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TETRAHEDRON

Styryl-Lactone Derivatives and Alkaloids from *Goniothalamus borneensis* (Annonaceae)

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Abstract: Twelve natural products, including two new compounds, goniothalesdiol **1a** and goniothalactam **2**, were isolated from the bark of the Malaysian tree *Goniothalamus borneensis* (Annonaceae) and evaluated for their cytotoxic activity. Their structures were elucidated by spectroscopy including 2D-NMR spectroscopy. **1a**, which was semisynthesised from (+)-goniothalenol (**3a**), was related to several cytotoxic styryl-lactones (**4** - **8**) also isolated from this species.

INTRODUCTION

Ethnobotanical uses of several species of the genus *Goniothalamus* are well known in Malaysia; many of these plants have provided bioactive acetogenins,[1,2] alkaloids,[3-5] styryl-lactones[6-10] and flavonoids.[11] Cytotoxic styryl-lactones or their derivatives, which have been reported in almost all the *Goniothalamus* species studied, are characteristic compounds of this genus. There has been continued interest in the cytotoxicity and synthesis of these and related compounds.[12,13] The alkaloids produced by many *Goniothalamus* species, being part of the Annonaceae family, are notably aporphines, some of which are known to be biologically active.[14] *Goniothalamus borneensis*, an exceptionally large tree of this genus, is found in Sabah, East Malaysia, and has been used as a mosquito repellent by the natives. Our chemical screenings[15] showed that this chemically unstudied plant contained cytotoxic and insecticidal principles. The bioassay-directed separation of the tree's bark extracts led to the isolation of seven cytotoxic compounds and five others, two of which are new natural products. The structure and stereochemistry of the new compounds were determined by IR, MS and NMR spectra as well as by chemical transformation and semisynthesis.

RESULTS AND DISCUSSION

Goniothalesdiol **1a** was isolated as a yellow oil, $[\alpha]_D^{25}$ +7.5 (c 0.23, EtOH). In the IR spectrum, absorption bands attributable to hydroxyl (3508 cm^{-1}) and carbonyl (1705 cm^{-1}) groups were present. The UV spectrum of **1a** had maxima at 236 and 280 nm. The EIMS showed only a weak parent ion ($M^{+\cdot}$ *m/z* 266, 0.01%) and the presence of two hydroxyl groups were confirmed by the preparation of a diacetate derivative (**1b**) which gave peaks at *m/z* 350 ($M^{+\cdot}$), 290 (M^+ - AcOH), 230(M^+ - 2AcOH) in the EIMS and two resonances at δ1.98 and 2.10 (3H each) in the ^1H NMR spectrum. The presence of a phenyl moiety was

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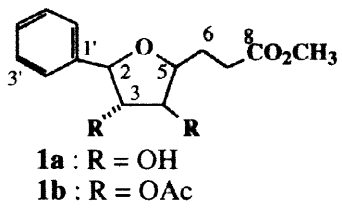
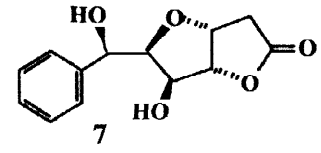
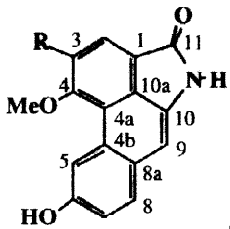
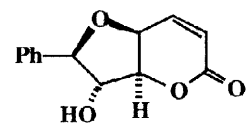
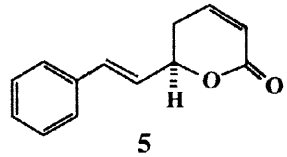
supported by ^{1}H NMR (δ 7.42, d, J = 7.0 Hz, H-2', 6' ; 7.35, t, J = 7.0 Hz, H-3', 5'; 7.28, d, J = 7.0 Hz, H-4') and ^{13}C NMR (δ 139.8, C-1'; 128.4, C-3', 5'; 127.7, C-4' and 126.0, C-2',6') (Table 1). Four oxygen-bearing carbons were also suggested by the ^{1}H NMR (δ 4.62, d, J = 4.5 Hz, H-2 and 4.08 - 4.17, m, H-3, 4 and 5) and ^{13}C NMR (δ 86.2, C-2; 85.2, C-3; 80.7, C-5 and 78.9, C-4). The ^{1}H NMR and ^{13}C NMR also showed the existence of two methylene groups (δ 2.56, m, H_2-6/δ 30.4, C-6; δ 2.12, m, H_2-7/δ 23.4, C-7). A singlet at δ 3.70 (3H) and a carbon resonance at δ 51.8 (C-9) indicated the presence of an -OMe group, and the resonance at δ 174.5 also suggested the existence of an ester group. With the molecular formula deduced as $C_{14}H_{18}O_5$ and the above spectral features, the structure **1a**, a substituted methyl tetrahydrofuranylpropanoate, may be assigned to goniothalesdiol.

