Asian Journal of Chemistry; Vol. 27, No. 5 (2015), 1947-1948

ASIAN JOURNAL OF CHEMISTRY

http://dx.doi.org/10.14233/ajchem.2015.17381

NOTE

A New Eremophilane-Type Sesquiterpene from Flue-Cured Tobacco and Its Anti-tobacco Mosaic Virus Activity

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Received: 13 February 2014; *Accepted*: 22 April 2014; *Published online*: 20 February 2015; AJC-16930

A new eremophilane-type sesquiterpene, tobterpene B (**1**), was isolated from the stems of flue-cured tobacco (a variety of *Nicotiana tabacum* L). Its structure was elucidated by spectroscopic methods, including extensive 1D and 2D NMR techniques. Compound **1** was tested for its anti-tobacco mosaic virus (anti-TMV) activity and it shows potential anti-tobacco mosaic virus activity with inhibition rates of 15.2 %.

Keywords: Sesquiterpene, Flue-cured tobacco, Anti-tobacco mosaic virus activity.

Nicotiana tabacum L. is the most commonly grown of all plants in the *Nicotiana* genus and its leaves are commercially grown in many countries to be processed into tobacco[1,2]. In addition to being used in cigarette industry, *N. tabacum* is also used as insecticide, anesthetic, diaphoretic, sedative and emetic agents in Chinese folklore medicine because of it containing many useful chemical compounds.[1,3-5]. In previous work, a number of bioactive compounds, such as terpenoids[6-8], alkaloids[9,10], lignans[11,12], flavonoid[13], phenylpropanoids[14] and the homologous, were isolated from this plant. In this study, we reported the isolation of a new eremophilane-type sesquiterpene, tobterpene B (**1,** Fig. 1), structure of this new compound was evaluated by spectroscopic methods, including HRMS and ^{1}D and ^{2}D NMR. In addition, its anti-tobacco mosaic virus (anti-TMV) activity was evaluated for compound **1**.

Optical rotations were obtained on a Perkin-Elmer 341 digital polarimeter; UV spectra were obtained using a Shimadzu UV-2401A spectrophotometer. IR spectra were obtained in KBr disc on a Bio-Rad Wininfmred spectrophotometer. ESI-MS were measured on a VG Auto Spec-3000 MS spectrometer. ^{1}H, ^{13}C and 2D NMR spectra were recorded on Bruker DRX-500 instrument with TMS as internal standard. Column chromatography was performed on silica gel (200-300 mesh), or on silica gel H (10-40 μm), Qingdao Marine Chemical Inc., China). Preparative HPLC was used an Agilent 1100 HPLC equipped with ZORBAX-C18 (21.2 mm × 250 mm, 7.0 mm) column and DAD detector.

Plant material: The stems of flue-cured tobacco were collected in Honghe Prefecture, Yunnan Province, P.R. China, in September 2012. The identification of the plant material was verified by Prof. Y.J. Chen (Yunnan University of Nationalities).

Extraction and isolation: The air-dried and powdered tobacco stems (2.5 kg) were extracted four times with 90 % methanol (4 × 3.5 L) at room temperature and filtered to yield a filtrate. The crude extract (55.6 g) was applied to silica gel (200-300 mesh) column chromatography, eluting with a chloroform-acetone system (20:1, 9:1, 8:2, 7:3, 6:4, 5:5), to give six fractions A-F. The further purification of fraction C (8:2, 22.4 g) by silica gel column chromatography, eluted with petroleum ether-acetone (9:1, 8:2, 7:3, 6:4, 5:5), yielded mixtures C1-C5. Fraction C-3 (7:3, 1.88 g) was subjected to preparative HPLC (55 % MeOH-H_2O, flow rate 12 mL/min) to yield compound **1** (11.6 mg).

