FOUR NEW 4-PHENYLCOUMARINS FROM CALOPHYLLUM DISPAR ISOLATION AND HEMISYNTHESIS

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Abstract- Four new 4-phenylcoumarins, disparinols A and B, isodisparinols A and B, having a 2-hydroxy-3-methylbut-3-enyl substituent were isolated from the bark of Calophyllum dispar (Clusiaceae). Their structures were established by means of spectroscopic studies and confirmed by hemisynthesis.

INTRODUCTION

The Clusiaceae (ex-Guttiferae) represent a botanical family known as an abundant source of xanthones,[1] biflavonoids[2] and coumarins.[3] Since the discovery of the anti-HIV calanolides isolated from a Malayan Calophyllum lanigerum,[4] this genus has been considered as a potential source of new bioactive compounds. As part of our cooperative phytochemical investigations on the Malayan flora,[5] we now report here results from our bioassay-directed fractionation of an ethyl acetate extract of the stem bark of C. dispar.

RESULTS and DISCUSSION

An ethyl acetate extract of the stem bark of C. dispar was selected for its cytotoxic activity towards KB cells. Bioassay-directed[6] chromatography of this extract then leads to the isolation of new regio-isomeric coumarins (**1-4**) exhibiting the same molecular ion on their high-resolution MS as well as strong similarities in their UV, IR and NMR data.

Compound (**1**) crystallized as yellow needles (mp = 115-116°C, hexane/ethyl acetate) and had the molecular formula $C_{25}H_{26}O_6$ (found 422.1726, calcd 422.1729) determined by HREIMS. UV and IR spectra of **1** exhibited λ (Table 2) and ν values (1700, 1622, 1580, 1559, 756, and 697 cm^{-1}) closely matching the ones reported for coumarin mammeisin (**5**).[7] Furthermore, the UV spectrum of **1** (Table **2**) underwent a bathochromic shift after addition of NaOH which was typical of a 6-acyl-5,7-dihydroxycoumarin.[8] Two phenolic hydroxyls, one of them being strongly chelated by the carbonyl function of the acyl group, were then in evidence at δ_H 14.43 and 10.18 ppm on the ^{1}H-NMR spectrum (500 MHz, $CDCl_3$, TMS = 0) of **1**. In agreement with the observations noticed by Crombie et al.,[9] these hydroxyl groups were thus located at the 5 and 7 positions of the benzopyrane ring. This fact was corroborated, on the ^{13}C-NMR spectrum, by the low field resonances of three carbons at δ_C 163.3, 162.5 and 157.7 ppm and the high field resonances of three others at δ_C 107.8, 104.7 and 102.0 ppm. Indeed, these shielding and deshielding effects could be explained by two meta oxygenated functions in the 5,7 position of the heterocycle.

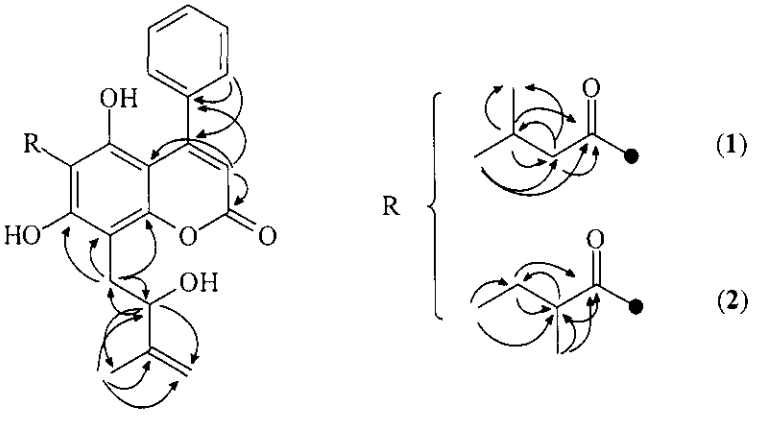
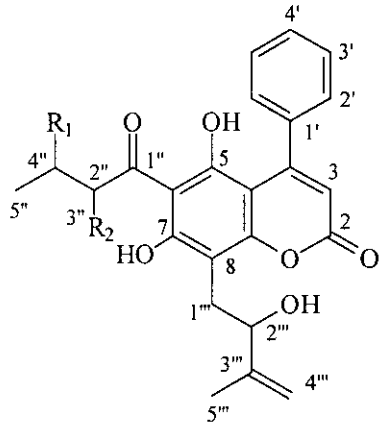
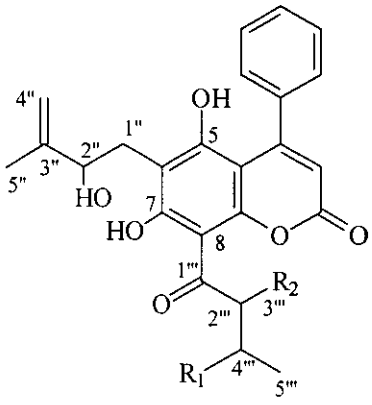