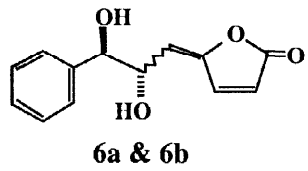












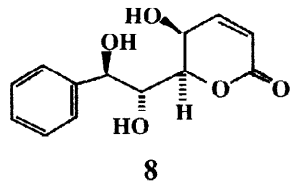


Table 1. ^{1}H and ^{13}C NMR data of **1a** ($CDCl_3$) and **2** (acetone-d_6)

H#	1a[a]	C#	1a[a]	H#	2[b]	C#	2[b]
2	4.62 d (4.5)[a]	2	86.2[c]	2	7.80 s	1	123.4
3 }		3	85.2[c]	5	8.77 d (2.6)	2	111.1
4 }	4.13 m	4	78.9[d]	7	7.16 dd (2.6, 8.6)	3	155.9
5 }		5	80.7[d]	8	7.75 d (8.6)	4	152.9
6	2.12 m	6	23.4	9	7.07 s	4a	121.9
7	2.56 m	7	30.4	NH	9.63 s	4b	129.6
2', 6'	7.42 d (7.0)	8	174.5	OMe (C-3)	4.12 s	5	113.9
3', 5'	7.35 t (7.0)	9	51.8	OMe (C-4)	4.10 s	6	157.1
4'	7.28 d (7.0)	1'	139.8			7	118.5
OMe	3.70 s	2', 6'	128.4			8	131.6
		3', 5'	126.0			8a	130.0
		4'	127.7			9	106.3
						10	134.4
						10a	126.0
						11	169.7
						OMe (C-3)	58.1
						OMe (C-4)	61.1

[a,b] **1a** obtained at 300 MHz; **2** at 500 MHz using TMS as internal reference; chemical shift δ, multiplicity (J Hz in parenthesis)
[c,d] assignments with same superscripts interchangeable

Further spectral evidence was required to confirm the structure and stereochemistry of **1a**. Because the peaks of H-3, H-4 and H-5 were overlapping considerably and appeared as a broad unresolved multiplet, **1a** was acetylated in order to make the assignments clearer. The HMBC spectrum of **1b** (Figure 1), which provided correlations from δ 4.90 (H-3) to δ 169.5 (C-10) and from δ 5.18 (H-4) to δ 169.5 (C-11), indicated that the two hydroxyl moieties of **1a** were located on C-3 and C-4. The observed three-bond correlation in the HMBC spectrum between, apparently, H-2 (δ 4.79) and C-2'/C-6' (δ 128.3) gave evidence that the phenyl moiety was located on C-2. The long-range correlations, C-2 (δ 84.0)/H-5 (δ 4.17) and C-5 (δ 79.3)/H-2 (δ 4.79), indicated that diol **1a** was a tetrahydrofuran derivative with a phenyl group among others.

