Antiviral Constituents against Respiratory Viruses from *Mikania micrantha*

Paul Pui-Hay But,*[,†] Zhen-Dan He,[†] Shuang-Cheng Ma,[†] Yiu-Man Chan,[†] Pang-Chui Shaw,[†] Wen-Cai Ye,[‡] and Ren-Wang Jiang*[,†,‡]

Departments of Biology and Biochemistry and Institute of Chinese Medicine, The Chinese University of Hong Kong, Shatin, Hong Kong, People's Republic of China, and Guangdong Province Key Laboratory of Pharmacodynamic Constituents of Traditional Chinese Medicine and New Drugs Research, Institute of Chinese Medicine and Natural Products, Jinan University, Guangzhou, People's Republic of China

Received August 26, 2008

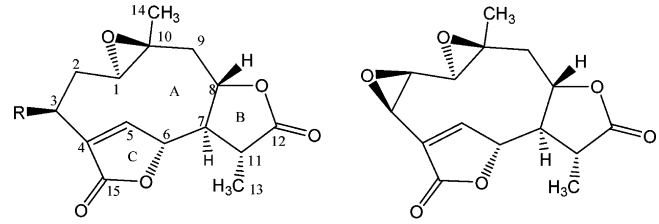
Phytochemical investigation of the dried aerial parts of *Mikania micrantha* led to the isolation of a new sesquiterpene, 3β-acetoxy-1,10-epoxy-4-germacrene-12,8;15,6-diolide (**1**), along with six known constituents: 1,10-epoxy-4-germacrene-12,8;15,6-diolide (**2**), dihydromikanolide (**3**), potassium mikanin 3-sulfate (**4**), mikanin (**5**), alpinetin (**6**), and ergosta-7,22-dien-3β-ol (**7**). Their structures were elucidated by spectroscopic methods, and the molecular structures and stereochemistry of sesquiterpene lactones **1**−**3** were revealed by single-crystal X-ray analysis. Compound **2** showed moderate activity against respiratory syncytial virus (IC_{50} = 37.4 uM) and parainfluenza type 3 virus (IC_{50} = 37.4 uM) with a therapeutic index (TI) of 16.0 for both compounds. Compound **4**, the main component of *M. micrantha*, exhibited inhibitory activity against parainfluenza type 3 virus with IC_{50} (19.7 uM) and TI (24.0) values comparable to those of ribavirin, serving as a positive control.

Mikania micrantha H.B.K. (Compositae) is a fast-growing, perennial creeping vine native to tropical America. It has spread to other tropical areas in the Old World and in Australia and the Pacific Islands. Away from its native homeland and natural enemies, this vine has become extremely weedy, reproducing rapidly both vegetatively and sexually, and thus smothering young tree crops and other plants in disturbed terrestrial habitats. Moreover, it contains allelopathic substances that inhibit the growth of other plants and reduce nitrification in soil.[1,2] It is known that herbicides such as glyphosate can kill it,[3] but large-scale herbicide application would certainly lead to widespread environmental pollution and damage to native vegetation. Various parasites have been suggested as potential biocontrol agents for this vine, but repeated releases of its natural enemy (*Liothrips mikaniae*) in Malaysia and Solomon Islands failed, while tests on other pests have not yet produced applicable results.[4−6] Therefore, in many delicate habitats such as mangroves in Hong Kong where migratory birds visit and breed, removing the weed by hand appears to be the only alternative. Indeed, in the mangrove reserves along the coast of Shenzhen, the city connected to Hong Kong, servicemen patrolling the border were called in to help remove the vine manually. These servicemen posted slogans in the mangrove, calling for "the whole nation to mobilize all hands to eradicate *Mikania*".[7] While weeding of *M. micrantha* is a legitimate approach, we are exploring the alternative of "turning this weed into drug". The importance of weeds to ethnopharmacology and therapeutic application is also noted by other researchers.[8]