Figure 1 : Main HMBC (J = 6 Hz) correlations of compounds (**1**) and (**2**)

Additional signal at δ_c 207.7 ppm confirmed the presence of a carbonyl function. Examination of the contour map of a gradient-selected DQF-COSY experiment then allowed to distinguish two different spin systems in the molecule. On one hand, an isobutyl moiety was characterized with chemical shifts at δ_H : 0.94 (6H, dd, J = 2.0 and 6.5 Hz), 2.23 (1H, m) and 3.02 ppm (2H, d, J = 6.5 Hz). Long-range ^{1}H-^{13}C couplings deduced from the HMBC spectrum of **1** (Figure 1) then revealed that this fragment was linked to the coumarinic heterocycle via the carbonyl function. The corresponding acyl chain was thus placed at position 6 as explained above.





(**1**) : $R_1 = CH_3$ $R_2 = H$ (**3**) : $R_1 = CH_3$ $R_2 = H$

(**2**) : $R_1 = H$ $R_2 = CH_3$ (**4**) : $R_1 = H$ $R_2 = CH_3$

	δ (ppm) and J (Hz)							
	1		**2**		**3**		**4**	
2		160.0		159.9		159.1		159.2
3	5.96 s	112.1	5.97 s	112.2	6.03 s	112.2	6.04 s	112.2
4		156.7		156.7		156.4*		156.4*
4a		102.0		102.1		102.2		102.2
5	OH 14.43 s	163.3	OH 14.33 s	163.4	OH 9.29 s	160.3	OH 9.22 s	160.2
6		107.8		107.4		109.8		109.9
7	OH 10.18 s	162.5	OH 10.20 s	162.1	OH 14.80 s	166.9	OH 14.80 s	167.0
8		104.7		104.7		104.1		103.7
8a		157.7		157.7		156.5*		156.3*
1'		139.3		139.4		139.8		139.8
2'- 6'	7.33 m	127.1	7.33 m	127.2	7.32 m	127.1	7.32 m	127.1
3'- 5'	7.42 m	127.7	7.41 m	127.7	7.41 m	127.8	7.41 m	127.8
4'	7.42 m	128.2	7.41 m	128.2	7.41 m	128.2	7.41 m	128.2
1''		207.7		212.2	2.77 dd (8.0/15.0)	28.6	2.78 dd (8.5/15.0)	28.6
					3.13 dd (2.0/15.0)		3.12 dd (2.0/15.0)	
2''	3.02 d (6.5)	53.5	3.84 m	46.6	4.32 br d (8.0)	76.8	4.32 br d (8.0)	76.8
3''			1.14 d (6.5)	16.4		146.4		146.4
4''	2.23 m	25.1	1.39 m	26.8	4.86 s	110.6	4.87 s	110.6
			1.79 m		4.93 s		4.94 s	
5''	0.94 dd (6.5/2.0)	22.7	0.90 t (7.0)	11.9	1.81 s	18.4	1.82 s	18.4
1'''	3.04 dd (8.0/15.5)	28.8	3.06 dd (15.0/7.5)	28.7		206.1		210.6
	3.32 dd (2.0/15.0)		3.30 dd (2.0/15.0)					
2'''	4.50 br d (7.5)	77.5	4.51 br d (6.0)	77.7	3.20 dd (1.5/6.5)	53.5	3.99 m	46.8
3'''		146.0		146.0			1.29 dd (8.0/6.5)	16.6
4'''	4.94 s	111.1	4.93 s	111.1	2.32 m	25.7	1.53 m	27.2
	5.03 s		5.02 s				1.95 m	
5'''	1.93 s	18.7	1.94 s	18.8	1.06 d (7.0)	22.7	1.03 t (7.5)	11.8

