Pergamon

PII: S0031-9422(97)01110-2

Phytochemistry, Vol. 48, No. 2, pp. 327–331, 1998

Printed in Great Britain
0031-9422/98 $19.00+0.00

FOUR STILBENOIDS FROM *PLEIONE BULBOCODIOIDES*

LI BAI, NORIKO MASUKAWA, MASAE YAMAKI* and SHUZO TAKAGI

Faculty of Pharmaceutical Sciences, Mukogawa Women's University, 11-68 Koshien Kyuban-cho, Nishinomiya Hyogo 663, Japan

(*Received* 10 *November* 1997)

Key Word Index—*Pleione bulbocodioides*; Orchidaceae; tubers; shanciol E, F; dihydrophenanthropyrans; bulbocodin C, D; bibenzyl.

Abstract—Two dihydrophenanthropyrans, shanciol E and F, and two bibenzyls, bulbocodin C and D, were isolated from tubers of *Pleione bulbocodioides*. The new structures were elucidated to be 3-hydroxy-11-methoxy-2-(4′-hydroxy-3′,5′-dimethoxyphenyl)-2,3,5,6-tetrahydro-4H-phenanthro[2,1-b] pyran-8-ol; 4-hydroxy-11-methoxy-3-(4′-hydroxy-3′-methoxyphenyl)-3,4,5,6-tetrahydro-2H-pehnanthro[2,1-b] pyran-8-ol; 3′,5-dihydroxy-3-methoxy-2,4-di(*p*-hydroxybenzyl)bibenzyl; 3,3′-dihydroxy-5-methoxy-2,4-di(*p*-hydroxybenzyl)bibenzyl; respectively, on the basis of spectroscopic data and chemical correlations.

INTRODUCTION

In our previous papers, the isolation and structural determination of some stilbenoids and lignans in *Pleione bulbocodioides* were described [1–3]. Further investigation of the same source has resulted in the isolation of two new dihydrophenanthropyrans, shanciol E (**1**) and F (**2**), and two bibenzyls, bulbocodin C (**3**) and D (**4**). The structures were determined on the basis of spectral data and chemical correlations.

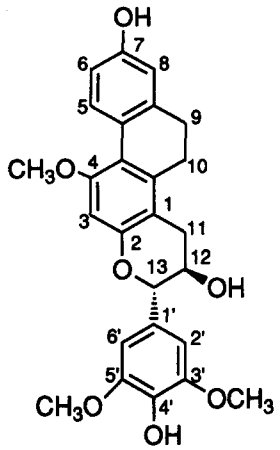
RESULTS AND DISCUSSION

Shanciol E (**1**) showed UV absorption maxima at 212, 281 and 300 nm indicative of a dihydrophenanthrene [4]. The IR spectrum exhibited absorptions at 3300 (OH), 1595 and 1450 cm^{-1} (benzenoid). The mass spectrum exhibited a $[M]^+$ at *m/z* 450 ($C_{26}H_{26}O_7$), a base peak at *m/z* 255 and a prominent peak at *m/z* 196, which were in complete agreement with a Retro-Diels-Alder fission as shown in Scheme 1. The ^{13}C NMR spectrum displayed signals for all 26 carbons in the molecule: one ethylene, one methylene, three methoxyls and two methines bearing oxygen, along with 18 aromatic carbons, of which six were protonated, six quaternary and six bearing oxygen. Acetylation of **1** afforded a triacetate ($[M]^+$ *m/z* 576), whose ^1H NMR spectrum contained three signals at δ 1.97, 2.25 and 2.27, suggesting the presence of one secondary and two phenolic hydroxyl groups. The ^1H-^1H COSY and ^1H NMR spectra (Table 1) showed that **1** had a dihydrophenanthropyran moiety: one multiplet at δ 2.60–2.67 (4H) due to H-9 and H-10, an ABX system at δ 8.00, 6.62 and 6.65 due to H-5, H-6 and H-8, and a singlet at δ 6.51 due to H-3, along with one methylene at δ 2.68 and 2.98 due to H-11, and two methines at δ 4.10 and 4.65 due to H-12 and H-13, respectively. The chemical shifts and the signal patterns closely resembled to those of a known dihydrophenanthropyran shanciol (**5**) [1], except for the difference in the splitting pattern of a phenyl group, whose signals presented as one singlet at δ 6.73 (2H), together with the signals due to three methoxyl groups at δ 3.85 (6H) and 3.81, suggesting that the phenyl group in **1** was substituted symmetrically with one hydroxyl and two methoxyl groups. This substitution pattern was consistent with the observations that the signals of H-2′ and H-6′ were enhanced on irradiation of the two methoxyl groups at C-3′ and C-5′ in NOE experiments.