Previous studies have reported that *M. micrantha* is used to treat skin itches and athlete's foot as an alternative medicine in Jamaica, and the sesquiterpenes mikanolide and dihydromikanolide show antibacterial and antimicrobial activities.[9] Chemical studies have established that *M. micrantha* contains essential oils and sesquiterpenoid lactones of the germacrane type.[10,11] Related species of the same genus have been reported to exhibit cytotoxic,[12] anticarcinogenic,[13] antiallergic,[14] antiulcer,[15] antiprotozoal,[16] insecticidal, and trypanocidal activities.[17]

In our screening tests, we found that the methanol extract of the dried aerial part of *M. micrantha* showed inhibitory activities against respiratory syncytial virus (RSV, IC_{50} = 60 μg/mL). Further work led to the isolation of a new sesquiterpene, 3β-acetoxy-1,10-epoxy-4-germacrene-12,8;15,6-diolide (**1**), as well as six known constituents: 1,10-epoxy-4-germacrene-12,8;15,6-diolide (**2**), dihydromikanolide (**3**), potassium mikanin 3-sulfate (**4**), mikanin (**5**), alpinetin (**6**), and ergosta-7,22-dien-3β-ol (**7**). We here report the structure elucidation and antiviral activity against respiratory viruses of these compounds.

Scheme 1. Structural Formulas of Compounds **1**−**3**



1 R= OAc
2 R= H

3

Compound **1** was obtained as colorless triangular plates. HR-FABMS analysis indicated a quasimolecular ion $[M + H]^+$ at *m/z* 337.3467, corresponding to a molecular formula $C_{17}H_{20}O_7$ with eight degrees of unsaturation. The formula was also confirmed by the EI mass spectrum (*m/z* 336 $[M]^+$). The IR bands at 1777 and 1750 cm^{-1} suggested the presence of acetyl and lactone functionalities. The ^1H and ^{13}C NMR spectral data (Tables 1) indicated three methyl groups at δ 1.25 (d, J = 7.2 Hz, H_3-13), 1.07 (s, H_3-14), and 2.16 (s, H_3-17), seven methines including four oxymethines at δ 2.96 (dd, J = 2.0, 10.4 Hz, H-1), 5.59 (br d, J = 2.0 Hz, H-3), 5.40 (s, H-6), and 4.58 (dt, J = 4.4, 11.2 Hz, H-8), an olefinic proton at 7.96 (s, H-5), two methylenes, and five quaternary carbons including one ester carbonyl at δ 169.1 (s, C−16), two lactone carbonyls at δ 175.6 (s, C-12) and 169.6 (s, C-15), one olefinic carbon at δ 130.9 (s, C-4), and a carbon bound to oxygen at δ 56.8 (s, C-10). HMBC correlations from the methyl group at δ 2.16 confirmed the location of the acetyl group at C-3. $^1H-^1H$ COSY, HSQC, and HMBC experiments facilitated assignment of all ^1H and ^{13}C NMR signals (Tables 1). An X-ray crystallographic study was performed to confirm the structure and relative configuration of **1** (Table 2). A perspective view of the molecular structure is given in Figure 1. Accordingly, compound **1** was characterized as 3β-acetoxy-1,10-

* Corresponding authors. (P.P.-H.B.) Tel: (852) 26096299. Fax: (852) 26035248. E-mail: paulbut@cuhk.edu.hk. (R.-W.J.) Tel: (8620) 85221016. Fax: (8620) 85221559. E-mail: rwjiang@jnu.edu.cn.
† The Chinese University of Hong Kong.
‡ Jinan University.