* resonances may be interchangeable

Table 1 : ^1H and ^{13}C NMR data taken in $CDCl_3$ of compounds (**1-4**)

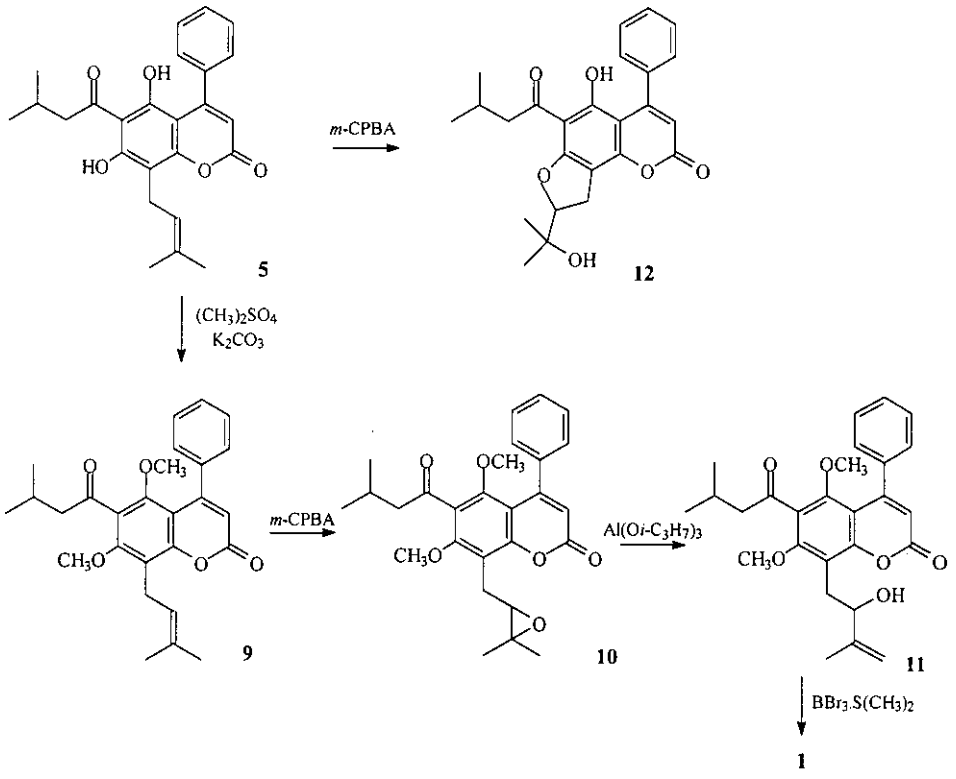
On the other hand, two singlets at δ_H 4.94 and 5.03 ppm were associated to a sp^2 methylene group, this fact being confirmed on the HMQC spectrum which showed that these protons were directly coupled with one carbon resonating at δ_c 111.1 ppm. The other elements of this spin system, deduced from the DQF-COSY, HMQC and HMBC data of **1**, were identified as an allylic methyl group [δ_H 1.93 (3H, s), δ_C 18.7 ppm], a secondary hydroxy group [δ_H 4.50 (1H, br d, 7.5 Hz), δ_C 77.5 ppm], a quaternary carbon at 146.0 ppm and a CH_2 [δ_H 3.04 (1H, dd, J= 8.0 and 15.5 Hz) and 3.32 (1H, dd, J= 2.0 and 15.0 Hz), δ_C 28.8 ppm] linked to the coumarin at the 8-position (Figure 1). The presence of the 2-hydroxy-3-methylbut-3-enyl moiety was confirmed by the observation of a base peak at m/z 351 ($C_{21}H_{19}O_5$) in the HREIMS of **1**. This fragment was attributed to the loss of an 1-hydroxy-2-methylprop-2-enyl radical by β-fission. In addition, absorptions at 756 and 697 cm^{-1} on the IR spectrum revealed the presence of a monosubstituted phenyl group and correlatively, five aromatic protons were in evidence at δ_H 7.33 (2H, m) and 7.42 ppm (3H, m) in the ^{1}H-NMR spectrum of **1**. Therefore **1** could be identified as 5,7-dihydroxy-8-(2'''-hydroxy-3'''-methylbut-3'''-enyl)-6-(3''-methyl-1''-oxobutyl)-4-phenyl-2H-benzopyran-2-one which we named disparinol A.