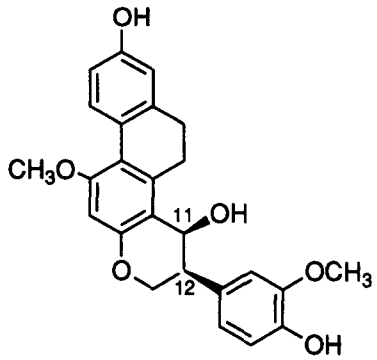
The relative stereochemistry of the C-12 and C-13 substituents was deduced to be *trans* from the coupling constant (J = 8.1 Hz) between H-12 and H-13 [5]. On the basis of these findings, shanciol E was assigned structure **1**.

Shanciol F (**2**) showed UV maxima at 211, 282 and 310 (sh) nm and the IR spectrum showed the presence of hydroxyl groups and benzenoids. The mass spectrum of **2** exhibited a $[M]^+$ at *m/z* 420 ($C_{25}H_{24}O_6$), 30 amu less than that of **1**, and a significant peak at *m/z* 402 formed by the loss of one molecule of H_2O, as shown in Scheme 1. Shanciol F also gave a triacetate on acetylation. The ^1H NMR spectrum of **2** showed

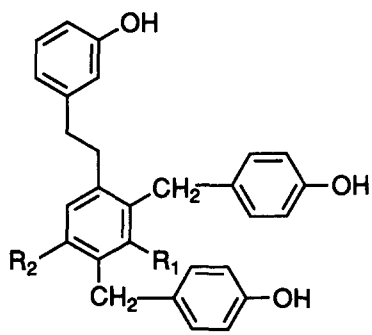
* Author to whom correspondence should be addressed.