10.1021/np800542t CCC: $40.75
Published on Web 03/06/2009

Table 1. ^{1}H and ^{13}C NMR Data of Compounds **1** and **2** in DMSO-d_6

	1		**2**	
position	δc, mult.	δ_H (*J* in Hz)	δc, mult.	δ_H (*J* in Hz)
1	57.9, CH	2.96, dd (2.0, 10.4)	60.9, CH	2.81, dd (2.0, 9.6)
2	28.8, CH_2	2.22, dt (2.0, 12.8) 2.62, dd (10.4, 12.8)	22.3, CH_2	1.29, m 2.05, m
3	66.6, CH	5.59, br d (2.0)	21.3, CH_2	2.49, m
4	130.9, qC		130.7, qC	
5	148.4, CH	7.96, s	149.3, CH	7.79, s
6	80.2, CH	5.40, s	79.4, CH	5.25, s
7	52.5, CH	1.65, dd (4.4, 12.0)	52.6, CH	2.56, m
8	77.2, CH	4.58, dt (4.4, 11.2)	77.2, CH	4.40, dt (4.4, 11.3)
9	41.9, CH_2	1.91, dd (4.4, 14.4) 2.04, dd (11.2, 14.4)	42.5, CH_2	1.87, dd (4.4, 14.0) 2.01, dd (11.3, 14.0)
10	56.8, qC		56.6, qC	
11	39.2, CH	2.90, dq (7.2, 12.0)	39.7, CH	2.93, dq(6.8, 12.8)
12	175.6, qC		175. 7, qC	
13	13.0, CH_3	1.25, d (7.2)	13.0, CH_3	1.25,d (6.8)
14	20.8, CH_3	1.07, s	19.9, CH_3	1.05, s
15	169.6, qC		171.8, qC	
Ac	169.1, qC 20.0, CH_3	2.16, s		

Table 2. Crystallographic Data of Compounds **1**, **2**, and **3**

	1	**2**	**3**
CCDC deposit no.	161088	161087	161086
color/shape	colorless/triangular plate	colorless/rectangular plate	colorless/plate
cryst dimens (mm^3)	0.09 × 0.46 × 0.65	0.28 × 0.40 × 0.62	0.21 × 0.34 × 0.66
chemical formula	$C_{17}H_{20}O_7$	$C_{15}H_{18}O_5$	$C_{15}H_{16}O6$
fw	336.33	278.25	292.28
temperature, K	293(2)	293(2)	293(2)
cryst syst	monoclinic	monoclinic	monoclinic
space group	$P2_1$	$P2_1$	$P2_1$
unit cell dimens	a = 6.280(1) Å	a = 9.464(1) Å	a = 9.225(1) Å
	b = 9.702(2) Å	b = 6.9503(8) Å	b = 7.128(2) Å
	c = 13.448(3) Å	c = 10.560(1) Å	c = 10.426(1) Å
	β = 93.421(5)°	β = 102.045(2)°	β = 99.645(9)°
volume, $Å^3$	819.1(3)	679.31(14)	675.9(2)
Z	2	2	2
density (calcd), g/cm^3	1.364	1.361	1.436
absorp coeff, mm^{-1}	0.106	0.102	0.112
diffractometer/scan	Bruker SMART1000 CCD/ω	Bruker SMART1000 CCD /ω	Bruker P4/ω
θ range for data collection, deg	1.51 to 23.24	1.97 to 23.27	1.98 to 24.99
reflns measd	3789	3217	1789
indep reflns	2152	1893	1497
obsd reflns	1971	1735	1344
data/restrains/params	2152/1/217	1893/1/181	1497/1/190
goodness of fit on F^2	1.009	1.023	1.057
final R indices [$I > 4\sigma(I)$]	0.0320	0.0322	0.0330
R indices (all data)	0.0371	0.0358	0.0390

epoxy-4-germacrene-12,8;15,6-diolide, an epimer of dihydroscandenolide isolated from *M. scandens*.[18]

Compound **2** was obtained as colorless plates. Its molecular formula was identified as $C_{15}H_{18}O_5$ from the EIMS spectrum (*m*/*z* 278 $[M]^+$) and HRFABMS spectrum (*m*/*z* 279.3118 $[M + H]^+$). UV (209 nm) and IR (1760 cm^{-1}) indicated the presence of lactone

