Pergamon

0031-9422(94)00626-1

Phytochemistry, Vol. 38, No. 2, pp. 437–442, 1995

0031-9422/95 $9.50 + 0.00

CYTOTOXIC *ENT*-KAURENE DITERPENOIDS FROM THREE *ISODON* SPECIES

HAN-DONG SUN, ZHONG-WEN LIN, FANG-DI NIU, LONG-ZE LIN,* HEE-BYUNG CHAI,* JOHN M. PEZZUTO* and GEOFFREY A. CORDELL*†

Laboratory of Phytochemistry, Kunming Institute of Botany, Academia Sinica, Kunming 650204, Yunnan Province, P.R. China; *Program for Collaborative Research in the Pharmaceutical Sciences, Department of Medicinal Chemistry and Pharmacognosy, College of Pharmacy, University of Illinois at Chicago, Chicago, IL 60612, U.S.A.

(*Received in revised form* 21 *June* 1994)

Key Word Index—*Isodon loxothyrsa*; *I. pleiophyllus*; *I. adenoloma*; Labiatae; *ent*-kaurene diterpenoids; loxothyrin A; adenolin B; longikaurin F; coetsoidin G; coetsoidin A; coetsoidin B; biological activities.

Abstract—From *Isodon loxothyrsa*, one new diterpenoid, loxothyrin A, together with one known diterpenoid, adenolin B, from *I. pleiophyllus*, three known diterpenoids, coetsoidins A, B and G, and from *I. adenoloma*, one known diterpenoid, longikaurin F, were isolated. The structure determination of loxothyrin A, and the unambiguous NMR spectral assignments of the known compounds were made by a combination of 1D and 2D NMR techniques and computer modelling calculations. The isolates showed potent cytotoxic activities.

INTRODUCTION

In a previous study of the chemical constituents of *Isodon loxothyrsa* (Hand.-Mazz.) H. Hara (Labiatae), we reported on the isolation of two new diterpenoids, rabdoloxins A and B [1]. Recently, we studied the same plant collected from a different location to yield loxothyrin A, a new diterpenoid, and adenolin B [2]. Meanwhile, we isolated the known compounds coetsoidins A, B and G [3] from *I. pleiophyllus*, and longikaurin F [4] from *I. adenoloma*. In this report, we present the isolation and structure elucidation of loxothyrin A, and the unambiguous assignment of the NMR spectral data by a combination of NMR techniques, including DEPT, COSY, ROESY [5–7, 9], HETCOR, FLOCK [8], HMBC [9, 10] and selective INEPT [11, 12], and computer modelling calculations, as well as the results of the biological evaluation of these diterpenes.

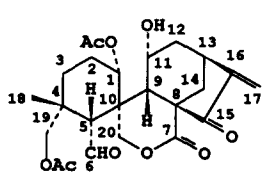
RESULTS AND DISCUSSION

An ethereal extract from the leaves of *I. loxothyrsa* was subjected to CC on silica gel, followed by recrystallization to yield loxothyrin A and adenolin B. In the same way, coetsoidins A, B and G were isolated from the leaves of *I. pleiophyllus*, and longikaurin F from the leaves of *I. adenoloma*.