Compound		λ_{max} nm (log ε)					
1	a	242 (4.09)		284 (4.42)	341 (4.00)		
	b	244 (4.21)		302 (4.21)		381 (4.05)	412 (4.07)
2	a	235 (4.00)		284 (4.24)	340 (3.82)		
	b	243 (4.09)		302 (4.07)		382 (3.92)	409 (3.93)
5	a	236 (4.13)		285 (4.28)	332 (3.85)		
	b	240 (4.21)		303 (4.15)		394 (3.88)	428 (4.01)
3	a	226 (4.38)		292 (4.32)	328 (4.10)		
	b	230 (4.29)	263 (4.08)		336 (4.39)		
4	a	228 (4.21)		289 (4.04)	328 (3.71)		
	b	229 (3.96)	262 (3.69)		335 (4.03)		
7	a	227 (4.01)		296 (3.92)	333 (3.81)		
	b	230 (4.21)	261 (3.94)		337 (4.39)		

Table 2 : UV data of compounds (**1**-**7**), a : in EtOH-HCl and b : in EtOH-NaOH

The structural hypothesis was finally confirmed through an hemisynthesis of **1** using mammeisin (**5**) as a starting material (Scheme 1). Oxidation of the prenyl moiety of **5** in a 2-hydroxy-3-methylbut-3-enyl group was achieved by an epoxide rearrangement following a procedure developped by Terao et al.[10]
It should be noticed that the 7-OH function of mammeisin had to be protected in order to prevent the formation of a 2-(1-hydroxy-1-methylethyl)dihydrofuran ring (**12**).[8] Epoxidation of the dimethyl ether

(**9**)[11] with m-CPBA afforded **10** wich was rearranged in **11** bearing the desired moiety. The deprotection of the methyl ether group led to a compound identical with **1** (TLC, ^{1}H and ^{13}C NMR).



Scheme 1 : Hemisynthetic pathway of compound (**1**)

Compound (**2**) was isolated as an oil with IR data closely matching that of **1**. HREIMS revealed the same molecular formula $C_{25}H_{26}O_6$ (found 422.1747, calcd 422.1729) and the same base peak at m/z = 351 ($C_{21}H_{19}O_5$). Compound (**2**) also exhibited alkaline shifts (Table 2) usually associated with a 6-acyl-5,7-dihydroxycoumarin chromophore. The ^{1}H -NMR data (Table 1) of this compound though revealed that **1** and **2** differed from each other in a methyl position on their acyl-chain. Indeed, interlocking DQF-COSY, HMQC and HMBC (Figure 1) data, an isopentanoyl moiety was characterized with proton chemical shifts at δ_H : 0.90 (3H-5'', t, J = 7.0 Hz), 1.39 (1H-4'', m), 1.79 (1H-4'', m), 1.14 (3H-3'', d, J = 6.5 Hz), 3.84 ppm (1H-2'', m) and carbon chemical shifts at δ_C : 11.9 (C-5''), 16.4 (C-3''), 26.8 (C-4''), 46.6 (C-2''), 212.2 ppm (C-1''). A second substituent corresponding to a 2-hydroxy-3-methylbut-3-enyl moiety was finally identified with δ_H and δ_C values almost superimposable with the ones observed for **1** (Table 1).