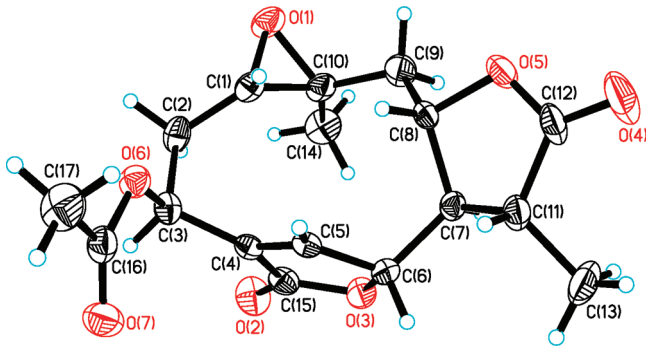


Figure 1. Molecular structure of compound **1** with atom-labeling scheme. The C and O atoms are drawn as 30% thermal ellipsoids.

functionalities in **2**. The ^{1}H and ^{13}C NMR spectra (Table 1) were similar to those of **1** except for the absence of an O-acetyl group at C-3. X-ray diffraction analysis revealed the complete structure and stereochemistry of **2**, showing the framework of a 10-membered ring fused with an expoxide and two lactone rings. A perspective view of the molecular structure of **2** is given in Figure 2. Therefore, compound **2** was identified as 1,10-epoxy-4-germacrene-12,8;15,6-diolide. Compound **2** was isolated from *M. periplocifolia* three decades ago.[19] However, the crystal structure and spectral data were not reported except for partial proton NMR data. We thus identified it from *M. micrantha* for the first time.

The known compound **3** was identified as dihydromikanolide by comparison of its physical data with reported values[18] and further confirmed by X-ray analysis. A perspective view of the molecular structure of **3** is given in Figure 3. The crystal structure of **3** was reported three decades ago, but with lower precision (R = 0.062 for 1486 reflections).[20] It was also identified from *M. micrantha* for the first time.

All three sesquiterpene lactones have a 10-membered ring, a saturated lactone ring, and an unsaturated lactone ring. The 10-membered ring A in **1** and **2** is fused with one epoxide ring, whereas in **3** it is fused with two epoxide rings, which make a dihedral angle of 145.7°. This structural difference leads to different

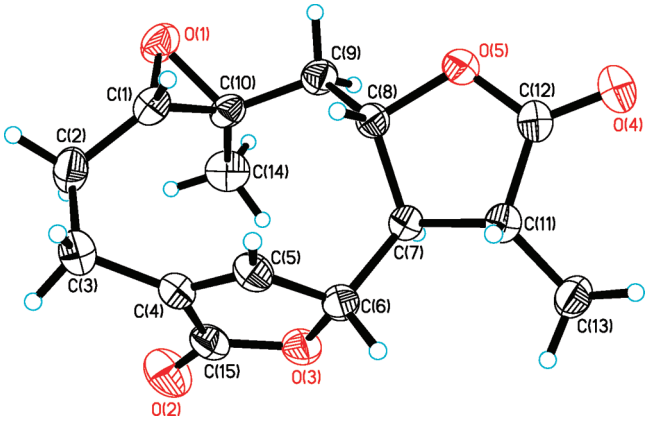


Figure 2. Molecular structure of compound **2** with atom-labeling scheme. The C and O atoms are drawn as 30% thermal ellipsoids.