1

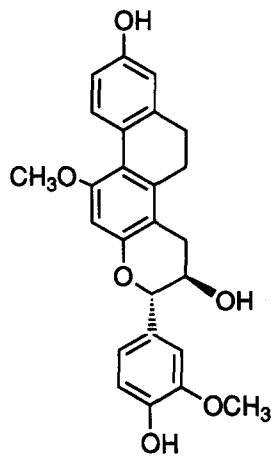


2



3 R_1=OCH_3 R_2=OH

4 R_1=OH R_2=OCH_3



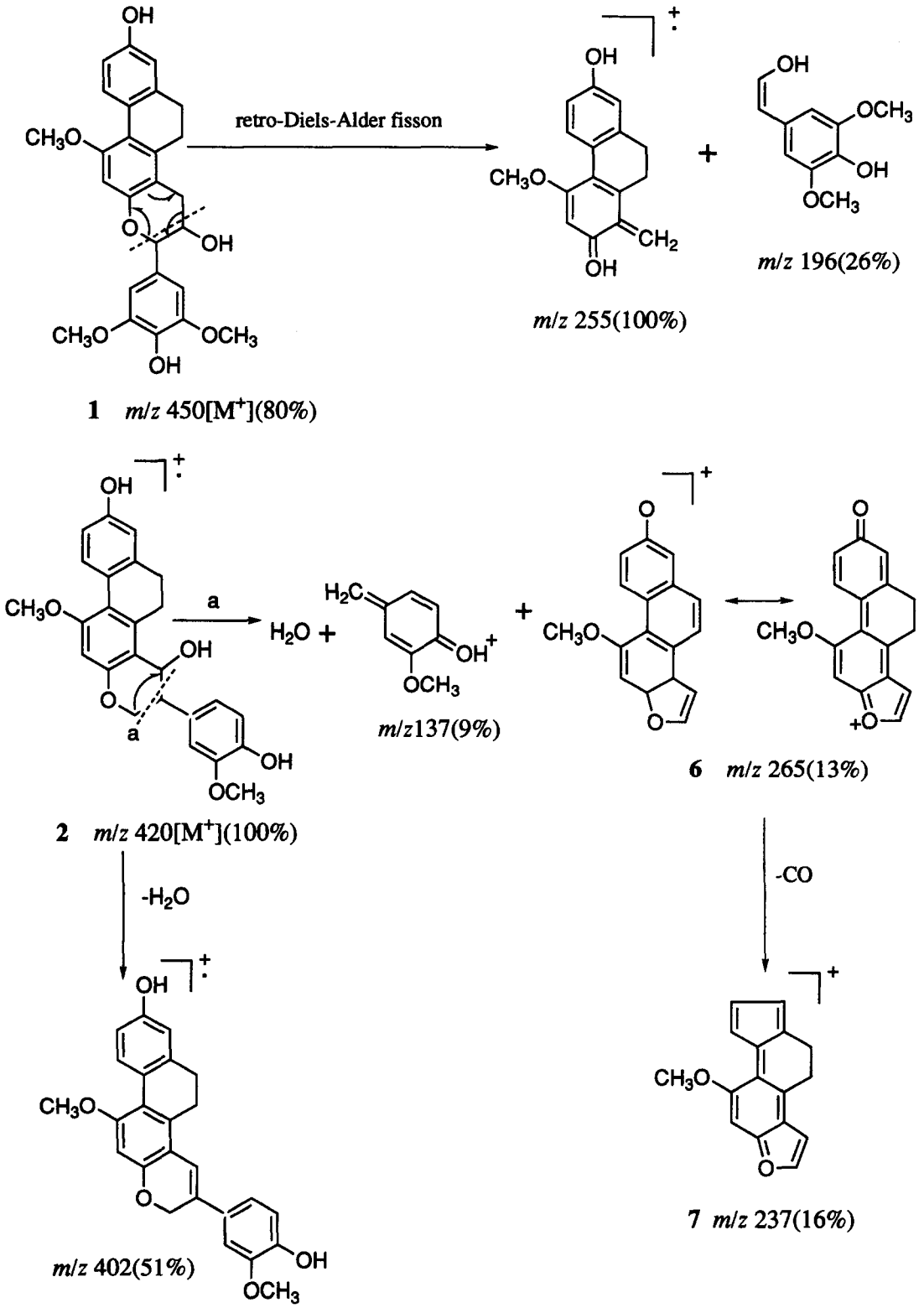
5

the signals of a dihydrophenanthrene moiety with the same substitution pattern as **1**, while the signals of a pyran ring appeared at δ 5.64 and 3.46 (*m*) due to two methines (H-11 and H-12), and at δ 3.60 and 3.85 (*m*), due to one methylene (H-13), suggesting that the hydroxyl and phenyl groups were placed at C-11 and C-12, respectively, as shown in **2**. This assumption was further supported by the mass spectrum, which contained prominent peaks at *m/z* 265 and 237 ascribed to ions **6** and **7**, obviously different from **1** (Scheme 1).

Additionally, in the ^{1}H NMR spectrum, the signals of the phenyl group appeared as one set of an ABX system at δ 6.90, 6.74 and 6.79 due to H-2′, H-5′ and H-6′. Only two methoxyls at δ 3.80 and 3.81 were observed, suggesting the phenyl group was disubstituted. Selective irradiation of the methoxyl group at δ 3.81 gave NOE enhancements of H-3 and H-5, while irradiation of the other at δ 3.80 only caused enhancement of the signal of H-2′. Thus, two methoxyls were at C-4 on the dihydrophenanthrene group and C-3′ on the phenyl group, respectively. Shanciol F had *cis* relative stereochemistry of the C-11 and C-12 substituents (J = 3.0 Hz). Thus, shanciol F was established to have structure **2**.

For comparison of the spectral data with other phenanthrenes, we have used the phenanthrene numbering system instead of the systematic nomenclature. Thus shanciol E and shanciol F should be called 3-hydroxy-11-methoxy-2- (4′-hydroxy-3′,5′-dimethoxyphenyl)-2,3,5,6-tetrahydro-4H-phenanthro[2,1-b] pyran-8-ol and 4-hydroxy-11-methoxy-3-(4′-hydroxy-3′-methoxyphenyl)-3,4,5,6-tetrahydro-2H-phenanthro[2,1-b] pyran-8-ol, respectively.