On the basis of these results, the structure of **2** was identified as 5,7-dihydroxy-8-(2'''-hydroxy-3'''-methylbut-3'''-enyl)-6-(2''-methyl-1''-oxobutyl)-4-phenyl-2H-benzopyran-2-one which we have named disparinol B.

HREIMS analysis of compounds (**3**) and (**4**) again showed that they had the same molecular formula $C_{25}H_{26}O_6$ as **1** and **2**. Extensive NMR analysis of **3**, isolated as an oil, showed that this coumarin had the same substituents as **1** since a phenyl, a 2-hydroxy-3-methylbut-3-enyl and a 3-methyl-1-oxobutyl substituent were revealed on its ^1H and ^{13}C NMR spectra (Table 1). However, UV spectrum of **3** (Table 2) exhibited alkaline shifts associated with 8-acyl-5,7-dihydroxycoumarin chromophores.[8] It was then deduced that **3** was a regioisomer of disparinol A (**1**). Inspection of the HMBC contour map of **3** confirmed this hypothesis (Figure 2) and, therefore, this compound was identified as a new 4-arylcoumarin, namely 5,7-dihydroxy-6-(2''-hydroxy-3''-methylbut-3''-enyl)-8-(3'''-methyl-1'''-oxobutyl)-4-phenyl-2H-benzopyran-2-one which we have named isodisparinol A.

On the basis of similar evidences (Table 1 and Figure 2), compound (**4**) appeared to be a regioisomer of **2** and **4** was identified as 5,7-dihydroxy-6-(2''-hydroxy-3''-methylbut-3''-enyl)-8-(2'''-methyl-1'''-oxobutyl)-4-phenyl-2H-benzopyran-2-one which we have named isodisparinol B.

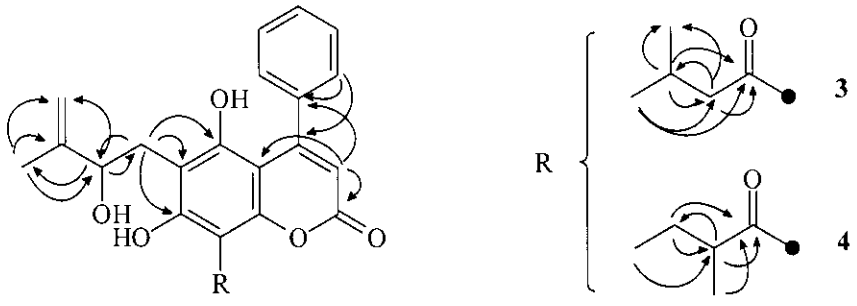
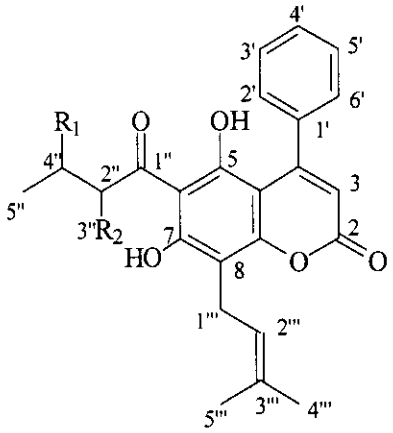


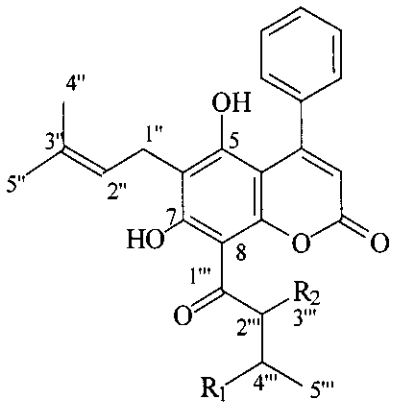
Figure 2 : Main HMBC (J = 6 Hz) correlations of compounds (**3**) and (**4**)

The known coumarins (**5-8**) were identified by comparaison of their spectral data previously described.[7] It may be noticed that the four new coumarins (**1-4**) did not present any significant optical activities ($[\alpha]_D$ and CD). This observation has already been reported for coumarins (**6**) and (**8**) and for a large number of derivatives belonging to this group.[7,9] However, weak optical activities were observed for some 4-alkylcoumarins in highly concentrated solutions[12,13] but not in our case. Then, coumarins (**1-4**) are not necessarily racemic but could exhibit a particular conformation leading to a very weak optical activity.