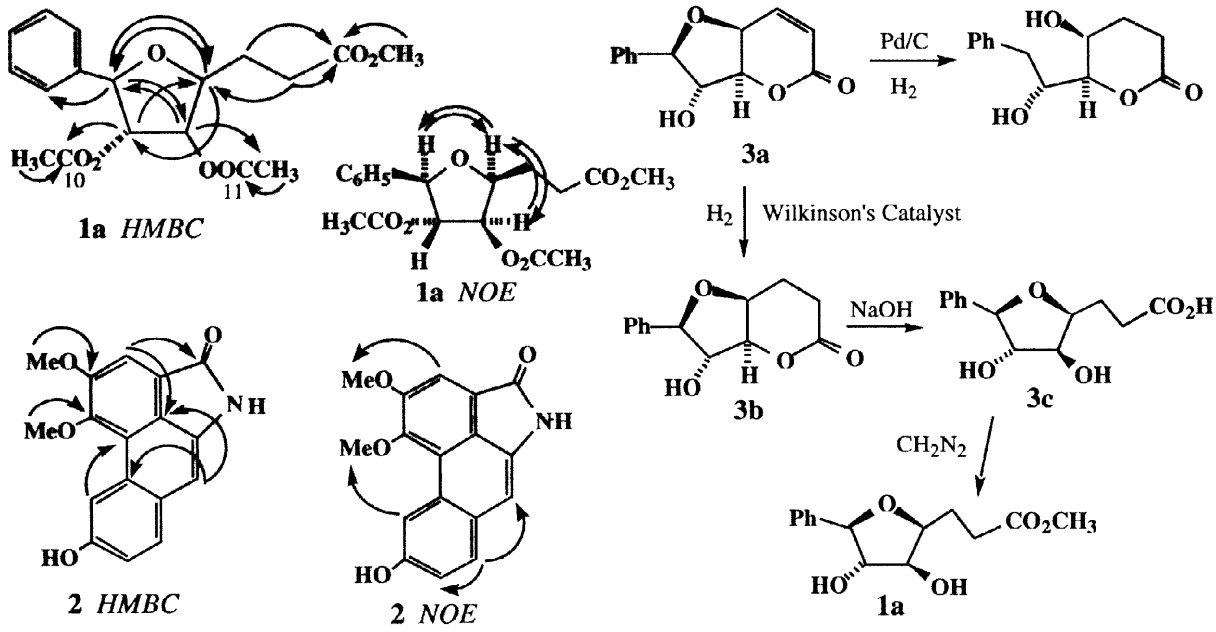


Figure 1. Selected HMBC and NOE Correlations of **1b** and **2**, and Semisynthesis of **1a**

Apart from the data of the IR absorption [1705 cm^{-1}], there was convincing evidence to support that **1a** was a methyl ester because in the HMBC spectrum of **1b** a cross peak was observed between the protons of the -OMe group (δ 3.59, 3H, s, OMe/δ 51.4, C-9) and the carbonyl group (δ 173.0, C-8). The ^1H-^1H COSY spectrum of **1b**, which clearly showed coupling correlations from H-2 *via* H-5, through H_2-6 to H_2-7 suggested that the substituent at C-5 on the furan ring was a 2-(methoxycarbonyl)ethyl group because the HMBC spectrum of **1b** provided correlations from H_2-6 (δ 1.93) (three-bond) and H_2-7 (δ 2.48, m) (two-bond) to C-8 (δ 173.0). The sprectral data indicated that goniothalesdiol is 2-phenyl-3,4-dihydroxy-5-(2-methoxycarbonylethyl)tetrahydrofuran (**1a**).

The relative stereochemistry of **1a** was assigned on the basis of NOE effects in the acetylated derivative **1b** (Figure 1). The assignment was confirmed by the synthesis of **1a** from (+)-goniothalenol (**3a**) (Figure 1). Hydrogenation of the double bond in the lactone ring with Pd/C opened the ring due to the cleavage of the benzylic ether bond. Also, bromination followed by tributyltin hydride reduction was unsuccessful. However, direct hydrogenation of (+)-goniothalenol with Wilkinson's catalyst gave the expected product, dihydrogoniothalenol (**3b**), which could be saponified by NaOH to provide the dihydroxy acid (**3c**). Esterification with diazomethane afforded the desired compound **1a**. Both the isolated natural product **1a** and the semisynthesised compound had the same rotation and identical NMR spectra.

