Available online at www.sciencedirect.com
ScienceDirect
Chinese Chemical Letters 21 (2010) 821–823
CHINESE CHEMICAL LETTERS
www.elsevier.com/locate/cclet

A new alkylene dihydrofuran glycoside with antioxidation activity from the root bark of *Morus alba* L.

Wei Fu, Yong Fang Lei, Ya Ling Cai, Dao Nian Zhou, Jin Lan Ruan *

Key Laboratory of Natural Medicinal Chemistry and Resource Evaluation of Hubei Province, College of Pharmacy, Tongji Medical Center, Huazhong University of Science and Technology, Wuhan 430030, China

Received 21 October 2009

Abstract

A new alkylene dihydrofuran glycoside (**1**) was isolated from the root bark of *Morus alba* L., along with moracin M-3′-*O*-β-D-glucopyranoside (**2**), and moracin M-6, 3′-di-*O*-β-D-glucopyranoside (**3**). Compound **1** was identified as 2-methylene-3-methoxy-2, 5-dihydrofuran-4-*O*-β-D-glucopyranoside on the basis of chemical and spectroscopic data including 1D and 2D NMR spectral analysis. In addition, the antioxidant activity of **1** was evaluated using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) and 2,2′-azinobis-3-ethylbenzothiazoline-6-sulphonic acid (ABTS) assay. The IC_{50} values were 2.49 and 0.45 mg/mL, respectively.

Keywords: *Morus alba* L.; Dihydrofuran glycoside; Spectral analysis; Antioxidation activity

The plant *Morus alba* L. is widely distributed in eastern countries and mainly found in China, Japan and Korea [1]. Its root bark ('Sang Bai Pi' in China) has been used as a traditional Chinese medicine for the treatment of lung-heat, cough, hematemesis, dropsy, and dysuria [2]. As a part of an investigation on *Morus* species, we examined the root bark of *M. alba* L., and obtained a new dihydrofuran glycoside (**1**), along with moracin M-3′-*O*-β-D-glucopyranoside (**2**), and moracin M-6, 3′-di-*O*-β-D-glucopyranoside (**3**). In this paper, we report the isolation, structure elucidation and radical scavenging abilities of compound **1** on 1,1-diphenyl-2-picrylhydrazyl (DPPH) and 2,2′-azinobis-3-ethylbenzothiazoline-6-sulphonic acid (ABTS) radicals.

The root bark of *M. alba* L. were collected at Kaili city, Guizhou Province, China, in December 2008. The plant was identified by Prof. Changgong Zhang at the College of Pharmacy, Tongji Medical Center, Huazhong University of Science and Technology. A voucher specimen (No. SBP081201) has been deposited at the Herbarium of the College of Pharmacy, Tongji Medical Center, Huazhong University of Science and Technology, Wuhan, China.

The dried root bark (3.7 kg) was extracted with 80% ethanol under reflux. The resulting extracts were combined and concentrated under reduced pressure to obtain a residue (620 g). The residue was suspended in water and partitioned with petroleum ether, EtOAc, and *n*-BuOH, successively. The *n*-BuOH fraction (85.4 g) was separated over silica gel column gradiently eluted with $CHCl_3$–MeOH (40:1 to 1:1) to give 10 fractions (fractions 1–10), fraction 4 (1.84 g) was separated further over silica gel column ($CHCl_3$:MeOH:H_2O, 5:1:0.1) and Sephadex LH-20 column (MeOH:H_2O, 1:1)

* Corresponding author.
E-mail address: jinlan8152@163.com (J.L. Ruan).

1001-8417/$ – see front matter
doi:10.1016/j.cclet.2010.02.016

Table 1
NMR spectral data of compound **1** in DMSO-d_6 (^{1}H: 400 MHz, ^{13}C: 100 MHz, δ ppm, J in Hz).

Position	δ_H	δ_C
2		154.4
3		132.8
4		153.5
5	3.72 (s, 2H)	56.2
6	6.37 (s, 2H)	94.8
1′	4.76 (d, 1H, J = 8.0)	101.5
2′	3.1–3.5 (m, 1H)	73.7
3′	3.1–3.5 (m, 1H)	77.7
4′	3.1–3.5 (m, 1H)	70.5
5′	3.1–3.5 (m, 1H)	77.3
6′	4.40 (dd, 1H, J = 12.0, 2.0) 4.22 (dd, 1H, J = 12.0, 6.4)	61.3
3-OMe	3.57 (s, 3H)	60.6

to yield compound **1** (115 mg), The separation of fraction 8 (1.63 g) was chromatographed on silica gel column ($CHCl_3$:MeOH:H_2O, 3:1:0.1), and Sephadex LH-20 column ($CHCl_3$:MeOH, 1:1) to yield **2** (38 mg) and **3** (45 mg).

