Tetrahedron Letters 56 (2015) 2163–2166

Contents lists available at ScienceDirect

Tetrahedron Letters

journal homepage: www.elsevier.com/locate/tetlet

Sweriyunnanlactone A, one unusual secoiridoid trimer from *Swertia yunnanensis*

Chang-An Geng, Xing-Long Chen, Xiao-Yan Huang, Yun-Bao Ma, Kang He, Ning-Jia Zhou, Tuan-Wu Cao, Xue-Mei Zhang, Ji-Jun Chen *

State Key Laboratory of Phytochemistry and Plant Resources in West China, Kunming Institute of Botany, Chinese Academy of Sciences, Kunming 650201, PR China

ARTICLE INFO

Article history:
Received 8 December 2014
Revised 10 March 2015
Accepted 13 March 2015
Available online 18 March 2015

Keywords:
Sweriyunnanlactone A
Secoiridoid trimer
Swertia yunnanensis
LC–MS guided isolation
Anti-HBV activity

ABSTRACT

One unusual secoiridoid trimer, namely sweriyunnanlactone A (**1**), was isolated from *Swertia yunnanensis* under the guidance of LC–MS analysis. Sweriyunnanlactone A with a phenyl ring (ring F) was the first example of secoiridoid trimer featuring a C_{28} skeleton. Its structure was determined by extensive HRESIMS, 1D and 2D NMR spectroscopic data, and GIAO ^{13}C NMR calculation. Compound **1** showed weak inhibition on HBV DNA replication with an IC_{50} value of 60.76 μM (SI = 12.6) on HepG 2.2.15 cell line in vitro.

Qing-Ye-Dan is a famous traditional Chinese herb used for treating hepatitis in Yunnan Province of China, which has been documented in every edition of Chinese Pharmacopoeia.[1] In order to clarify its anti-HBV constituents, our previous investigation on *Swertia mileensis* resulted in a series of novel lactones,[2–6] of which four secoiridoid trimers, namely swerilactones H–K, have aroused much interest in scientists due to their unprecedented architectures and promising activities.[7] Biogenetically, swerilactones H–K with a polycyclic C_{29} skeleton are polymerized by three molecules of secoiridoids (two C_{10} and one C_9). In addition to swerilactones H–K, no additional secoiridoid trimer has been reported. Therefore, it will be very interesting to demonstrate whether other types of secoiridoid trimers still exist in Qing-Ye-Dan related herbs from both biosynthetic and biological points of views. However, these compounds are always present as trace constituents in plants, and the low content brings great difficulties to classical isolation.

The Shimadzu UFLCMS-IT-TOF apparatus equipped with an electrospray ionization source coupled to ion-trap and time-of-flight mass analyzers (ESI–IT–TOF) which enables high-resolution mass spectra in both positive and negative modes simultaneously, is effective for characterizing trace components in the complex mixture of natural products.[8–12]

Swertia yunnanensis as the congener plant of *S. mileensis* is always used as the alternative of Qing-Ye-Dan for treating jaundice, hepatitis, and cholecystitis in Yunnan, Sichuan, and Guizhou Provinces of China. Our previous investigation on this plant gave rise to a series of xanthones, triterpenoids, secoiridoids, and flavonoids that were common in *Swertia* plants.[13,14]