Unlike dipyranocoumarins such as calanolides and inophyllums generally isolated from Calophyllum genus,[4,14] compounds (**1-8**) are here closely related to saturated acyl substituted coumarins usually reported in Mammea americana,[12,15] M. africana,[16] M. longifolia,[17] and Mesua ferrea.[18]



(**5**) : $R_1 = CH_3$ $R_2 = H$
(**6**) : $R_1 = H$ $R_2 = CH_3$



(**7**) : $R_1 = CH_3$ $R_2 = H$
(**8**) : $R_1 = H$ $R_2 = CH_3$

EXPERIMENTAL

General. HREIMS were recorded on Varian MAT 311 spectrometer at 70 eV. NMR spectra were recorded in $CDCl_3$ solutions on JEOL GSX WB 270 MHz and Bruker Avance DRX 500 MHz instruments using TMS as an internal standard. Optical activities were recorded on a Schmidt-Haensch polarimeter, IR spectra were recorded on a Perkin Elmer 580 spectrophotometer and UV spectra on a Shimadzu UV-1601 spectrophotometer. Melting points were determined with a Electrothermal 8100 melting point apparatus and are uncorrected.

Plant material. The stem bark of Calophyllum dispar was collected by one of us (C. W.) from Terrengganu, Malaysia in October, 1994. An herbarium specimen (KL 4430) is deposited at the University of Kuala-Lumpur, Malaysia and at the FRIM Herbarium-Kepong .

Extraction and isolation. The dried and powdered stem bark (5 kg) was extracted with ethyl acetate (7 L) for 72 h in a Soxhlet apparatus. After concentration, 87.5 g of the residue were repeatedly chromatographied on silica gel by MPLC using hexane gradually enriched with ethyl acetate and methanol, to give coumarins : disparinol A (**1**) (350 mg), disparinol B (**2**) (120 mg), isodisparinol A (**3**) (44 mg), isodisparinol B (**4**) (8 mg), **5** (6.5 g), **6** (400 mg), **7** (110 mg) and **8** (36 mg).

Disparinol A (**1**), 5,7-dihydroxy-8-(2'''-hydroxy-3'''-methylbut-3'''-enyl)-6-(3''-methyl-1''-oxobutyl)-4-phenyl-2H-benzopyran-2-one, $[\alpha]_D$ = 0° (c = 3.02, $CHCl_3$), mp 115-116°C (hexane/ethyl acetate : 90/10). HREIMS : $[M]^+$ 422.1726 (Calcd 422.1729 for $C_{25}H_{26}O_6$), m/z (rel. int.) : 422 (4), 404 (8), 389 (5), 352 (84), 351 (100), 347 (9), 333 (21), 295 (14). IR (film): ν_{max} 1700, 1622, 1580, 1559, 756, 697 cm^{-1}. The ^1H, ^{13}C NMR and UV data are respectively listed in Tables 1 and 2.

Disparinol B (**2**), 5,7-dihydroxy-8-(2'''-hydroxy-3'''-methylbut-3'''-enyl)-6-(2''-methyl-1''-oxobutyl)-4-phenyl-2H-benzopyran-2-one: $[\alpha]_D$ = 0° (c = 2.40, $CHCl_3$), yellow oil, HREIMS : $[M]^+$ 422.1747 (Calcd 422.1729 for $C_{25}H_{26}O_6$), m/z (rel. int.) : 422 (6), 404 (17), 378 (53), 363 (37), 352 (53), 351 (82), 347 (58), 333 (32), 321 (100), 295 (30). IR (film) ν_{max} : 3420, 1718, 1618, 1584, 758, 702 cm^{-1}. The ^1H, ^{13}C NMR and UV data are listed respectively in Tables 1 and 2.