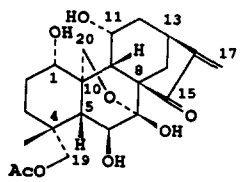
Loxothyrin A, $C_{22}H_{30}O_4$ (HRMS), crystals, mp 242–244°, showed IR and UV absorptions for the existence of a five-membered ring ketone conjugated with an exo-methylene, a six-membered lactone and acetyl groups (229 nm; 1740, 1735, 1730, 1710, 1690 and 1640 cm^{-1}) [3]. The ^1H, ^{13}C, DEPT NMR and HETCOR spectra of **1** showed signals for a B-*seco*-*ent*-kaurane-skeleton like rabdosichuanin A [3]; the differences are that **1** has one more acetyl, one more hydroxyl function, and one more exomethylene. The COSY and HETCOR spectra of **1** indicated the existence of the following fragments: $-CHCH_2CH_2-$, $-CHCH(OH)CH_2CHCH_2-$, $-CH_2O-$ and OHCCH <, each of which was connected to quaternary carbon atoms at both ends, suggesting that **1** had a B-*seco*-*ent*-kaurane skeleton with a secondary hydroxyl at C-11, an aldehyde at C-5, two acetyls at C-1 and C-19, and a δ-lactone ring at C-7 (20). The H-9 signal appeared as a broad signal at δ 2.63, and was coupled to only one proton (H-11); C-11 appeared as a methine carbon in the DEPT and HETCOR spectra, supporting the placement of the secondary hydroxyl function at the C-11 position. The above suggestions were further confirmed by a reverse detected proton–carbon long-range chemical shift correlation 2D NMR spectrum (HMBC), to assign the quaternary carbons, and thus to formulate the skeleton, and confirm the assignments of the protons and carbons.

In the HMBC [10] spectrum of **1** (Table 1), H-20a was coupled to C-7, C-9 and C-10, and H-20b was coupled to C-1, which served to establish the closure of the lactone ring between C-7 and C-20. The olefinic proton (H-17) signals were coupled to C-13, C-15 and C-16, the H_2-14

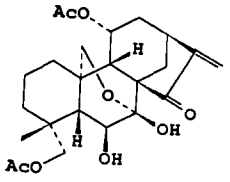
† Author to whom correspondence should be addressed.



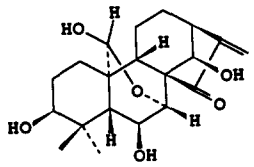
Loxothyrin A (1)



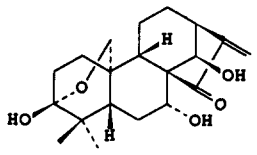
Adenolin B (2)



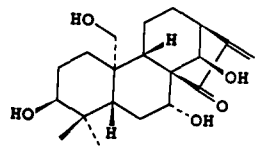
Longikaurin F (3)



Coetsoidin G (4)



Coetsoidin A (5)



Coetsoidin B (6)

Table 1. Major correlations from the ROESY and HMBC spectra of loxothyrin A (1)*†

Proton	ROESY (Proton)	HMBC (Carbon)
1β	2β, 3β, 5β, 11β	20, 1α-OCOMe
2α	3α, 19a, 20a	4, 10
2β	1β, 3α, 3β	4, 10
3α	2α, 2β, 18	n.o.
3β	1β, 2β, 18	n.o.
5β	1β, 3β, 6, 9β, 11β, 18	(4), 6, (10), 18, 19
6	5β, 9β, 18, 19a, 20b	5, 6.
9β	1β, 5β, 6, 11β, 12β, 20b	1, 7, (8), (10), 12
11β	1β, 5β, 9β, 12α, 12β	n.o.
12α	11β, 13α, 14α	9, (11), (13), 16
12β	9β, 11β, 13α	14, 16
13α	12α, 12β, 14β, 17b	8, 11, (12), (14), (16), 17
14α	1α-OAc, 12α	(8), 12, (13), 15, 16
14β	13α	(8), 9, 12, (13), 15, 16
17a	17b	13, 15, (16)
17b	13α, 17a	13, 15
18	5β, 6, 19b	3, (4), 5, 19
19a	2a, 6, 20a, 19b	(4), 18, 19-O-COMe
19b	18, 19a	(4), 5, 18, 19-OCOMe
20a	2α, 19a, 19b	7, 9, (10)
20b	6, 9β	1, 7, 9, (10)
1α-OCOMe	14α	1α-OCOMe
19-OCOMe	n.o.	19-OCOMe

* ROESY experiment was performed at 500.1 MHz with a spin-lock time of 300 msec, and a spin lock field strength of 5 kHz [14, 15].