Tobterpene B: Obtained as a white amorphous powder; $[\alpha]_D^{23.6}$ -115 (*c* 0.20, MeOH); UV (MeOH), λ_{max} (log ε) 232 (4.23), 285 (3.90), 325 (3.68) nm; IR (KBr, ν_{max}, cm^{-1}) 3385, 2962, 2925, 1690, 1615, 1562, 1473, 1454, 1330, 1225, 1148, 1122, 993, 860; ^{1}H NMR and ^{13}C NMR data (C_5D_5N, 500 MHz and 125 MHz, respectively), (Table-1); ESIMS (negative ion mode), *m/z* 271 [M-H]$^-$; HRESIMS (negative ion mode), *m/z* 271.0602 [M-H]$^-$ (calcd. 271.0606 for $C_{15}H_{11}O_5$).

Compound **1** was obtained as white amorphous powder. Its molecular formula was determined as $C_{15}H_{12}O_5$ by HRESIMS, *m/z* 271.0602 [M-H]$^-$ (calcd. 271.0606), corres-

TABLE-1
^{1}H AND ^{13}C NMR DATA OF COMPOUND **1** (δ in ppm, IN C_5D_5N, 500 AND 125 MHz)

Position	δ_C (m)	δ_H (m, *J*, Hz)	Position	δ_C (m)	δ_H (m, *J*, Hz)
1	196.2 s		9	142.2 s	-
2	56.7 d	3.82 d (4.5)	10	119.0 s	-
3	58.2 d	4.02 dd (4.5, 2.5)	11	113.8 s	-
4	33.5 d	4.12 dq (6.8, 2.5)	12	146.9 d	7.56 s
5	137.5 s	-	13	68.6 t	5.22, 5.28 s
6	124.7 s	-	14	62.2 t	5.19, 5.23 s
7	132.2 s	-	15	19.5 q	1.22 (d) 6.8
8	145.4 s	-	Ar-OH		12.28 s

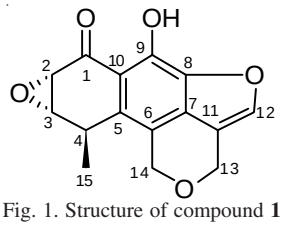

Fig. 1. Structure of compound **1**

ponding to ten degrees of unsaturation. Its ^{1}H and ^{13}C NMR spectral data (Table-1) showed signals to 12 hydrogens and 15 carbons, respectively, corresponding to eight aromatic carbons (δ_C 137.5, 124.7, 132.2, 145.4, 142.2, 119.0, 113.8, 146.9) with one aromatic praton (δ_H 7.56), one methane group (δ_C 33.5, δ_H 4.12), two oxygened methane groups (δ_C 56.7, 58.2; δ_H 3.82, 4.02), two oxygened methylene groups (δ_C 68.6, 62.2; δ_H 5.22, 5.28, 1H each and 5.19, 5.23 1H each), a methyl group (δ_C 19.5, δ_H 1.22), a carbonyl group (δ_C 196.2) and a phenolic hydroxy group (δ_H 12.28). Strong absorption bands accounting for hydroxyl (3385 cm^{-1}), carbonyl (1690 cm^{-1}) and aromatic group (1615, 1562, 1473 cm^{-1}) could be observed in its IR spectrum. The UV spectrum of compound **1** showed absorption maxima at 325, 285 and 232 nm also confirmed the existence of the aromatic function. Both the ^{1}H and ^{13}C NMR spectra (Tables-1) of compound **1** were similar to the published values for 14-angeloyloxy-2R,3R-epoxy-1-oxo-*O*-methylcacalol[15]. The noted differences between two compounds were due to the disappearance of an angeloyloxy and a methoxy group and the appearance of a phenolic hydroxy group. In addition, a methyl was also be oxidized to a methylene in compound **1**. The HMBC correlations (Fig. 2) between the phenolic hydroxy signal at δ_H 12.28 and C-8 (δ_C 145.4), C-9 (δ_C 142.2) and C-10 (δ_C 119.0) confirmed the hydroxy groups attached to C-9. C-13 linked to C-14 by an oxygen atom was supported by the HMBC correlations of H-14 (δ_H 5.19, 5.23) to C-13 (δ_C 68.6) and H-13 (δ_H 5.22, 5.28) to C-14 (δ_C 62.2). Forthure more, the existence of an oxygen ring was also supported by the ten degrees of unsaturation in this compound. Thus, the structure of compound **1** was established as shown in Fig. 2.