The air-dried and powdered whole plant of *S. yunnanensis* (1.0 kg) was extracted with EtOH (10 L) at room temperature for 3 times. The combined EtOH extract was condensed in vacuo and partitioned between H_2O and EtOAc (2 L × 3). The EtOAc part (41 g) was separated by silica gel column chromatography (Si CC) eluted with $CHCl_3$–MeOH gradient to give six fractions, Frs. A1–A6. Fr. A3 was loaded on a RP-18 CC and eluted with a gradient of MeOH–H_2O system (15:85–90:10) to provide fractions A3-1 to A3-5. Fr. A3-2 was further purified with Sephadex LH-20 CC eluted with $CHCl_3$–MeOH (1:1) system to give three fractions A3-2-1 to A3-2-3. Fr. A-3-2-1 was analyzed by UFLC-MS-IT-TOF to recognize one chromatographic peak with the molecular formula of $C_{29}H_{30}O_8$ that was determined from the $[M+Na]^+$ ion (*m*/*z* 529.1843) in positive mode, and $[M+HCOO]^-$ (*m*/*z* 551.1923) and $[M-H]^-$ (*m*/*z* 505.1823) ions in negative mode. Consequently, this peak was purified by a semi-preparative HPLC apparatus on a Waters XTerra Prep RP-18 column (7.8 × 300 mm, 10 μm), eluted with acetonitrile–H_2O system (26:74, flow rate = 3.0 ml/min) to obtain the targeted compound **1** (2.5 mg, R_t = 19.5 min). Structurally, compound **1** featured a heptacyclic system which is the first example of

* Corresponding author. Tel.: +86 871 65223265; fax: +86 871 65227197.
E-mail address: chenjj@mail.kib.ac.cn (J.-J. Chen).

http://dx.doi.org/10.1016/j.tetlet.2015.03.057
0040-4039/

secoiridoid trimer with a C_{28} skeleton, and named as sweriyunnanlactone A (**1**) (Fig. 1).

Compound **1**[15] was isolated as white powder with the molecular formula of $C_{29}H_{30}O_8$, corresponding to 15 degrees of unsaturation. The IR spectrum indicated the presence of hydroxyl (3435 cm^{-1}), carbonyl (1717 cm^{-1}) groups, and an aromatic ring (1632, 1463 and 1408 cm^{-1}) in the structure. In accordance with its molecular formula, all the 29 carbons were well resolved in the ^{13}C NMR spectrum, which were recognized as three methyls (including one methoxy), six methylenes, eight methines, and twelve quaternary carbons by ^{13}C NMR (DEPT) and HSQC experiments. Two ester carbonyl groups (δ_C 161.0 and 164.3) and twelve olefinic carbons (from δ_C 155.6 to 122.5) were characterized in the down-field region of ^{13}C NMR spectrum, of which three groups of tri-substituted double bonds were revealed based on three singlets at δ_H 6.41 (1H, s, H-7), 7.67 (1H, s, H-11), and 7.33 (1H, s, H-13) in the ^1H NMR spectrum. One dioxygenated methine [δ_C 95.1 (d, C-25) and δ_H 5.19 (1H, s, H-25)] and one methoxy [δ_C 55.0 (q) and δ_H 3.39 (3H, s)] were deduced to be an acetal group by the obvious HMBC correlations from OMe to C-25 and from H-25 to OMe. In the ^1H NMR spectrum, two groups of doublets at δ_H 1.07 (3H, d, J = 6.2 Hz) and 1.16 (3H, d, J = 6.6 Hz) suggested that two methyls were respectively linked with a methine, which was confirmed by ^1H ^1H COSY correlations of H-31/H-19 and H-32/H-23. In addition, six methylenes involving three oxygenated ones were readily observed in the ^{13}C NMR spectrum, which was characteristic for the presence of δ-lactone moieties.[4] Based on the above analyses, compound **1** was supposed to be the analogue of secoiridoid trimer, with some similarity with the previously reported swerilactones H–K.[3]

Although, the framework of compound **1** has been tentatively proposed as a secoiridoid trimer, it is still not an easy work to totally construct its structure by spectroscopic analyses, due to the complicated construction of secoiridoid trimers. Fortunately, the partial structure **1a** (rings A–E) of compound **1** was clearly established to be the same with that of swerilactone H, based on their almost identical ^1H and ^{13}C NMR spectroscopic data, and detailed ^1H ^1H COSY, HMBC, and ROESY analyses (Fig. 2). Compared with swerilactone H,[3] the fragment **1c** of compound **1** was characterized with an additional methoxy group to form an acetal motif, which was confirmed by the down-field shift of C-25 (from δ_C 89.4 to 95.1) as well as the ^1H ^1H COSY (H-23/H-32 and H-27/H-28) and HMBC (H-32 with C-23 and C-24; H-23 with C-25 and C-29; H-25 with OMe, C-23, C-27 and C-29; and H-27 with C-29 and C-25) experiments. From the above analyses, the structural difference between compound **1** and swerilactone