† HMBC experiment was performed at 500.1/125.8 MHz with $J = 6$ Hz [14, 15]; two-bond correlations are put in parentheses; n.o. indicates no clear HMBC contours with this proton.

protons coupled to C-8, C-9, C-12, C-13, C-15 and C-16, and the H-9 proton coupled to C-1, C-7, C-8, C-10, C-12 and C-15, which led to a conclusion that the ketone function was at C-15, the double bond between C-16 and C-17, and that C-7, C-8, C-9, C-10 and C-20 constituted the six-membered lactone ring. Similarly, from the observations that H-5 was coupled to the aldehyde carbonyl, that H-1 was coupled to the acetyl carbonyl at δ170.2, and that the H_2-19 protons were coupled to another acetyl carbonyl signal at δ170.1, the two acetyl groups should be placed at the C-1 and C-19 positions, and the aldehyde function at C-5. Thus, all of the carbons could be unambiguously assigned, as shown in Table 2.

The principal results (Table 1) from the ROESY experiment [5–7] suggested that the isolate had the stereochemistry shown in Fig. 1. Based on the information from ^{1}H, COSY and ROESY spectra, a computer-assisted 3D structure (Fig. 2) was obtained using the molecular modelling program PCMODEL 386 V 4.0, using MMX force field calculations for energy minimization. This structure shows that the six-membered ring (A-ring), and the six membered lactone ring (B-ring) are in a deformed chair conformation, but that the other six-membered ring (C-ring) is in a boat conformation. The calculated distances between H-1β and H-3β (2.40 Å), H-1β and H-5β (2.40 Å), H-3β and H-5β (2.58 Å), H-5β and H-9β (2.97 Å), H-5β and H-11β (2.33 Å), H-5β and H-18 (2.33 Å), H-9β and H-12β (2.94 Å), H-13α and H-17b (2.58 Å), H-14α and 1α-OAc (2.54 Å), H-19a and H-20a (1.98 Å), H-20a and H-2α (2.21 Å), H-19a and H-2α (2.32 Å), H-20b and H-9β (2.32 Å), H-20b and H-6 (2.36 Å), are all less than 3.00 Å, which was consistent with the well-defined ROESY correlations observed between each of these proton pairs and confirmed the A-, B- and C-rings to occupy a chair, a chair and a boat conformation, respectively. The data also supported the α-orientation of the C-11 hydroxyl and C-1 acetyl functions. Thus, loxothyrin A (1) was identified as 1α,19-diacetyl-11α-hydroxy-6,15-dione-6,7-*seco-ent*-kaur-16-en-7,20-olide, with a conformation as shown in Fig. 2.

Molecular dynamics simulation also afforded information about the average dihedral angles and the corresponding J values between each of the vicinal proton pairs.

Table 2. ^{13}C NMR data of diterpenoids 1–6*

C	1	2	3	4	5	6
1	77.9	73.2	30.9	25.7	30.4	28.7
2	24.0	29.0	18.5	23.6	35.1	26.8
3	35.4	33.8	36.0	74.4	98.0	74.8
4	39.2	37.6	37.2	39.8	40.7	43.7
5	61.5	62.3	60.9	53.5	48.7	47.6
6	201.6	74.2	74.0	70.0	31.0	29.8
7	170.8	96.4	96.3	70.0	72.8	75.3
8	58.6	59.9	59.1	62.9	61.2	62.1
9	47.2	54.8	53.2	52.1	48.5	55.7
10	44.6	43.1	37.3	39.8	37.2	38.0
11	63.6	67.0	69.4	20.1	18.4	18.9
12	42.4	39.4	37.9	31.8	30.9	31.1
13	34.9	34.8	34.2	44.6	46.4	47.4
14	29.9	27.2	27.7	71.0	76.5	76.7
15	202.1	211.4	209.7	212.1	207.5	209.0
16	150.8	154.3	153.0	153.8	149.7	150.7
17	118.7	115.7	117.6	118.5	116.5	115.8
18	28.4	27.9	28.6	29.9	27.3	29.6
19	69.0	66.6	67.0	23.3	19.5	23.2
20	67.8	66.4	68.6	94.2	68.1	60.3
1-COMe	170.2	—	—	—	—	—
1-COMe	20.4	—	—	—	—	—
11-COMe	—	—	169.9	—	—	—
11-COMe	—	—	21.7	—	—	—
19-COMe	170.1	170.8	170.9	—	—	—
19-COMe	21.4	20.8	20.8	—	—	—