Bulbocodin C (**3**) showed UV maxima at 210 and 280 nm, suggesting **3** to be a bibenzyl [6]. The IR spectrum exhibited absorptions at 3250 (OH), 1590 and 1500 cm^{-1} (benzenoid). The mass spectrum exhi-



Scheme 1. Mass spectral fragmentation of compounds **1** and **2**.

bited a $[M]^+$ at *m/z* 456 ($C_{29}H_{28}O_5$) and significant peaks at *m/z* 350 and 243 formed by sequential cleavages of two hydroxybenzyl groups. Acetylation of **3** gave a tetraacetate ($[M]^+$ *m/z* 624) indicating the presence of four hydroxyl groups. The ^1H NMR spectrum (Table 2) showed three doublets at δ 6.64 (4H), 6.85,

Table 1. ^{1}H NMR data of shanciol E (**1**) and F (**2**) and their acetates*

H	**1**	**1** acetate	**2**	**2** acetate
3	6.51 *s*	6.63 *s*	6.54 *s*	6.55 *s*
5	8.00 *d* (8.5)	8.19 *d* (8.6)	7.99 *d* (9.4)	8.19 *d* (8.6)
6	6.62 *dd* (8.5, 2.6)	6.91 *dd* (8.6, 2.6)	6.62 *m*	6.95 *dd* (8.6, 2.6)
8	6.65 *d* (2.6)	6.95 *d* (2.6)	6.61 *d* (2.6)	6.94 *d* (2.6)
9,10	2.60–2.67 *m*	2.61–2.64 *m* 2.69–2.73 *m*	2.56–2.70 *m*	2.71 *m*
11	2.68 *dd* (15.5, 8.6) 2.98 *dd* (15.5, 5.7)	2.83 *dd* (16.2, 6.8) 2.96 *dd* (16.2, 5.3)	5.64 *d* (3.0)	5.60 *d* (3.4)
12	4.10 *ddd* (8.6, 8.1, 5.7)	5.41 *ddd* (6.8, 6.4, 5.3)	3.46 *m*	3.71 *m*
13	4.65 *d* (8.1)	5.14 *d* (6.4)	3.60 *dd* (11.1, 9.0) 3.85 *m*	4.12 *dd* (11.3, 9.4) 4.46 *dd* (11.3, 4.2)
2′	6.73 *s*	6.77 *s*	6.90 *d* (1.9)	6.92 *d* (1.8)
5′	—	—	6.74 *d* (8.3)	7.00 *d* (8.1)
6′	6.73 *s*	6.77 *s*	6.79 *dd* (8.3, 1.9)	6.90 *dd* (8.1, 1.8)
4-OMe	3.81 *s*	3.87 *s*	3.81 *s*	3.88 *s*
3′-OMe	3.85 *s*	3.77 *s*	3.80 *s*	3.80 *s*
5′-OMe	3.85 *s*	3.77 *s*	—	
OCOMe	—	1.97, *s*, 2.25 *s*, 2.27 *s*	—	2.09 *s*, 2.29 *s*, 2.30 *s*

* Coupling constants (J in Hz) are given in parentheses.

Table 2. ^{1}H NMR data of bulbocodin C (**3**) and D (**4**) and their acetates*

H	**3**	**3** acetate	**4**	**4** acetate
6	6.56 *s*	6.75 *s*	6.33 *s*	6.69 *s*
2′	6.50 *m*	6.67 *t* (2.1, 1.7)	6.53 *m*	6.77 *t* (1.7)
4′	6.57 *dd* (8.4, 2.2)	6.84 *d* (8.1)	6.57 *dd* (8.2, 2.3)	6.88 *br d* (7.7)
5′	7.01 *t* (8.4)	7.22 *t* (8.1)	7.02 *t* (8.2)	7.22 *t* (7.7)
6′	6.50 *m*	6.87 *d* (8.1)	6.53 *m*	6.88 *br d* (7.7)
2″,6″	6.85 *d* (8.5)	6.94 *d* (8.6)	6.90 *d* (8.5)	7.04 *d* (8.6)
3″5″	6.64 *d* (8.5)	7.08 *d* (8.6)	6.62 *d* (8.5)	6.92 *d* (8.6)
2‴,6‴	7.03 *d* (8.5)	6.93 *d* (8.6)	7.01 *d* (8.5)	7.12 *d* (8.1)
3‴,5‴	6.64 *d* (8.5)	7.06 *d* (8.6)	6.65 *d* (8.5)	6.94 *d* (8.1)
-CH_2-CH_2-	2.59 *m*	2.42 *m*	2.59 *m*	2.71 *m*
	2.66 *m*	2.82 *m*	2.77 *m*	2.86 *m*
2-CH_2-	3.86 *s*	3.91 *s*	3.94 *s*	3.80 *br s*
4-CH_2-	3.91 *s*	4.09 *s*	3.90 *s*	3.80 *br s*
3-OMe	3.44 *s*	3.80 *s*	—	—
5-OMe	—	—	3.68 *s*	3.73 *s*
OCOMe	—	2.16 *s*, 2.23 *s*, 2.26 *s*	—	2.08 *s*, 2.23 *s*, 2.24 *s*