Isodisparinol A (**3**) : 5,7-dihydroxy-6-(2''-hydroxy-3''-methylbut-3''-enyl)-8-(3'''-methyl-1'''-oxobutyl)-4-phenyl-2H-benzopyran-2-one : $[\alpha]_D$ = 0° (c = 0.88, $CHCl_3$), yellow oil, HREIMS : $[M]^+$ 422.1726 (Calcd 422.1729 for $C_{25}H_{26}O_6$), m/z (rel. int.) : 422 (6), 404 (12), 389 (6), 352 (57), 351 (100), 347 (9), 333 (30), 305 (10), 295 (15). IR (film) ν_{max} : 3407, 1717, 1620, 1597, 1559, 758, 702 cm^{-1}. The ^1H, ^{13}C NMR and UV data are listed respectively in Tables 1 and 2.

Isodisparinol B (**4**) : 5,7-dihydroxy-6-(2''-hydroxy-3''-methylbut-3''-enyl)-8-(2'''-methyl-1'''-oxobutyl)-4-phenyl-2H-benzopyran-2-one : $[\alpha]_D$ = 0° (c = 0.16, $CHCl_3$), yellow oil, HRLSIMS : $[M+H]^+$ 423.1808 (Calcd 423.1808 for $C_{25}H_{27}O_6$). IR (film) ν_{max} :3431, 1700, 1605, 771, 694 cm^{-1}. The ^1H, ^{13}C NMR and UV data are listed respectively in Tables 1 and 2.

5,7-Dimethoxy-8-(2''',3'''-epoxy-3'''-methylbutyl)-6-(3''-methyl-1''-oxobutyl)-4-phenyl-2H-benzopyran-2-one (**10**) : m-Chloroperbenzoic acid (170 mg, 0.98 mmol) was added to a solution of 5,7-dimethoxy-6-(3''-methyl-1''-oxobutyl)-8-(3'''-methylbut-2'''-enyl)-4-phenyl-2H-benzopyran-2-one (**9**) (307 mg, 0.70 mmol) in $CHCl_3$ (5 mL). The mixture was stirred at rt for 2 h and then washed twice (5 mL) with a saturated aqueous solution of $NaHCO_3$. The organic layer was dried over anhydrous sodium sulfate and evaporated. The residue was chromatographed on silica gel (cyclohexane-ethyl acetate 8:2) to yield 265 mg (83%) of **10** as an oil wich on standing gave white crystals, mp 75-77°C. UV (MeOH) λ_{max} (logε) : 208 (2.29), 251 (0.78), 300 (0.79) nm. IR (film) ν_{max} : 3059, 2959, 2874, 1736, 1705, 1581, 1458, 1402, 1373, 1198, 1144, 1113 cm^{-1} . ^1H NMR δ : 0.97 (6 H-4'', d, J = 6.7 Hz), 1.36 (3H-4''', s), 1.52 (3H-4''', s), 2.22 (1H-3'', m), 2.67 (2H-2'', d, J = 6.8 Hz), 2.96 (3H-OMe, s), 3.04 (1H-2''', m), 3.14 (1H-1''', d, J = 4.9 Hz), 3.17 (1H-1''', d, J = 4.9 Hz), 3.84 (3H-OMe, s), 6.22 (1H-2, s), 7.41 ppm

(5H-phenyl, m). ^{13}C NMR δ : 19.0 (C-4'''), 22.4 (C-4''), 23.6 (C-1'''), 23.8 (C-3''), 24.8 (C-4'''), 54.0 (C-2''), 59.6 (C-3'''), 62.9 (C-2'''), 63.8 (C-OMe), 64.2 (C-OMe), 110.1 (C-4a), 116.1 (C-2), 117.1 (C-8), 127.6 (C-2',5'), 127.8 (C-3',5'), 128.7 (C-4'), 137.8 (C-1'), 153.7 (C-4), 154.4 (C-5 or 7), 154.5 (C-7 or 5), 158.4 (C-8a), 159.6 (C-2), 203.3 ppm (C-1''). HREIMS : [M^+] 450.2028 (calcd 450.2042 for $C_{27}H_{30}O_6$), m/z (rel. int) : 450 (54), 419 (36), 393 (85), 379 (29), 335 (100), 307 (22). Anal. Calcd for $C_{27}H_{30}O_6$: C, 71.98 ; H, 6.71 ; O, 21.31 ; Found : C, 71.92 ; H, 6.74 ; O, 21.26.