A summary of some of the styryl-lactone derivatives and bioactivity are given in the Table 2. Some of these compounds were strikingly similar to those isolated from *Goniothalamus giganteus* of Thailand reported in a series of papers.[1,2,8-10] Goniothalesdiol **1a** is a new addition to this series and the structure as determined from spectral data and semisynthesis is (2*R*,3*R*,4*S*,5*S*)-2-phenyl-3,4-dihydroxy-5-(2-methoxycarbonylethyl)tetrahydrofuran. As goniothalesdiol **1a** was structurally similar to the other styryl-lactones but with the lactone ring opened, it may be suggested to be an artifact. However since the side chain on C-5 was saturated, it is probably not; thus this finding constitutes the first report of a 2-phenylfuranyl derivative from the genus *Goniothalamus*.

Goniothalactam **2**, with the molecular formula $C_{17}H_{13}NO_4$ as deduced by HREIMS, was a phenolic lactam, and its UV spectrum resembled closely that of aristolactam-AIII.[14] The prominent peaks (3226 and 1651 cm^{-1}) in the IR spectrum confirmed the presence of a lactam ring and are similar to those seen in the spectrum of aristolactam-AIII (**4**),[14] also isolated from this plant. The proton NMR spectrum exhibited signals at δ 4.10 (3H, s, OMe-C-4) and 4.12 (3H, s, OMe-C-3) indicative of two OMe groups. Apart from

two singlet protons at δ 7.07 (1H, s, H-9) and 7.80 (1H, s, H-2), it also showed the presence of an AMX coupling system with signals at δ 8.77 (1H, d, J = 2.6 Hz, H-5), 7.75 (1H, d, J = 8.6 Hz, H-8) and 7.16 (1H, dd, J = 2.6, 8.6 Hz, H-7). The proton signal at δ 9.63 (1H, s, NH) attributed to the NH appeared as a short broad singlet. The positions of the two methoxy groups were unambiguously established by recording the NOE difference spectra (Fig. 1). Irradiation of the signal at δ 8.77 (H-5) led to the enhancement of the signal at δ 4.10 (OMe-C-4). NOE relationships were also observed between the singlet at δ 7.80 (H-2) and the other methoxyl protons at δ 4.12 (OMe-C-3), and between the signals at δ 7.75 (H-8) and the signals at δ 7.07 (H-9)/7.16 (H-7). On the basis of above results, **2** was structurally established as 10-amino-6-hydroxy-3, 4-dimethoxy-phenanthrene-1-carboxylic acid lactam. Its structure was confirmed by the observed long range correlations in the HMBC spectrum as illustrated in Figure 1.

Table 2. Cytotoxicity Bioassay Results (ED_{50})[a]

Compound	P388	WEHI164	THP-1	MOLT4	Others
1a	>30	-[a]	-	-	-
2	>30	-	-	-	-
3a	>30	>30	>30	-	2[b]
4	5.2	24	29.5	4.3	-
Me-**4**[c]	10.7	18	4.95	2.25	-
5	0.75	1.70	-	<1	0.7-1[d]
6a	6.0	22	2.45	5.1	-
7	-	-	-	-	4.76[e]
8	9.2	27	30	8.1	-

[a] ED_{50} values in μg/ml, ED_{50} < 30 considered cytotoxic, if not determined no value (-) is given; P388= mouse leukemia, WEHI1640 = mouse fibrosarcoma, THP-1 = human monocytic leukemia, MOLT4 = lymphoblastic leukemia, human; [b] 9KB[6]; [c] Methylated **4**; [d] HGC-27, HeLa and MCF-7 cell lines[16]; [e] A-549[8]