* Recorded in pyridine-d_5, chemical shift values reported as δ (ppm) from TMS at 125.8 MHz.

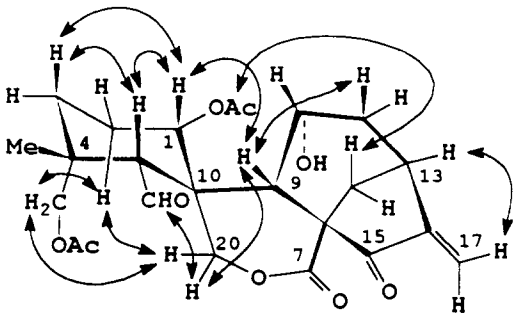


Fig. 1. Expression of the major ROESY correlations in loxothyrin A (1).

These data were used to check the J values from the NMR measurements, to assign the complex coupling patterns, and to determine the stereochemistry of the functional groups. The H-12 proton of 1 appearing in the rear of the plane should be H-12α, having J values of *ca* 1.65 Hz (dihedral angle of *ca* 79°) with H-11β, and 8.91 Hz (21°) with H-13α, but its partner H-12β has J = 5.48 Hz (34°) with H-11β, and J = 1.05 Hz (92°) with H-13α. In practice, the H-12 proton signal appearing at δ2.37 as a clear double doublet showing J = 9.5 Hz with H-13α, and J = 13.5 Hz with its geminal proton, should be assigned to H-12α; the remaining H-12 signal at δ1.66 (*m*), which overlapped with H-3α, should be assigned to its partner H-12β. Similarly, H-14α has a J = 1.48 Hz (73°) with H-13α, and H-14β has a J = 5.21 Hz (45°) with H-13α which served to assign the double doublet signal at δ2.54 (J = 11.5, 4.5 Hz) to H-14β, and the doublet signal at δ3.37 (J = 11.5) to H-14α. Analysis in the same way permitted all of the remaining coupled vicinal protons to be assigned stereotopically (Table 3).

Compounds **2**–**6** were identified as adenolin B (**2**), longikaurin F (**3**), coetsoidin G (**4**), coetsoidin A (**5**) and coetsoidin B (**6**) by direct comparison with authentic samples and reported data in refs [2–4]. Because their previous reported NMR data were not unambiguously assigned, their ^{1}H and ^{13}C NMR data were unambiguously assigned by the use of DEPT, COSY, ROESY, HETCOR, FLOCK and selective INEPT NMR techniques in the same way as described above.

Compounds **1**–**6** were subjected to anticancer, antimalarial and HIV RT inhibitory tests [16–19]; none of them showed any HIV RT inhibitory and antimalarial activity. However, **1**–**6** showed potent cytotoxic activity (Table 4).

