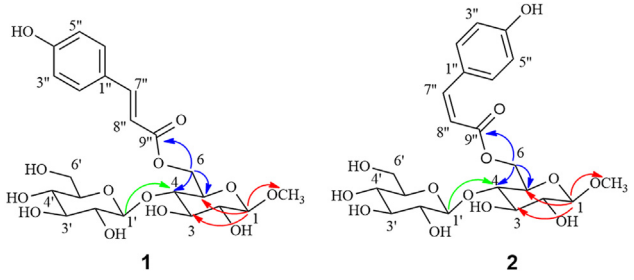

Figure 2 Key HMBC (arrows, from ^{1}H NMR to ^{13}C NMR) correlations of compounds **1** and **2**.

2. Results and discussion

Compound **1** was obtained as a white amorphous powder. The UV spectrum showed absorption maximums at 210, 228 and 314 nm. In the IR spectrum, absorption bands at 3375, 2901, 1604, 1515, and 1449 cm^{-1} were observed. These data indicated the presence of hydroxyl, benzene, and carbonyl groups in **1**. The molecular formula was determined to be $C_{22}H_{30}O_{13}$ on the basis of HR-ESI-MS *m/z* 525.1588 $[M+Na]^{+}$ (Calcd. for $C_{22}H_{30}O_{13}Na$ 525.1579). In the ^{1}H NMR spectrum of **1**, a set of AB-type signals at δ_H7.56 (2H, d, J=8.4 Hz, H-2″, 6″) and δ_H 6.57 (2H, d, J=8.4 Hz, H-3″, 5″) were observed, which suggested the existence of a 1,4-disubstituted benzene ring. Additionally, a methoxy signal at δ_H 3.35 (3H, s, OMe) and two anomeric proton signals at δ_H 4.25 (1H, d, J=8.0 Hz), δ_H 4.16 (1H, d, J=8.0 Hz) with large coupling constants suggested β-glucosidic linkages. From the hydrolysate of **1**, a neutral residue containing sugars was obtained by extraction and evaporation. The sugar residue and authentic D-glucose were separately allowed to react with L-cycteine methyl ester and *N*-trimethylsilylimidazole (Section 4.4). Subsequent GC analysis indicated that two sugar derivatives from the sugar residue had retention time (t_R) identical to that of authentic D-glucose. This verified that both glycosyl units in **1** possessed the D-configuration. We can also find *trans*-disubstituted double bond at δ_H 7.55 (1H, d, J=16.0 Hz) and δ_H 6.43 (1H, d, J=16.0 Hz), which suggests that the compound is *E* isomer. The ^{13}C NMR spectrum showed 22 carbon signals. An α,β-unsaturated carbonyl group was demonstrated at δ_C 166.5. These spectroscopic data indicates that **1** has a *trans-p*-coumaroyl and two β-glucopyranosyl groups, for which the structure was further elucidated by 2D NMR data analysis.

The proton-bearing carbon signals in the NMR spectra were assigned by cross-peaks in the HSQC spectra. HMBC correlations from H-1 to C-3, C-5, C–OCH_3; from H-6 to C-5, C-4, and C-9″ (Fig. 2); together with their chemical shifts, revealed the presence of a methoxy group at C-1 and a *trans-p*-coumaroyl group at C-6. In the NOE spectra (Fig. 3), an enhancement of the proton signal at the H-OCH_3/H-5 on irradiation of the H-1, and at the H-5 on irradiation of the H-6 revealed that H-OCH_3 and the coumaroyl groups are linked on the same glucopyranosyl moiety. The HMBC correlations from H-1′ to C-4 demonstrated that two β-glucopyranosyl groups were connected through a 1,4-linkage. Therefore, the structure of **1** was elucidated as methyl-6-*O*-[(*E*)-3-(4-hydroxyphenyl)prop-2-enoyl]-4-*O*-β-D-glucopyranoside-β-D-glucopyranoside, and named cuneataside E.

Compound **2** was obtained as a white powder, whose molecular formula was determined to be $C_{22}H_{30}O_{13}$ on the basis of HR-ESI-MS. The UV spectrum showed absorption maximums at 210, 227 and

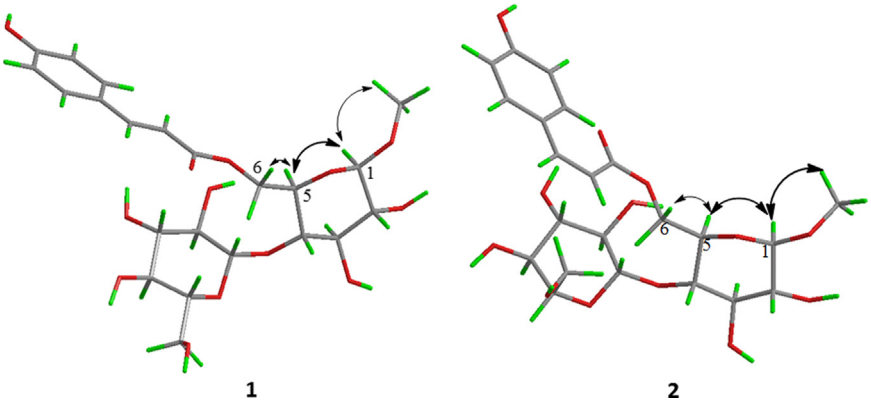

Figure 3 The NOE enhancements induced by irradiation of H-1 and H-6 for compounds **1** and **2**.

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