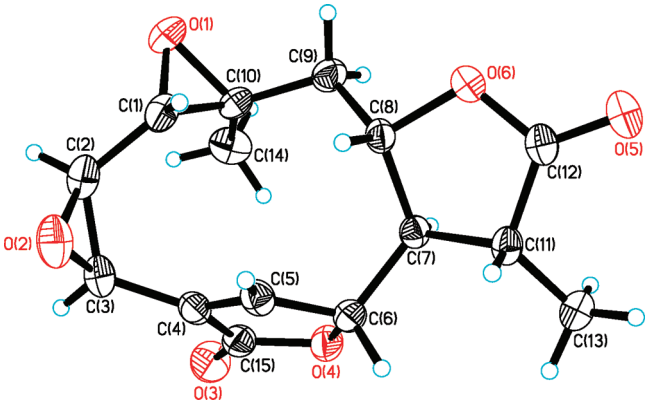


Figure 3. Molecular structure of compound **3** with atom-labeling scheme. The C and O atoms are drawn as 30% thermal ellipsoids.

conformations of the 10-membered ring. The 10-membered ring A in **1** and **2** exists in a boat−chair−chair conformation (Figure 4a). In contrast, in **3** it can be divided into two parts: fragment C-5−C-6−C-7−C-8−C-9−C-10−C-1 exists in a boat conformation and fragment C-1−C-2−C-3−C-4 lies on a plane with a maximum deviation of 0.008Å (Figure 4b). The conformational difference is also indicated by the torsion angle C-1−C-2−C-3−C-4 of 42.6°, 41.6°, and −2.3°, for **1**, **2**, and **3**, respectively. The conformations of the 10-membered rings in **1**, **2**, and **3** are significantly different from the chair−boat−chair conformation of macrocaesalmin, which is also due to the different fusion environment.[21] Though investigations of the conformation of germacrane sesquiterpenes based on NMR data[22] and molecular mechanics[23] were reported before, conformational comparison of the macro-rings by X-ray analysis is presented for the first time. The saturated lactone ring B adopts an envelope conformation with C-7 displaced by 0.542, 0.490, and 0.527 Å from the least-squares plane of the remaining four atoms for **1**, **2**, and **3**, respectively. The unsaturated lactone ring C is planar for all three compounds. The C−C bond distances in the epoxide ring of the three compounds (**1**: 1.480 Å, **2**: 1.466 Å, and **3**: 1.478 Å) are significantly shorter than the C(sp^3)−C(sp^3) distances but similar to the single-bond values between two sp^2 carbon atoms.[24]

The known constituents **4**−**7** were identified by comparison of their physical data with reported values, as potassium mikanin 3-sulfate (**4**),[25] mikanin (**5**),[25] alpinetin (**6**),[26] and ergosta-7,22-dien-3β-ol (**7**).[27]

Compounds **1**−**7** were screened for inhibitory effects against RSV, parainfluenza type 3 (Para 3), and influenza type A (Flu A) in cell culture monolayers employing cytopathagenic effect reduc-

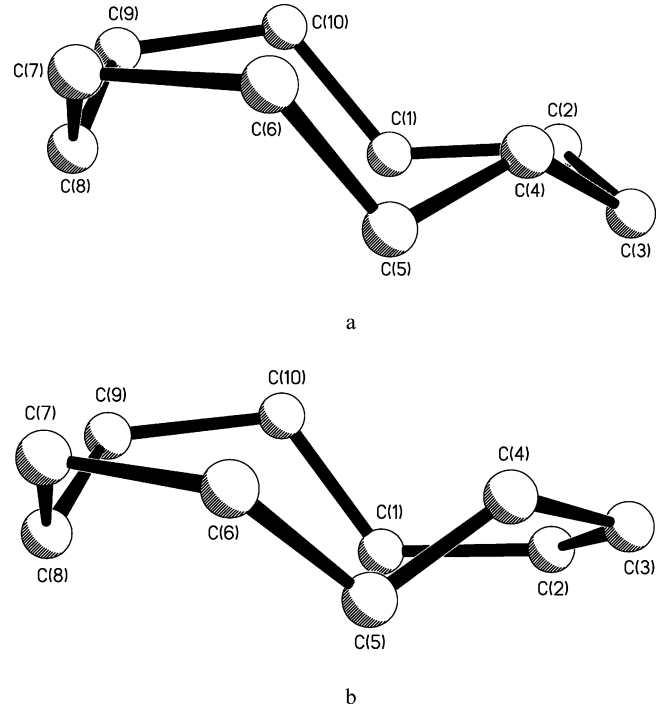


Figure 4. Conformation of the 10-membered ring in compounds **1** and **2** (Figure 4a) and **3** (Figure 4b).