EXPERIMENTAL

General. Mps are uncorr. ^{1}H, ^{13}C DEPT, COSY, DQF-COSY, ROESY, HETCOR and FLOCK spectra were taken on a GE OMEGA 500 instrument operating

Table 3. ^{1}H NMR data of diterpenoids **1–6***

Proton	1	2	3	4	5	6
1α	—	—	1.43 (*m*)	1.99 (*m*)	2.26 (*m*)	2.15 (*m*)
1β	5.78 (*dd*, 9.0, 9.0)	3.90 (*dd*, 8.3, 8.3)	1.26 (*m*)	1.93 (*m*)	2.00 (*m*)	1.52 (*m*)
2α	2.00–2.10 (*m*)	1.92 (*m*)	1.23 (*m*)	2.13 (*m*)	0.94 (*m*)	2.11 (*m*)
2β	2.00–2.10 (*m*)	1.86 (*m*)	1.29 (*m*)	2.18 (*m*)	2.04 (*m*)	1.79 (*m*)
3α	1.60 (*m*)	1.89 (*m*)	1.77 (*m*)	3.67 (*t*,2.5)	—	3.62 (*br*, *s*)
3β	1.34 (*m*)	1.24 (*m*)	1.00 (*m*)	—	—	—
5β	2.93 (*br s*)	1.70 (*d*,7.5)	1.56 (*d*,7.5)	2.08 (*d*, 8.0)	1.51 (*m*)	2.10 (*dd*, 12.5, 8.0)
6α	10.90 (*s*)	4.45 (*dd*, 11.0, 7.5)	4.37 (*dd*, 10.5, 7.5)	4.50 (*dd*, 8.0, 3.0)	2.18 (*m*)	2.35 (*dt*, 12.5, 2.5)
6β	—	—	—	—	2.18 (*m*)	2.14 (*m*)
7β	—	—	—	4.86 (*d*, 3.0)	4.55 (*dd*, 11.0, 5.0)	4.96 (*dd*, 12.0, 4.5)
9β	2.63 (*br*, *s*)	1.62 (*d*, 4.5)	1.75 (*d*, 5.0)	1.83 (*dd*, 13.0, 6.0)	1.51 (*m*)	1.76 (*d*, 9.0)
11α	—	—	—	2.99 (*dddd*, 13.0, 13.0, ,8.5, 4.5)	1.40 (*m*)	1.97 (*m*)
11β	4.64 (*m*)	4.53 (*t*, 4.5)	5.39 (*t*, 5.0)	1.53 (*m*)	1.34 (*m*)	1.43 (*ddd*, 12.0, 9.0, 7.0)
12α	2.37 (*dd*, 13.5, 9.5)	2.57 (*dd*, 15.0, 9.5)	2.45 (*dd*, 15, 9.5)	2.29 (*ddd*, 13.5, 8.5, 5.0)	1.46 (*m*)	2.81 (*ddd*, 12, 7.0, 5.0)
12β	1.66 (*m*)	1.69 (*m*)	1.73 (*m*)	1.40 (*m*)	1.55 (*m*)	1.63 (*ddd*, 12, 10.0, 7.0)
13α	3.07 (*dd*, 9.0, 4.5)	3.09 (*dd*, 9.5, 4.0)	3.01 (*dd*, 9.5, 4.5)	3.19 (*d*, 10)	3.12 (*br*, *s*)	3.27 (*br*, *s*)
14α	3.37 (*d*, 11.5)	3.59 (*d*, 11.5)	2.91 (*d*, 12.5)	5.74 (*br*, *s*)	4.98 (*s*)	5.67 (*s*)
14β	2.54 (*dd*, 11.5, 4.5)	2.51 (*dd*, 11.5, 4.0)	2.51 (*dd*, 12.5, 4.5)	—	—	—
17a	5.97 (*s*)	5.92 (*s*)	5.95 (*s*)	6.16 (*s*)	6.25 (*s*)	6.28 (*s*)
17b	5.36 (*s*)	5.27 (*s*)	5.30 (*s*)	5.45 (*s*)	5.31 (*s*)	5.34 (*s*)
18	1.02 (*s*)	1.41 (*s*)	1.32 (*s*)	1.57 (*s*)	1.16 (*s*)	1.14 (*s*)
19a	4.51 (*d*, 11.5)	4.75 (*d*, 11.0)	4.60 (*d*, 10.5)	1.09 (*s*)	1.28 (*s*)	0.99 (*s*)
19b	4.16 (*d*, 11.5)	4.42 (*d*, 11.0)	4.31 (*d*, 10.5)	—	—	—
20a	5.45 (*d*, 11.5)	5.19 (*d*, 10.0)	4.39 (*d*, 9.5, 2.5)	6.05 (*s*)	4.99 (*dd*, 8.5)	4.33 (*d*, 12.0)
20b	5.34 (*d*, 11.5)	4.32 (*d*, 10.0)	4.12 (*d*, 9.5)	—	3.85 (*dd*, 8.5, 1.5)	4.26 (*d*, 12.0)
1-OAc	2.11 (*s*)	—	—	—	—	—
11-OAc	—	—	2.05 (*s*)	—	—	—
19-OAc	1.87 (*s*)	1.91 (*s*)	1.90 (*s*)	—	—	—