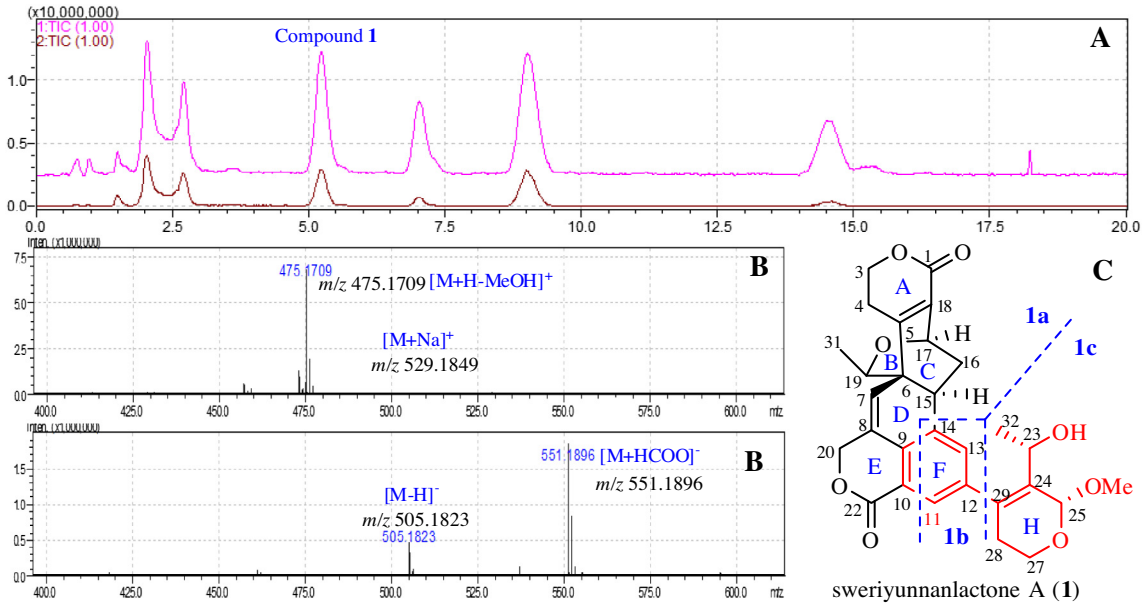

Figure 1. UFLC–MS profiles (A), (±) HRESIMS (B), and structure (C) of compound **1**.

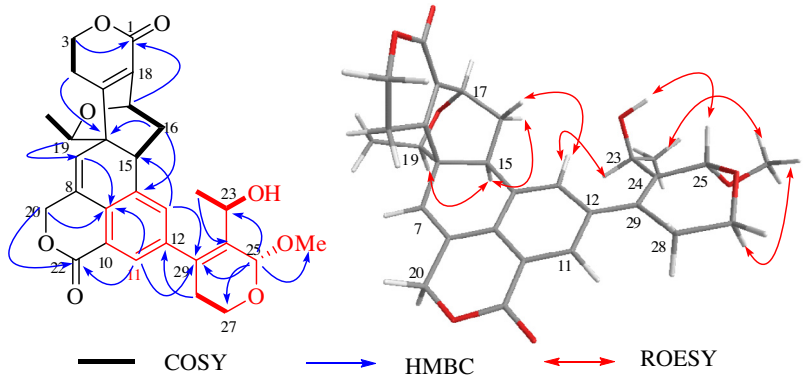

Figure 2. Key 2D correlations of compound **1**.