Asian Journal of Chemistry; Vol. 26, No. 21 (2014), 7165-7167

ASIAN JOURNAL OF CHEMISTRY

http://dx.doi.org/10.14233/ajchem.2014.16538

A New Cytotoxic Prenylated Chalcone from *Desmodium renifolium*

JIE LOU, GUI-YOU LIU, HUAN WANG, JUAN-XIA YANG, LIMEI LI, QIU-FEN HU, YAN-QING YE and XUE-MEI GAO*

Key Laboratory of Chemistry in Ethnic Medicinal Resources, State Ethnic Affairs Commission and Ministry of Education, Yunnan University of Nationalities, Kunming 650031, P.R. China

*Corresponding author: E-mail: gao_xuemei@hotmail.com

Received: 17 October 2013; *Accepted*: 17 April 2014; *Published online*: 30 September 2014; AJC-16094

A new prenylated chalcone, renifchalcone A (**1**) was isolated from the whole plants of *Desmodium renifolium*. Its structure was determined by spectroscopic methods, including 1D and 2D-NMR techniques. Compounds **1** was tested for their cytotoxicity against five human tumor cell lines, NB4, A549, SHSY5Y, PC3 and MCF7 and it showed potential cytotoxicity against A549 and MCF7 cell lines with IC_{50} values of 5.8 and 6.2 μM, respectively.

Keywords: *Desmodium renifolium*, Prenylated chalcone, Cytotoxicity.

INTRODUCTION

Desmodium renifolium (Linn.) Schindl is a subshrubs belongs to the *Desmodium genus*, Leguminosae family. This plant extends from India, Burma, Thailand, Vietnam, Laos, Malaysia, to Indonesia and across the Pacific Islands[1]. In Yunnan province, it has been widely used for scattered willow, diuretic, antiinflammation, detoxification by Dai people living in Xishuangbanna prefecture[2]. The chemical compositions of *D. renifolium* had not been reported in literature. However, a series of bioactive compounds, such as flavonoids[3-5], alkaloids[5], terpenoids[5], steroides[5], phenylpropanoids[5], has been described from Desmodium genus. Continuing the efforts to discover more bioactive metabolites from local plants, we now investigate the chemical constituents of the whole plant of *D. renifolium* growing in Dehong prefecture. As a result, a new prenylated chalcone (**1**) was isolated from this plant. In this paper the structure elucidation of **1** and its biological evaluation are described.

EXPERIMENTAL

Optical rotations were measured with a Horiba SEPA-300 polarimeter. UV spectra were obtained using a Shimadzu UV-2401A spectrophotometer. A Tenor 27 spectrophotometer was used for scanning IR spectra. 1D- and 2D NMR spectroscopic data were recorded on a DRX-500 or DRX-400 NMR spectrometer with TMS as internal standard. Chemical shifts (δ) are expressed in ppm with reference to the TMS signal. HRESIMS was performed on a VG Autospec-3000 spectrometer. Semi-preparative HPLC was performed on a Shimadzu LC-8A preparative liquid chromatograph with Zorbax PrepHT GF (21.2 mm × 25 cm) or Venusil MP C_{18} (20 mm × 25 cm) columns. Column chromatography was performed using silica gel (200-300 mesh, Qing-dao Marine Chemical, Inc., Qingdao, China), Lichroprep RP-18 gel (40-63 μm, Merck, Darmstadt, Germany), Sephadex LH-20 (Sigma-Aldrich, Inc, USA), or MCI gel (75-150 μm, Mitsubishi Chemical Corporation, Tokyo, Japan). Column fractions were monitored by TLC visualized by spraying with 5 % H_2SO_4 in EtOH and heating.

Plant material: Whole plants of *Desmodium renifolium* (Linn.) Schindl., were collected in Honghe Prefecture, Yunnan province in September 2012. The species was identified by Prof Yuan Ning. A voucher specimen (YNNI 12-9-41) was deposited in the herbarium of the Yunnan University of Nationalities.

Extraction and Isolation: The samples (0.8 kg) were crushed to 30 mesh and the powders were extracted with 70 % aqueous acetone (4 × 2 L) at room temperature and filtered. The filtrate was evaporated under reduced pressure and the crude extract (72.2 g) was applied to a silica gel (150-200 mesh) column eluted with $CHCl_3$-MeOH gradients (9:1, 8:2, 7:3, 6:4, 5:5) to afford six fractions (A-E). Further separation of fraction A (42.1 g) by silica gel column chromatography, eluted with petroleum ether-acetone (9:1-1:2), yielded sub-fractions A1-A6. Subfraction A3 (2.21 g) was separated on a silica gel column eluted by petroleum ether-EtOAc, followed by semi-preparative HPLC (60 % MeOH-H_2O, flow rate 12 mL/min) to give 1 (5.8 mg).