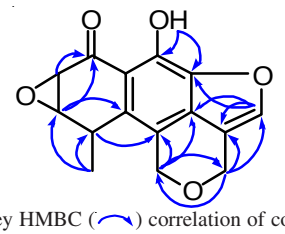

Fig. 2. Key HMBC ( ) correlation of compound **1**

Compounds **1** was tested for it anti-tobacco mosaic virus activity. The anti-TMV activities were tested using the half-leaf method[16]. Ningnanmycin (2 % water solution), a commercial product for plant disease in China, was used as a positive control. The results showed that compound **1** exhibited inhibition rates of 15.2 %.

ACKNOWLEDGEMENTS

This project was supported financially by the National Natural Science Foundation of China (No.31360081), the Basic Research Foundation of Yunnan Province (2013FB097) and the Basic Research Foundation of Yunnan Tobacco Industry Co. Ltd (2012JC01).

REFERENCES

1. The Editorial Committee of the Administration Bureau of Flora of China, Flora of China, 67 vols. (Beijing Science and Technology Press, Beijing, China (2005).
2. T.W. Hu and Z. Mao, *Tob. Control*, **15(suppl. 1)**, i37 (2006).
3. A. Rodgman and T.A. Perfetti, The Chemical Components of Tobacco and Tobacco Smoke, CRC Press, Taylor and Francis Group, Boca Raton, Florida (2008).
4. A.P. Cavender and M. Alban, *J. Ethnobiol. Ethnomed*., **5**, 3 (2009).
5. A. Inta, P. Shengji, H. Balslev, P. Wangpakapattanawong and C. Trisonthi, *J. Ethnopharmacol*., **116**, 134 (2008).
6. X. Feng, J.-S. Wang, J. Luo and L.-Y. Kong, *J. Asian Nat. Prod. Res*., **12**, 252 (2010).
7. Y. Shinozaki, T. Tobita, M. Mizutani and T. Matsuzaki, *Biosci. Biotechnol. Biochem*., **60**, 903 (1996).
8. T. Pettersson, A.M. Eklund and I. Wahlberg, *J. Agric. Food Chem*., **41**, 2097 (1993).
9. X.C. Wei, S.C. Sumithran, A.G. Deaciuc, H.R. Burton, L.P. Bush, L.P. Dwoskin and P.A. Crooks, *Life Sci*., **78**, 495 (2005).
10. T. Braumann, G. Nicolaus, W. Hahn and H. Elmenhorst, *Phytochemistry*, **29**, 3693 (1990).
11. Y.K. Chen, X.S. Li, G.Y. Yang, Z.Y. Chen, Q.F. Hu and M.M. Miao, *J. Asian Nat. Prod. Res*., **14**, 450 (2012).
12. Q.-F. Hu, G. Yang, X. Li, X. Yang, H. Mu, Y. Chen and X.-M. Gao, *Heterocycles*, **85**, 147 (2012).
13. Z.Y. Chen, J.L. Tan, G.Y. Yang, M.M. Miao, Z.Y. Chen and T.F. Li, *Phytochem. Lett*., **5**, 233 (2012).
14. Y. Chen, T. Li, Z.-Y. Chen, G. Yang, M. Miao and J. Tan, *Heterocycles*, **83**, 2381 (2011).
15. G.D. Huang, Y.J. Yang, W.S. Wu and Y. Zhu, *J. Nat. Prod*., **73**, 1954 (2010).
16. Q.F. Hu, B. Zhou, X.M. Gao, L.Y. Yang, L.D. Shu, Y.Q. Shen, G.P. Li, C.T. Che and G.Y. Yang, *J. Nat. Prod*., **75**, 1909 (2012).