* Recorded in pyridine-d_5, chemical shift values are reported as δ (ppm) from TMS at 500.1 HMz; signal multiplicity and coupling constants (Hz) are shown in parentheses.

at 500.1 MHz for ^{1}H and homonuclear 2D NMR spectra, 125.8 MHz for ^{13}C and DEPT spectra, and 500.1/125.8 MHz for heteronuclear 2D spectra with a long-range coupling constant of $J = 6$ Hz, using standard GE programs in pyridine-d_5 soln, and have been described in detail previously [13, 14].

Plant material. The plant materials of *I. loxothyrsa*, *I. pleiophyllus* and *I. adenoloma* were collected from Yunnan Province, P.R. China, in 1990, and identified by Prof. H.-W. Li. The voucher specimens of *I. loxothyrsa*, *I. pleiophyllus* and *I. adenoloma* are deposited in the Herbarium of the Department of Taxonomy, Kunming Institute of Botany, Academia Sinica, Kunming, P.R. China.

Extraction and isolation. The powdered air-dried leaves (4.29 kg) of *I. loxothyrsa* were extracted with Et_2O and the solvent removed under vacuum. The residue (635 g) was subjected to CC on silica gel, eluted with petrol, $CHCl_3$, $CHCl_3$–Me_2CO mixt. in increasing proportions of Me_2CO. Frs were collected, and combined by monitoring with TLC, followed by recrystallization to yield loxothyrin A (**1**, 205 mg, 0.00048%) and adenolin B (**2**, 750 mg, 0.0018%).

Treated in the same way, the dried leaves (1.44 kg) of *I. pleiophyllus* offered coetsoidin G (**4**, 100 mg, 0.0069%), coetsoidin A (**5**, 120 mg, 0.0083%) and coetsoidin B (**6**, 121 mg, 0.0084%), and the dried leaves (1.0 kg) of *I. adenoloma* offered longikaurin F (**3**, 210 mg, 0.021%). All of the known compounds were identified by direct comparison of their mp, mixt. mp, TLC, IR and ^{1}H NMR data with the authentic sample, and the reported data in refs [2–4], respectively.

Loxothyrin A (**1**). Obtained as crystals; mp 242–244°; $[\alpha]_D$ −76.3° (MeOH; *c* 0.05); UV λ_{max}^{MeOH} (logε): 229 (4.07) nm; IR ν_{max}^{KBr}: 3460, 1740, 1735, 1730, 1710, 1690, 1640, 1488, 1390, 1250, 1240, 1230, 1080, 1040 and 985 cm^{-1}: ^{1}H NMR data, see Table 3; ^{13}C NMR data, see Table 2; EIMS *m/z* (100%): 462 ([M]$^+$, 18), 425 (26),