Renifchalcone A (1): It was obtained as a pale yellow gum: $[\alpha]_D^{24.6}$ -1.98 (*c* 0.18, MeOH), UV (MeOH) λ_{max} (log ε)

210 (4.38), 225 (4.09), 295 (3.87) nm; IR (KBr, ν_{max}, cm^{-1}): 3387, 3142, 3075, 2954, 2867, 1641, 1602, 1567, 1463, 1345, 1192, 1063, 870; ^1H and ^{13}C NMR data (500 and 120 MHz, acetone-d_6) (Table-1); Positive ESIMS *m/z* 377 $[M + Na]^+$; Positive HRESIMS *m/z* 377.1360 $[M + Na]^+$ ($C_{21}H_{22}NaO_5$, calcd for: 377.1365).

RESULTS AND DISCUSSION

The whole plants of *D. renifolium* were extracted with 70 % aqueous acetone, followed by repeatedly column chromatography on silica gel, Sephadex LH-20 and RP-18 silica gel. Final purification by semi-preparative RP-HPLC afforded 15 chalcones. Its structure was shown in Fig. 1 and its ^1H and ^{13}C NMR data were given in Table-1.

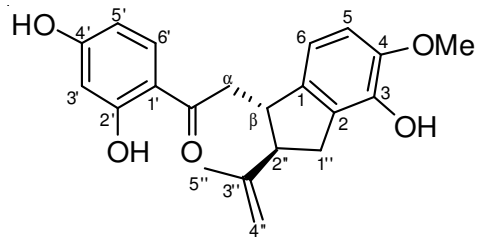

Fig.1. Structure of compound **1**

Compound (1) was obtained as pale yellow gum: The molecular formula $C_{21}H_{22}O_5$, with eleven degrees of unsaturation, was established based on HRESIMS (377.1360 $[M + Na]^+$; calcd for $C_{21}H_{22}NaO_5$, 377.1365) and NMR spectroscopy data (Table-1. The ^1H NMR (Table-1) spectrum of **1** showed the presence of a 1,2,4-tri-substituted benzene ring [δ_H 6.22 (1H, d, 1.6 Hz), 6.36 (1H, dd, 8.8, 1.6 Hz) and 7.63 (1H, d, 8.8 Hz)], as well as an AB-type spin system at [δ_H 6.47 (1H, d, 7.8 Hz) and 6.62 (1H, d, 7.8 Hz)]. The ^{13}C NMR and DEPT spectra (Table-1) revealed the presence of 21 carbons, which were assigned as twelve aromatic (five methines), two methyl (one methoxy), three methylene (one terminal double bond), two methine, two quaternary carbons (one carbonyl), suggesting that **1** is a prenylated chalcone with a tricyclic C20 skeleton[12]. The positions of the methoxy groups at C-4 was supported by the HMBC correlations (Fig. 2) of a the methoxy proton signal (δ_H 3.79) with C-4 (δ_C 148.8) and the other proton signal (δ_H 12.50) with C-3', C-4' and C-5'. Additionally, the positions of three phenolic hydroxy groups at C-3, C-2' and C-4' were also supported by the HMBC correlations of the hydroxy proton signals with corresponding carbons. The HMBC spectrum of **1** showed correlations from H_3-5" (δ_H 1.67) and H_2-4" (δ_H 4.88) to C-2" and C-3", from H-2" (δ_H 3.16) to C-1, C-1", C-2, C-4", C-5", C-α and C-β, from H_2-α (δ_H 2.99 and 2.93) to C-1', C-β, C-1, C=O and C-2", from H-β (δ_H 3.88) to C-1, C-1", C-α, C-2 and C-2" and from H-1" (δ_H 2.91) to C-1, C-2, C-2", C-β and C-3", coupled with a spin system ($CH_2CHCHCH$, H_2-α/H-β/H-2"/H_2-1") established by ^1H-^1H COSY correlations gave rise to a prenyl structural fragment with its C-2" conected with C-β in dihydrochalcone skeleton to form a carbocyclic of five members[6].

The relative configurations of **1** were deduced by the comparison of the optical rotation, coupling constants and ROESY correlations with these of antiarone J[6], of which relative configurations were unambiguously established by X-ray structure analysis. In the ROESY spectrum of **1**, the NOE correlations suggested that relative configurations of compound **1** are β for H-β and a for H-2". On the basis of the foregoing account, the structure of compound **1** was determined and gives the trivial name of renifchalcone A.