tion assay. The three viruses are often the cause of severe respiratory diseases, particularly in infants and young children.[28] Our results demonstrated that compounds **1**−**7** showed different levels of antiviral activities *in vitro* against these viruses. Compound **2** showed moderate activity against RSV (IC_{50} = 37.4 uM) and Para 3 (IC_{50} = 37.4 uM) with a therapeutic index (TI, the ratio of TC_{50}/IC_{50}) of 16.0 for both viruses (Tables S1 and S2, Supporting Information). Compound **4** (yield 0.30%), the main component of *M. micrantha*, exhibited inhibitory activity against Para 3 with IC_{50} (19.7 uM) and TI (24.0) values that were comparable to those of ribavirin, serving as a positive control (Table S2). Compounds **1**−**7** showed weak anti-Flu A activities (Table S3, Supporting Information). There is potential to develop *M. micrantha* into an antiviral remedy, so as to "turn this weed into drug". The antiviral activity of this herb is reported for the first time.

Experimental Section

General Experimental Procedures. Optical rotations were measured with a JASCO DIP-370 digital polarimeter. UV spectra were recorded using a Shimadzu UV-3100PC spectrophotometer. IR absorption spectra were obtained using an IR-450 instrument. ESIMS was recorded on a Finnigan TSQ 7000 mass spectrometer. NMR spectra were obtained with a Bruker 400 spectrometer operating at 400 MHz for ^{1}H and 100 MHz for ^{13}C, respectively. Column chromatographies were performed with silica gel (Qingdao Haiyang Chemical Group Co. Ltd., China), reversed-phase C-18 (60u, Merck), and Sephadex LH-20 (Pharmacia Fine Chemical Co. Ltd.). TLC was performed on precoated silica gel 60 F_{254} plates (0.2 mm thick, Merck).

Plant Material. The dried aerial parts of *M. micrantha* growing in Hong Kong were collected in June 2000. A voucher specimen (Chan 2000-1) was deposited in the Herbarium, Department of Biology, Chinese University of Hong Kong.

Extraction and Isolation. The dried aerial part of *M. micrantha* (650 g) was extracted with hot MeOH (2500 mL × 3) under reflux condtions. The IC_{50} of the MeOH extract against RSV was 60 μg/mL. Compound **4** (1.9535 g) was directly crystallized from the methanol extract. The remaining extract was concentrated to give a residue (100 g), which was suspended in H_2O (800 mL) and partitioned with *n*-hexane and $CHCl_3$ to afford three fractions. The *n*-hexane, $CHCl_3$, and aqueous fractions were tested against RSV, and the IC_{50} values were 133.3, 19.4, and 60.2 μg/mL, respectively. Further purification

of the $CHCl_3$ fraction by column chromatography led to the isolation of **1** (15.1 mg), **2** (20.1 mg), **3** (39.3 mg), **6** (4.8 mg), and **7** (9.2 mg), and purification of the aqueous fraction led to the isolation of **5** (70.1 mg). Crystals of **1**−**3** were obtained from $CHCl_3$−MeOH (1:3) solution. For detailed isolation procedures, see the Supporting Information.

3*β*-Acetoxy-1,10-epoxy-4-germacrene-12,8;15,6-diolide (1): colorless triangular plates, mp 271−273 °C; $[\alpha]^{20}_D$ +54 (*c* 0.25, $CHCl_3$); UV ($CHCl_3$) λ_{max} (log ϵ) 205 (3.47) nm; IR (KBr) ν_{max} 1777 and 1750 cm^{-1}; ^1H and ^{13}C NMR data (see Table 1); EIMS *m*/*z* 336 $[M]^+$, 318 $[M - H_2O]^+$, 292 $[M - 44]^+$; HRFABMS *m*/*z* found 337.3467 $[M + H]^+$ ($C_{17}H_{21}O_7$ calcd 337.3495).