Unlike aristolactam-AIII (**4**) which was strongly cytotoxic, **2** was only weakly active. Several of the other styryl-lactone derivatives isolated, however, had strong cytotoxic activities (Table 2). Among the family of various styryl-lactones found from *Goniothalamus* species, it can be seen that a large number of these compounds has been elaborated by *G. borneensis*. Compounds **3a**, **5**, **6a**, **6b**, **7** and **8** were also isolated previously from a closely related species *G. giganteus* found in Thailand.[10,17] This latter tree also furnished a prolific number of such compounds including isoaltholactone and 8-membered cyclic gonioheptolides.[10,17] Goniopypyrones as bicyclic Michael adducts were also elaborated by other *Goniothalamus* species.[18]

EXPERIMENTAL

General. A Reichert-Jung hot-stage microscope was used to measure melting points (uncorrected). EIMS were run on a Micromass VG 7035 mass spectrometer at 70 ev. NMR spectra were recorded by Bruker ACF 300 [300 MHz (^{1}H) and 75 MHz (^{13}C)] and AMX 500 [500 MHz (^{1}H) and 125 MHz (^{13}C)] instruments using $CDCl_3$, Me_2CO-d_6 or DMSO-d_6 solutions with TMS as an internal standard unless otherwise stated. IR spectra were recorded on a Bio-Rad FTIR spectrometer and UV spectra were recorded on a Hewlett Packard 8452A diode array spectrometer. Liquid chromatography was performed on silica gel (Kieselgel 60, particle size 0.040-0.063 mm) and Sephadex LH-20. TLC was run on silica gel pre-coated glass plates (Merck silica gel 60 F_{254}).

Plant material. The specimen of *Goniothalamus borneensis* was collected from Sabah, Malaysia, in June 1996 and identified by J. T. Pereira and L. Madani. Malaysian *Goniothalamus* species will be updated soon.[19] A voucher specimen (SAN 135154) was deposited at the herbarium of the Forest Research Centre,

Sepilok, Sandakan, Sabah, Malaysia.

Extraction and isolation. The dried and powdered bark material (2 kg) of *G. borneensis* was extracted with MeOH (24 h, 6L x 5) in a Soxhlet apparatus. The MeOH extract was evaporated under reduced pressure and the residue (304 g) was suspended in 5% MeOH/H_2O and extracted successively with hexane and chloroform to provide two portions of crude extracts after removal of solvents.

Stigmasterol was isolated as the major component in the hexane extract. The chloroform extract (183 g) was chromatographed on silica gel (5 kg) and eluted with hexane-acetone (1:20 to 1:1) system to give eight fractions. Fraction 1 (about 50 g) was mainly pinocembrin (40 g, 2%), while fraction 2 (about 60 g) was an optically active oil whose major component was (+)-goniothalenol (**3a**, 50 g, 2.5%) and also a minor amount of cinnamyl cinnamate (20 mg, 1 x 10^{-5} %). Fraction 3 (about 30 g) was chromatographied on silica gel (1.5 kg) with hexane-EtOAc (20:1 to 1:1) as the eluent, and the fractions were further subjected to sephadex LH-20 column chromatography followed by preparative silica gel TLC to yield six compounds: goniothalamin (**5**, 30 mg, 1.5 x 10^{-5} %), goniothalesdiol (**1a**, 60 mg, 3 x 10^{-5} %), goniobutenolide A (**6a**, 10 mg, 5 x 10^{-6} %) and B (**6b**, 10 mg, 5 x10^{-6} %), goniofufurone (**7**, 10 mg, 5 x 10^{-6} %) and goniotriol (**8**, 50 mg, 2.5 x 10^{-5} %). Fraction 4 (20 g) was chromatographed on silica gel and eluted with hexane-EtOAc (10:1 to 1:1) to furnish two alkaloids, goniothalactam (**2**, 10 mg, 5 x 10^{-6} %) and aristolactam A-III (**4**, 10 g, 0.005%).