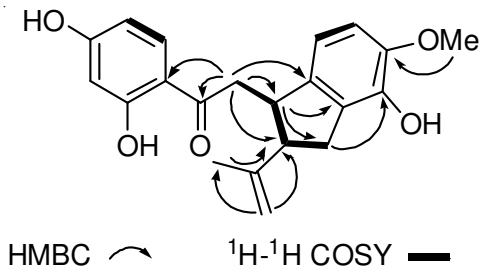

Fig. 2. ^1H-^1H COSY and key HMBC correlation of **1**

Many chalcone derivatives are known to be cytotoxic[7-10]. Compounds **1** was tested for its cytotoxicity against five human tumor cell lines (NB4, A549, SHSY5Y, PC3 and MCF7) using the MTT method, with paclitaxel as the positive control[11]. The results shown that compounds **1** exhibited potential cytotoxicity against A549 and MCF7 cell lines with IC_{50} values of 5.8 and 6.2 μM, respectively.

TABLE-1
^1H AND ^{13}C NMR DATA OF COMPOUND **1** (RECORDED AT 500 MHZ, ACETONE-d_6)

No.	δ_C	δ_H (m, J, Hz)	No.	δ_C	δ_H (m, J, Hz)
1	139.0 s	-	1″	31.5 t	2.91, overlapped
2	129.2 s	-	2″	52.2 d	3.16, br d (6.7)
3	141.3 s	-	3″	146.0 s	-
4	148.8 s	-	4″	112.6 t	4.88, d (8.0)
5	113.6 d	6.62, d (7.8)	5″	22.8 q	1.67, s
6	115.5 d	6.47, d (7.8)	α	38.5 t	2.99, dd (17.3, 6.0)
1′	113.4 s	-	β	42.3 d	2.93, dd (17.3, 7.3)
2′	164.5 s	-	C=O	205.6 s	3.88, br d (6.0)
3′	103.1 d	6.22, d (1.6)	4-OMe	55.8 s	3.79 s
4′	165.6 s	-	3-OH	-	12.68, s
5′	108.3 d	6.36, dd (8.8,1.6)	2′-OH	-	12.82, s
6′	133.2 d	7.63, d (8.8)	4′-OH	-	12.57, s

ACKNOWLEDGEMENTS

This project was supported partially by the National Natural Science Foundation of China (No. 21302164), the Excellent Scientific and Technological Team of Yunnan High School (2010CI08), the Yunnan University of Nationalities Green Chemistry and Functional Materials Research for Provincial Innovation Team (2011HC008) and Open Research Fund Program of the Key Laboratory of Ethnic Medicine Resource Chemistry (Yunnan University of Nationalities) (2010XY08).

REFERENCES

1. Editorial Committee of China Flora, Flora China, Vol. 41, p. 35 (1955).
2. S.W. Zhao and Z.Y. Dao, *Records for Dai Medicine of Xishuangbanna*, **2**, 263 (1981).
3. H. Sasaki, Y. Kashiwada, H. Shibata and Y. Takaishi, *Phytochemistry*, **82**, 136 (2012).
4. H. Sasaki, H. Sato, Y. Kashiwada, K. Kawazoe, K. Murakami, H. Shibata, H.D. Sun, S.L. Li and Y. Takaishi, *Planta Med*., **78**, 1851 (2012).
5. X. Ma, C. Zheng, C. Hu, K. Rahman and L. Qin, *J. Ethnopharmacol*., **138**, 314 (2011).
6. Y. Hano, P. Mitsui, T. Nomura, T. Kawai and Y. Yoshida, *J. Nat. Prod*., **54**, 1049 (1991).
7. B.P. Bandgar, S.S. Jalde, L.K. Adsul, S.N. Shringare, S.V. Lonikar, R.N. Gacche, N.A. Dhole, S.H. Nile and A.L. Shirfule, *Med. Chem. Res*., **21**, 4512 (2012).
8. P. Champelovier, M. Mininno, E. Duchamp, E. Nicolle, V. Curri, A. Boumendjel and J. Boutonnat, *Anticancer Res*., **31**, 3213 (2011).
9. M.M. Wu, L.Q. Wang, Y. Hua, Y.G. Chen, Y.Y. Wang, X.Y. Li, Y. Li, T. Li, X.Y. Yang and Z.-R. Tang, *Planta Med*., **77**, 481 (2011).
10. J.H. Lee, N.I. Baek, S.H. Kim, H.W. Park, J.H. Yang, J.J. Lee, S.J. Kim, S. Jeong, C.-H. Oh, K.-H. Lee and D.K. Kim, *Arch. Pharm. Res*., **30**, 408 (2007).
11. X.M. Gao, R.R. Wang, D.Y. Niu, C.Y. Meng, L.M. Yang, Y.T. Zheng, G.Y. Yang, Q.F. Hu, H.D. Sun and W.L. Xiao, *J. Nat. Prod*., **76**, 1052 (2013).