5 ,7-Dimethoxy-6-(3''-methyl-1''-oxobutyl)-8-(3'''-hydroxy-2'''-methylbut-3'''-enyl)-4-phenyl-2H-benzopyran-2-one (**11**) : A mixture of **10** (40 mg, 0.088 mmol) and aluminum isopropoxide (18 mg, 0.088 mmol) was refluxed in xylene (7.5 mL) for 10 h. After cooling, 10 mL of 2 N HCl were added. The organic layer was separated, washed with brine (10 mL), dried over anhydrous sodium sulfate and evaporated. The residue was chromatographed on silica gel TLC (cyclohexane-ethyl acetate 3 : 2) to yield 21 mg (53 %) of **11** : mp 115 °C (from cyclohexane-ethyl acetate), UV (MeOH) λ_{max} (logε) : 206 (1.65), 254 (0.47), 302 (0.46) nm. IR (film) ν_{max} : 3439, 3055, 2984, 1728, 1709, 1645, 1581, 1454, 1425, 1111 cm^{-1}. ^{1}H NMR δ : 0.97 (6 H-5'', d, J = 7.0 Hz), 1.93 (3H-5''', s), 2.22 (1H-4'', m), 2.67 (2H-2'', d, J = 7.0 Hz), 2.95 (3H-OMe, s), 3.04 (1H-1''', dd, J = 9.1 and J = 13.6 Hz), 3.17 (1H-1''', dd, J = 4.1 and J = 13.9 Hz), 3.84 (3H-OMe, s), 4.43 (1H-2''', dd, J = 4.0 and J = 9.1 Hz), 4.89 (1H-4''', s), 5.04 (1H-4''', s), 6.21 (1H-2, s), 7.41 ppm (5H-phenyl, m). ^{13}C NMR δ : 18.1 (C-4'''), 22.4 (C-5''), 23.8 (C-4''), 30.2 (C-1'''), 54.0 (C-2''), 63.6 (C-OMe), 63.8 (C-OMe), 75.0 (C-2'''), 110.0 (C-4a), 110.7 (C-4'''), 116.1 (C-2), 117.7 (C-8), 127.5 (C-2', 6'), 127.8 (C-3', 5'), 128.5 (C-4'), 137.8 (C-1'), 147.3 (C-3'''), 153.5 (C-4), 154.3 (C-5 or 7), 154.6 (C-5 or 7), 158.4 (C-8a), 159.6 (C-2), 203.5 ppm (C-1''). HREIMS : [M^+] 450.2024 (calcd 450.2042 for $C_{27}H_{30}O_6$), m/z (rel. int.) : 450 (5), 380 (98), 379 (100), 365 (42), 323 (94), 307 (10), 127 (32), 57(54). Anal. Calcd for $C_{27}H_{30}O_6$: C, 71.98 ; H, 6.71 ; O, 21.31 ; Found : C, 71.95 ; H, 6.69 ; O, 21.27.

Obtention of compound (**1**) : A solution of **11** (85 mg, 0.19 mmol) in dichloromethane (5 mL) at 0°C was added to a solution of boron tribromide dimethyl sulfide complex (0.4 mL of 1M solution in dichloromethane). The mixture was stirred for 3 h at 0°C and then allowed to warm up to rt overnight. The reaction mixture was washed twice with water (10 mL). The organic layer was separated, dried over anhydrous sodium sulfate, and evaporated. The residue was chromatographed on silica gel TLC (cyclohexane-ethyl acetate 3 : 2) to give **1** (4.5 mg, 6%) The spectral data (^{1}H and ^{13}C NMR) were identical to those of the natural product.

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