1,10-Epoxy-4-germacrene-12,8;15,6-diolide (2): colorless rectangular plates, mp 215−217 °C; $[\alpha]_D^{20}$ +71 (*c* 0.31, $CHCl_3$); UV ($CHCl_3$) λ_{max} (log ϵ) 209 (3.47) nm; IR (KBr) ν_{max} 1760 and 1650 cm^{-1}; ^1H and ^{13}C NMR data (see Table 1); EIMS *m*/*z* 278 $[M]^+$, 260 $[M - H_2O]^+$, 234 $[M - 44]^+$; HRFABMS *m*/*z* found 279.3118 $[M + H]^+$($C_{15}H_{19}O_5$ calcd 279.3129).

X-ray Analysis. X-ray diffraction data of **1** and **2** were collected on a Bruker SMART1000 CCD diffractometer, and those of **3** were collected on a Bruker P4 four-circle diffractometer using Mo Kα radiation (0.71073 Å). The structures were solved by direct methods (SHELXTL version 5.1) and refined by full-matrix least-squares on F^2. The crystallographic data of compound **1**, **2**, and **3** are displayed in Table 2.

Antiviral Assay. The antiviral activities of isolated compounds **1**−**7** were evaluated against RSV, Flu-A, and Para-3 viruses, using a cytopathagenic effect reduction assay as described previously.[29]

Acknowledgment. Partial support was received from the Kong Kong Innovation and Technology Fund (Industrial Support Fund AF/281/97), Direct grant of the Chinese University of Hong Kong (CUHK), and starting fund (51208005) and 211 project of Jinan University. Assistance from Prof. T. T. W. Mak, Prof. S. H. S. Lee, and Prof. S. F. Lee is deeply appreciated.