Goniothalesdiol (1a). An optically active oil; $[\alpha]_D^{25}$ +7.5° (EtOH, c 0.23); UV (EtOH) λ_{max} (nm) 222 (2.80), 258 (2.30), 302 (1.80); IR (film) ν_{max} (cm^{-1}) 3508, 2973, 1705, 1450. 1360, 1239, 1175, 1046, 949, 917, 759, 699; EIMS *m/z* (rel. int.) 266 [M^+](0.01), 234 (56), 216 (44), 206 (22), 188 (50); HR-EIMS [M^+] *m/z* 266.1151 (calcd. for $C_{14}H_{18}O_5$, 266,1154); the ^1H and ^{13}C NMR spectral data are listed in Table 1.

Goniothalesdiol diacetate 1b. 1a (20 mg) was dissolved in acetic anhydride: pyridine (1:1, 5 ml) and the reaction mixture left at room temperature for 12 hr. Workup with water, extraction with chloroform and purification by silica gel PTLC with hexane: EtOAc (5:1) gave an optically active oil (**1b**, 22 mg, 85%). $[\alpha]_D^{25}$ +6.9° (EtOH, c 4.9); EIMS *m/z* (rel. int.) 350 [M^+] (0.2), 319 (5), 290 (7), 248 (10), 231 (50), 204 (8); HR-EIMS [M]$^+$ *m/z* 350.1337 (calcd. for $C_{18}H_{22}O_7$, 350.1366); ^1H NMR δ (ppm) (500 MHz) (DMSO-d_6): 1.93 (2H, m, H_2-6), 1.98 (3H, s, OAc), 2.10 (3H, s, OAc), 2.48 (2H, m, H_2-7), 3.59 (3H, s, OMe), 4.17 (1H, ddd, *J* = 3.6, 12.0, 5.0 Hz, H-5), 4.79 (1H, d, *J* = 3.6 Hz, H-2), 4.90 (1H, *J* = 1.2, 3.6 Hz, H-3), 5.18 (1H, dd, *J* = 1.2, 3.6 Hz, H-4), 7.30 (1H, d, *J* = 7.2 Hz, H-4'), 7.36 (2H, t, *J* = 7.2 Hz, H-3', 5'), 7.39 (2H, d, *J* = 7.2 Hz, H-2', 6'); ^{13}C NMR (125 MHz) (DMSO-d_6): δ 20.4 [OMe (C-10 or C-11)], 20.7 [OMe (C-11 or C-10)], 23.5 (C-6), 30.0 (C-7), 51.4 (C-9), 77.2 (C-4), 79.3 (C-5), 83.2 (C-2), 84.0 (C-2), 126.3 (C-3', 5'), 127.8 (C-4'), 128.3 (C-2', 6'), 139.2 (C-1'), 169.5 (C-10 and C-11), 173.0 (C-8).

Goniothalactam (2). Light yellow needles, m.p. 257-259 °C; UV (EtOH) λ_{max} (nm) 236 (4.58), 256 (4.45), 262 (4.48), 294 (4.35), 320 (4.21), 342 (4.10) and 400 (3.90); IR (film) ν_{max} (cm^{-1}) 3394, 3226, 1651, 1611, 1503, 1457, 1408, 1376, 1308, 1241, 1059, 1013, 951, 872; EIMS *m/z* (rel. int.) 295 [M^+](100), 280 (12), 265 (44), 252 (40); HR-EIMS [M]$^+$ *m/z* 295.0846 (calcd. for $C_{17}H_{13}NO_4$, 295.0845); the ^1H and ^{13}C NMR spectral data are listed in Table 1.