Compounds **1** was obtained as yellow powder, mp 188–190 °C, $[\alpha]_D^{20} + 42.5°$ (c0.10, MeOH), Its molecular formula was determined as $C_{12}H_{18}O_8$ by HR-EI-MS ([M+Na]$^+$ m/z 313.2132, calcd. 313.2062). The λ_{max} at 300 nm in UV spectrum revealed a conjugated double bond. The IR absorptions indicated the presence of methylene group (2936 cm^{-1}) and double bond (1654 cm^{-1}). The ^{1}H and ^{13}C NMR spectra (see Table 1), in combination with HSQC experiment, showed the presence of one terminal double bond [δ_H 6.37 (s, 2H, H-6), δ_C 154.4 (C-2), δ_C 94.8 (C-6)], one double bond [δ_C 132.8 (C-3), δ_C 153.5 (C-4)], one methyleneoxy group [δ_H 3.72 (3H, s, H-5), δ_C 60.6 (C-5)], one methoxyl group [δ_H3.57 (2H, s), δ_C 56.2], and a sugar group. The aglycone of **1** may be a substituted dihydrofuran, this assumption was subsequently confirmed by HMBC correlations: H-5/C-2, C-4; and H-6/C-2, C-3 (Fig. 1). Thus, the aglycone of **1** was elucidated as a methylene dihydrofuran. Compound 1 was hydrolyzed with 10% hydrochloric acid to yield glucose identified by comparing with the authentic samples on TLC, and D-glucose was confirmed by GC analysis after acid hydrolysis. The ^{13}C NMR data and the ^{1}H NMR signal at δ 4.76 (d, 1H, J = 8.0 Hz, H-1′) suggested the glucose was β configurated. The 4-*O*-glycopyranosyl and 3-OMe were determined on the HMBC correlations H-1′ (δ 4.76) and C-4 (δ 153.5), OMe (δ 3.57) and C-3 (δ 132.8). Therefore, compound **1** was identified as 2-methylene-3-methoxy-2, 5-dihydrofuran-4-*O*-β-D-glucopy-ranoside (Fig. 1). To the best of our knowledge, it is the first report of alkylene dihydrofuran glycoside as natural product.

The antioxidant activity of the compound **1** was investigated using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) and 2,2′-azinobis-3-ethylbenzothiazoline-6-sulphonic acid (ABTS) method [3,4], with TRLOX as positive controls (IC_{50} 1.90 mg/mL for DPPH, 0.29 mg/mL for ABTS). Compound **1** showed potential activity, with the IC_{50} values of 2.49 and 0.45 mg/mL, respectively.

Moracin M-3′-*O*-β-D-glucopyranoside (**2**), and moracin M-6, 3′-di-*O*-β-D-glucopyranoside (**3**) were identified by comparing their spectral data with those reported [5].

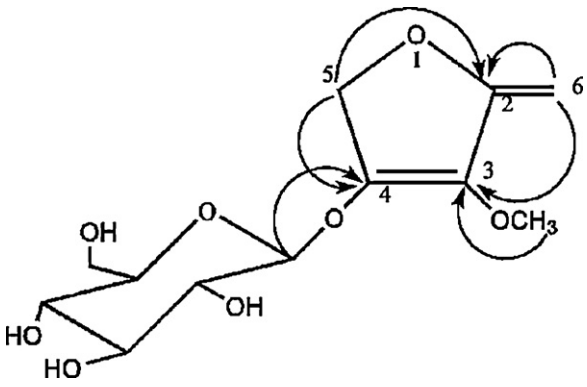

Fig. 1. Structure and key HMBC correlations of compound 1.

Acknowledgment

The author is grateful to Ping Lang (Department of Analysis, Huazhong University of Science and Technology) for recording NMR spectra.

References

[1] M.L. Xia, L.B. Qian, X.M. Zhou, et al. J. Ethnopharmacol. 120 (2008) 442.
[2] Pharmacopoeia Commission of People's Republic of China, The Pharmacopoeia of the People's Republic of China (part one), 2000 ed., Chemical Industry Press, Beijing.
[3] K. Shimada, K. Fujikawa, K. Yahara, et al. J. Agric. Food. Chem. 40 (1992) 945.
[4] R. Re, N. Pellegrini, A. Proteggente, et al. Free. Radic. Biol. Med. 26 (1999) 1231.
[5] T. Kanchanapoom, K. Suga, R. Kasai, Chem. Pharm. Bull. 50 (2002) 863.