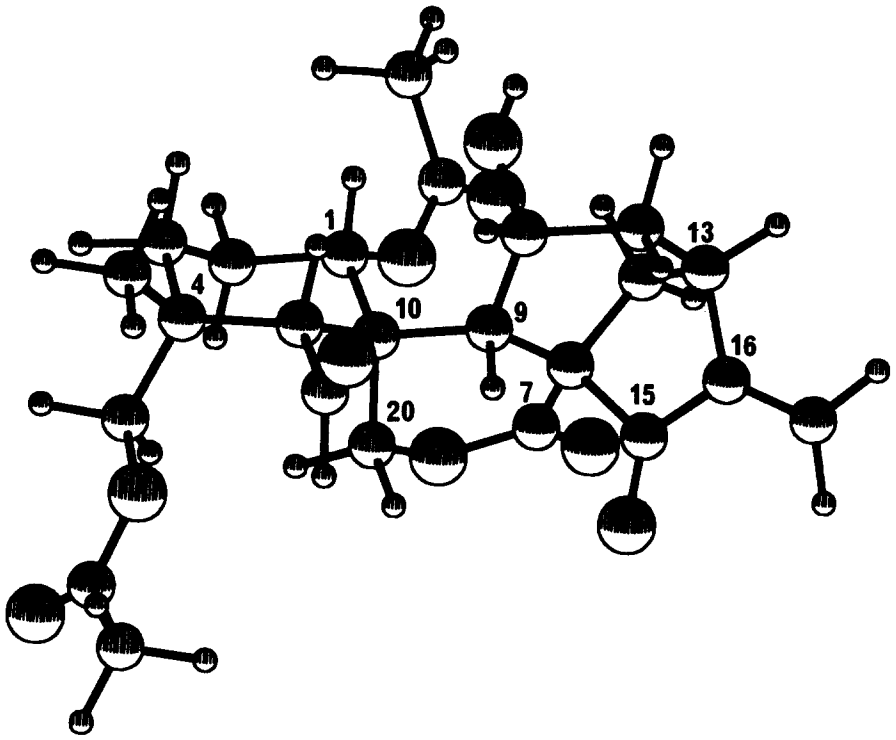


Fig. 2. Stereoview of loxothyrin A (**1**) generated from computer modelling.

Table 4. Evaluation of the cytotoxic activity of **1–6***

	Cell lines (ED_{50}, $\mu g\,ml^{-1}$)						
Compounds	Lu-1	KB	KB-V (+VLB)	KB-V (−VLB)	LNCaP	ZR-75-1	ASK†
1	>20	>20	>20	>20	13.5	7.2	neg.
2	>20	>20	>20	>20	8.9	4.8	neg.
3	0.1	1.1	14.2	>20	0.3	0.3	neg.
4	2.4	7.2	>20	>20	5.4	7.5	neg.
5	0.4	0.6	9.6	>20	0.7	0.8	neg.
6	2.7	4.4	>20	>20	3.7	4.8	neg.

* Lu-1 = human lung cancer, KB = human oral epidermoid carcinoma, KB-V = vinblastine-resistant KB evaluated in the presence (+ VLB) or absence (− VLB) of vinblastine (1 $\mu g\,ml^{-1}$), LNCaP = hormone dependent human Prostatic cancer, ZR-75-1 = hormone-dependent human breast cancer, ASK = rat astrocytoma.

† neg. = Negative, indicates no antimitotic activity with the ASK cell line.

424 (100), 342 (28), 284 (21), 283 (40), 248 (80), 207 (26), 203 (26), 162 (25), 149 (20), 149 (20), 133 (28), 119 (25), 109 (20), 107 (20), 105 (30), 97 (28), 95 (31), 91 (31), 84 (23), 83 (33), 81 (33), 71 (27), 69 (38), 57 (37) and 55 (45); HRMS: observed 462.1893 for $C_{24}H_{30}O_9$, calcd 462.1889.

Cytotoxicity, antimalarial and HIV-1 RT inhibitory assays. The biological evaluations for cytotoxic, antimalarial and HIV-1 RT inhibitory activities of these compounds were carried out according to established protocols [15–18].