Preparation of 1a from 3a. A 100 mg sample of (+)-goniothalenol was hydrogenated using Wilkinson's catalyst (10 mg) in anhydrous EtOAc at room temperature and at atmospheric pressure for two hours. The reaction mixture was chromatographed on silica gel eluted with hexane-EtOAc (3: 1) and furnished compound **3b** (80 mg, 80%). **3b** (20 mg) was further hydrolysed with NaOH to give compound **3c** (20 mg, 90%). The latter (10 mg) was esterified with excess CH_2N_2 to yield **1a** (10 mg, 90%). Compound **3b**: an optically active oil; $[\alpha]_D^{25}$ +14.2° (EtOH, c 0.55); UV (EtOH) λ_{max} (nm) 222 (2.81), 258 (2.28), 286 (1.76); IR (film) ν_{max} (cm^{-1}) 3419, 2889, 1746, 1457, 1363, 1275, 1056; EIMS *m/z* (rel. int.) 234 [M^+] (12), 216 (11), 147 (66), 107 (76), 91 (64), 77 (100); HREIMS [M]$^+$ *m/z* 234.0902 (calcd. for $C_{13}H_{14}O_4$, 234.0892); ^1H NMR δ (ppm) (300 MHz) ($CDCl_3$): 2.17 (2H, m, H_2-4), 2.44 (1H, m, Ha-3), 2.67 (1H, m, Hb-3), 4.17 (1H, dd, *J* = 6.0, 2.1 Hz, H-7), 4.43 (1H, dt, *J* = 4.9 Hz, H-5), 4.61 (1H, d, *J* = 6.0 Hz, H-8), 4.74 (1H, dd, *J* = 4.9, 2.1 Hz, H-6), 7.28-7.37 (5H, m, H-2'-6'); ^{13}C NMR (75 MHz) ($CDCl_3$): d 22.9 (C-4), 26.1 (C-3), 72.2 (C-6), 84.1 (C-5), 85.5 (C-7), 88.2 (C-8), 126.0 (C-4'), 128.5 (C-3', 5'), 128.6 (C-2', 6'), 138.5(C- 1'), 170.9 (C-2).

Compound 3c. An optically active oil; $[\alpha]_D^{25}$ +11.6° (EtOH, c 0.28); UV (EtOH) λ_{max} (nm) 224 (2.74), 264 (2.23), 284 (2.02); IR (film) ν_{max} (cm^{-1}) 3297, 1746, 1241, 1167, 1086, 1039, 965, 905; EIMS

m/z (rel. int.) 252 [M$^+$](0.1), 234 (14), 216 (12); ^{1}H NMR δ (ppm) (300 MHz) (CD_3OD): 2.04 (2H, m, H_2-6), 2.44 (2H, m, H_2-7), 3.30 (3H, s, OMe), 3.94 (1H, dd, J = 4.7, 1.9 Hz, H-3), 4.01 (2H, m, H-4 and 5), 4.52 (1H, d, J = 4.7 Hz, H-2), 7.24 (1H, d, J = 7.4, H-4'), 7.31 (2H, t, J = 7.4 Hz, H-3', 5'), 7.44 (2H, d, J = 7.4 Hz, C-2', 6'); ^{13}C NMR (75 MHz) (CD_3OD): δ 25.7 (C-6), 33.1 (C-7), 79.9 (C-4), 82.7 (C-5), 86.6 (C-3), 88.1 (C-2), 127.6 (C-3', 5'), 128.5 (C-4'), 129.2 (C-2', 6'), 142.1 (C-1'), 175.0 (C-8).

Compounds 4 - 8. These compounds were identified from their NMR spectra and compared to the reported data.[1,2,6,8-10] Stigmasterol, pinocembrin and cinnamyl cinnamate had EIMS and NMR spectral data identical to those reported.[8,11]

Bioassays. The following cell lines were used: P388 (mouse lymphocytic leukemia), WEHI1640 (Mouse fibrosarcoma), THP-1 (human monocytic leukemia), MOLT4 (human lymphoblastic leukemia). Cell survival was evaluated by using MTT-tetrazolium assay as described previously.[20] Results are given in Table 2; according to the criterion set by the National Cancer Institute, USA, ED_{50} values of less than 30 mg/ml are considered cytotoxic.[21]