Supporting Information Available: Crystal data of **1**−**3** in standard CIF format have been deposited with the Cambridge Crystallographic Data Centre with CCDC numbers shown in Table 2. Copies of the data can be obtained, free of charge, on application to CCDC, 12 Union Road, Cambridge CB2 1EZ, UK (fax: +44-1223-336033; e-mail: deposit@ccdc.cam.ac.uk). Detailed isolation procedures, tables of antiviral properties of **1**−**7** against RSV, Para 3, and Flu A viruses (S1−S3), and tables of crystal data, bond distances, bond angles, and torsion angles for compound **1**−**3** are available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Ismail, B. S.; Mah, L. S. *Plant Soil* **1993**, *157*, 107–113.
(2) Ni, G. Y.; Li, F. L.; Chen, B. M.; Song, L. Y.; Peng, S. L. *Allelopathy J.* **2007**, *19*, 287–295.
(3) Hu, Y. J.; But, P. P. H. *Acta Sci. Nat. Univ. Sunyatseni* **1994**, *33* (4), 88–95.
(4) Shen, H.; Ye, W. H.; Hong, L.; Cao, H. L.; Wang, Z. M. *J. Exp. Bot.* **2006**, *56*, 1277–1284.
(5) But, P. P. H., Hu, Y. J.; Chu, L. M.; Wan, C. H.; Woo, T. K., *Abstracts of the 5th International Weed Science Congress*; June 23−27, 2008, Vancouver, Canada, 2008; p 68.
(6) Cook, M. J. W.; Ellison, C. A.; Evans, H. C.; Ooi, P. A. C. In *Proceedings of the X International Symposium on Biological Control of Weeds*, July 4−14, 1999; Spencer N. R., Ed.; Montana State University: Bozeman, Montana, 2000; pp 155−167.
(7) Xue, Y. M. "Rescue mangroves in Shenzhen", *Nanfang Ribao (Southern Daily)*, 2000, August 8, p D4.
(8) Stepp, J. R.; Moerman, D. E. *J. Ethnopharmcol.* **2001**, *75*, 19–23.
(9) Facey, P. C.; Pascoe, K. O.; Porter, R. B.; Jones, A. D. *J. Pharm. Pharmacol.* **1999**, *51*, 1455–1460.
(10) Huang, H.; Ye, W.; Wu, P.; Lin, L.; Wei, X. *J. Nat. Prod.* **2004**, *67*, 734–736.
(11) Cuenca, M. D. R.; Bardon, A.; Catalan, C. A. N.; Kokke, W. C. M. C. *J Nat. Prod.* **1988**, *51*, 625–626.
(12) Barrero, A. F.; Oltra, J. E.; Rodriguez-Garcia, I.; Barragan, A.; Alvarez, M. *J. Nat. Prod.* **2000**, *63*, 305–307.
(13) Bishayee, A.; Chatterjee, M. *Cancer Lett.* **1994**, *81*, 193–200.
(14) Fierro, I. M.; da Silva, A. C.; Lopes, C. S.; de Moura, R. S.; Barja-Fidalgo, C. *J. Ethnopharmacol.* **1999**, *66*, 19–24.
(15) Paul, R. K.; Jabbar, A.; Rashid, M. A. *Fitoterapia* **2000**, *71*, 701–703.
(16) Alves, T. M. A.; Chaves, P. P. G.; Santos, L. M. S. T.; Nagem, T. J.; Murta, S. M. F.; Ceravolo, I. P.; Romanha, A. J.; Zani, C. L. *Planta Med.* **1995**, *61*, 85–87.
(17) Rojas, D. A. A.; Ferro, E.; Inchausti, A.; Ascurra, M.; Acosta, N.; Rodriguez, E.; Fournet, A. *J. Ethnopharmcol.* **1995**, *45*, 35–41.
(18) Herz, W.; Subramaniam, P. S.; Santhanam, P. S.; Aota, K.; Hall, A. L. *J. Org. Chem.* **1970**, *35*, 1453–1464.
(19) Gutierrez, A. B.; Oberti, J. C.; Kulanthaivel, P.; Herz, W. *Phytochemistry* **1985**, *24*, 2967–2971.
(20) Cox, P. J.; Sim, G. A. *J. Chem. Soc., Perkin Trans. 2* **1974**, *1355*, 1359.
(21) Jiang, R. W.; But, P. P. H.; Ma, S. C.; Ye, W. C.; Chan, S. P.; Mak, T. C. W. *Tetrahedron Lett.* **2002**, *43*, 2415–2418.
(22) Tashkhodzhaev, B.; Abduazimov, B. K. *Chem. Nat. Compd.* **1997**, *33*, 382–388.
(23) Edil'baeva, T. T.; Turdybekov, K. M. *Chem. Nat. Compd.* **2001**, *37*, 143–147.
(24) Jiang, R. W.; Ming, D. S.; But, P. P. H.; Mak, T. C. W. *Acta Crystallogr.* **2000**, *C56*, 594–595.
(25) El-Sayed, N. H.; Norris, J. A.; Ahmed, A. A.; Mabry, T. J. *Phytochemistry* **1990**, *29*, 2364–2365.
(26) Rao, C. B.; Rao, T. N.; Suryaprakasam, S. *Planta Med.* **1976**, *29*, 391–392.
(27) Mercer, E. I.; Bartlett, K. *Phytochemistry* **1974**, *13*, 1099–105.
(28) Robert, B. C. In *Antiviral Agents and Human Viral Diseases: Respiratory Virus Infections*, 4th ed.; Galasso, G. J., Whitley, R. J., Merigan, T. C., Eds.; Lippincott-Raven Publishers: Philadelphia, 1997; pp 369−413.
(29) Li, Y. L.; But, P. P. H.; Ooi, V. E. C. *Antiviral Res.* **2005**, *68*, 1–9.

NP800542T