Acknowledgements— This work was supported, in part, by grants from the Laboratory of Phytochemistry, Kunming Institute of Botany, Academia Sinica, P.R. China, to the Chinese research group, and a grant (CA-20164) from the Division of Cancer Treatment, National Cancer Institute, Bethesda, MD, U.S.A., to the U.S.A. research group. The authors thank Prof. H.-W. Li for the identification of the plant material, Mr R. Dvorak for MS data, Dr C. K. Angerhofer, Ms T. Pengsuparp, Ms V. Wongpanich, Ms M. You and Ms E. Pisha for the biological tests, Dr G. Doss (formerly A.N. Abdel-Sayed) for the initial implementation of the selective INEPT technique at UIC [19], and the Research Resources Center of the University of Illinois at Chicago for the provision of NMR spectroscopic facilities.

REFERENCES

1. Sun, H.-D., Lin, Z.-W., Shen, X.-Y., Takeda, Y. and Fujita, T. (1991) *Phytochemistry* **30**, 603.
2. Zhang, R.-P., Zhang, H.-L., Lin, Z.-W. and Sun, H.-D. (1992) *Phytochemistry* **31**, 4237.
3. Huang, H., Zhang, Y.-L. and Sun, H.-D. (1992) *Phytochemistry* **28**, 2753.
4. Fujita, T., Takeda, Y. and Shingu, Y. (1981) *Heterocycles* **16**, 227.
5. Rance, M., Sørnensen, O. W., Bodenhausen, G., Wagner, G., Ernst, R. R. and Wüthrich, K. (1983) *Biochem. Biophys. Res. Commun.* **117**, 479.
6. Bothner-By, A. A., Stephens, R. L., Lee, J., Warren, C. D. and Jeanloz, R. W. (1984) *J. Am. Chem. Soc.* **106**, 811.
7. Griesinger, C. and Ernst, R. R. (1987) *J. Magn. Reson.* **75**, 261.
8. Reynolds, W. F., McLean, S., Perpick-Dumont, M. and Enriques, R. G. (1989) *Magn. Reson. Chem.* **27**, 162.
9. Summers, M. E., Marzilli, L. G. and Bax, A (1986) *J. Am. Chem. Soc.* **108**, 4285.
10. Bax, A., Aszalos, A., Dinys, Z. and Suda, K. (1986) *J. Am. Chem. Soc.* **108**, 8056.
11. Bax, A. (1984) *J. Magn. Reson.* **57**, 314.
12. Cordell, G. A. and Kinghorn, A. D. (1991) *Tetrahedron* **47**, 3521.
13. Lin, L.-Z., Lin, L.-J., Cordell, G. A., Luo, S.-Q. and Jin, H.-F. (1992) *Magn. Reson. Chem.* **30**, 1097.
14. Luo, S.-Q., Lin, L.-Z., Cordell, G. A., Xue, L. and Johson, M. E. (1993) *Magn. Reson. Chem.* **31**, 215.
15. Lin, L.-Z., Shieh, H.-L., Angerhofer, C.K., Pezzuto, J. M., Cordell, G. A., Xue, L., Johnson, M. E. and Ruangrungsi, N. (1993) *J. Nat. Prod.* **56**, 22.
16. Likhitwitayawuid, K., Angerhofer, C. K., Chai, H., Pezzuto, J. M. and Cordell, G. A. (1993) *J. Nat. Prod.* **56**, 30.
17. Tan, G. T., Pezzuto, J. M. and Kinghorn, A. D. (1991) *J. Nat. Prod.* **54**, 143.
18. Tan, G. T., Pezzuto, J. M. and Kinghorn, A. D. (1994) in *Natural Products as Antivirus Agents* (Chuc and Cutler H., eds), Plenum Press (in press).
19. Abdel-Sayed, A. N. and Bauer, L. (1986) *Tetrahedron Letts* **27**, 1003.