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REFERENCES

1. Gu, Z. M.; Fang, X. P.; Zeng, L.; McLaughlin, J. L. *Tetrahedron Letters* **1994**, *35*, 5367-5368.
2. Fang, X. P.; Anderson, J. E.; Smith, D. L.; Wood, K. V.; McLaughlin, J. L. *J. Nat. Prod.* **1992**, *55*, 1655-1663.
3. Talapatra, S. K.; Basu, D.; Chattopadhyay, P.; Talapatra, B. *Phytochemistry* **1988**, *27*, 903-906.
4. Din, L. B.; Colegate, S. M.; Razak, D. A. *Phytochemistry* **1990**, *29*, 346-348.
5. Omar, S.; Chee, C. L.; Ahmad, F.; Ni, J. X.; Jaber, H.; Huang, J.; Nakatsu, T. *Phytochemistry* **1992**, *31*, 4395-4397.
6. El-Zayat, A. A. E.; Ferrigni, N. R.; McCloud, T. G.; McKenzie, A. T.;Byrn, S. R.; Cassady, J. M.; Chang, C. J.; McLaughlin, J. L. *Tetrahedron Letters* **1985**, *26*, 955-956.
7. Sam, T. W.; Chew, S. Y.; Matsjeh, S.; Gan, E. K.; Razak, D.;Mohamed, A. L. *Tetrahedron Letters* **1987**, *28*, 2541-2544.
8. Fang, X. P.; Anderson, J. E.; Chang, C. J.; Fanwick, P. E.; McLaughlin, J. L. *J. Chem. Soc. Perkin Trans. 1* **1990**, 1655 -1661.
9. Fang, X. P.; Anderson, J. E.; Chang, C. J.; Mclaughlin, J. L. *Tetrahedron* **1991**, *4*, 9751-9758.
10. Fang, X. P.; Anderson, J. E.; Qiu, X. X.; Kozlowski, J. F.; Chang, C. J.; Mclaughlin, J. L. *Tetrahedron* **1993**, *49*, 1563-1570.
11. Talapatra, B.; Deb, T.; Talapatra, S. K. *Indian Journal of Chemistry* **1985**, *24B*, 561.
12. Surivet, J. P.; Vatele, J. M. *Tetrahedron Letters* **1996**, *37*, 4373-4376.
13. Mukai, C.; Hirai, S; Kim, I. J.; Kido, M.; Hanaoka, M. *Tetrahedron* **1996**, *52*, 6547-6560.
14. Mix, D. B.; Guinaudeau, H.; Shamma, M. *J. Nat. Prod.* **1982**, *45*, 657-666.
15. S. H. Goh et al, unpublished; Goh, S. H.; Lee, K. H.; Chuah, C. H.; Ong, H. C.; Madani, L.; Pereira, J. T. *J. Herbs, Spices & Med. Plants* **1997**, *5*, 29-51.
16. Ali, A. M.; Mackeen, M.M.; Mamid, M.; Aun, O. B.; Zaugah, Y.; Azimahtol, H. L. P.; Kawazu, K. *Planta Medica* **1997**, 63, 81-83.
17. Colegate, S. M.; Din, L. B.; Latiff, A.; Salleh, K. M.; Samsudin, M. W.; Skelton, B. W.; Tadano, K.; White, A. H; Zakaria, Z. *Phytochemistry* **1990**, *29*, 1701-1704.
18. Goh, S. H.; Ee, G. C. L.; Chuah, C. H.; Chen Wei *Aust. J. Chem.* **1995**, *48*, 199-205.
19. Kamarudin Mat Saleh (Universiti Kebangsaan Malaysia), *Private communication*.**1997**.
20. Mossman, T. *J. Immun. Methods* **1983**, *65*, 55-63.
21. Geran, R. I.; Greenberg, N. H.; MacDonald, M. M.; Schumacher, A. M.; Abbott, B. *J. Cancer Chemotherapy Reports Part 3*, **1972**, 3(2), *1-17*, 59-61.