* Coupling constant (J in Hz) are given in parentheses.

and 7.03 due to two pairs of A_2B_2 systems characteristic of a *p*-substituted aromatic ring, and two singlets at δ 3.86 and 3.91 due to two benzylic methylenes, supporting the presence of two *p*-hydroxybenzyl groups. In addition, the ^{1}H NMR spectrum contained the signals of one methoxyl at δ 3.44 and two methylenes at δ 2.59 (2H) and 2.66 (2H), along with five aromatic protons for the bibenzyl groups. Of these, four appeared at δ 6.50 (2H), 6.57 and 7.01 assignable to H-2′, H-6′, H-4′ and H-5′ on one aromatic ring based on their chemical shifts and coupling patterns [2, 6], the remaining one appeared as a singlet at δ 6.56 due to a proton on the other ring. In a NOE experiment, irradiation of the methylene at δ 2.66 caused NOEs with the other methylene (10%), H-6 (9%) and one of the benzylic methylenes at δ 3.86 (4%). In turn, irradiation of the methoxyl caused NOEs with two benzylic methylenes at δ 3.86 (2%) and 3.91 (3%), indicating the methoxyl and two *p*-hydroxybenzyls at C-3, C-2 and C-4, respectively, on the same aromatic ring and one hydroxyl at C-5. The remaining hydroxyl group as placed at C-3′, which was confirmed by a comparison with the splitting pattern of the known 3′-hydroxybibenzyls [2, 3, 6], and by the downfield shifts of H-2′ and H-4′ in its acetate (Table 2). On the basis of the above findings, the

structure of **3** was established as 2,4-bis(*p*-hydroxybenzyl)-3′,5-dihydroxy-3-methoxybibenzyl. Bulbocodin (**4**) showed the same [M$^+$] at *m/z* 456 ($C_{29}H_{28}O_5$), and two intense peaks as in **3**. The UV, IR and the ^{1}H NMR (Table 2) data were almost identical to those of **3**, except for the signals for H-6 at high field (Δ0.23) and methoxyl at lowfield (Δ0.24), respectively. These being attributable to the shielding of adjacent groups. In a NOE experiment, irradiation of the methoxyl group at δ 3.68 enhanced the signals due to H-6 (12%) and only one benzylic methylene at δ 3.90 (1%). This finding indicated that the hydroxyl and methoxyl group were interchanged with each other at C-3 and C-5. The ^{13}C NMR spectrum of **4** and its acetate supported these deductions. Thus, the structure of **4** was assigned to be 2,4-bis(*p*-hydroxybenzyl)-3,3′-dihydroxy-5-methoxybibenzyl.

EXPERIMENTAL

Mps.; uncorr.; IR: KBr; UV: MeOH; ^{1}H NMR and ^{13}C NMR: 500 and 125 MHz, respectively, MeOH-d_3 with TMS. The peaks marked with an asterisk are overlapped and not resolved. MS; EIMS, 70 eV. CC and TLC: Merck silica gel.

Plant materials

See Ref. [1].

Extraction and isolation

See Ref. [1]; Fr. 5 was rechromatographed over silica gel, LH-20 and Cosmosil C_{18} to give **1** (5 mg) and **2** (7 mg), and a mixt. of **3** and **4** which was separated on Cellulofine to give **3** (3 mg) and **4** (8 mg).

Compound **1**. Colourless plates from MeOH, mp 244–246°, $[\alpha]_D$ −11.8 (MeOH). IR ν_{max} cm^{-1}: 3300, 1595, 1450; UV λ_{max} nm (log ε): 212 (4.72), 281 (4.26), 300 (4.11); MS *m/z* (rel. int.): 450 (80), 432 (2), 255 (100), 196 (26); ^{1}H NMR: Table 1; ^{13}C NMR: δ 26.5 (*t*, C-9), 30.7 (*t*, C-10), 32.6 (*t*, C-11), 56.1 (*q*, 4-OMe), 56.9 (*q*, 3′,6′-OMe), 69.4 (*d*, C-12), 83.2 (*d*, C-13), 99.8 (*d*, C-3), 106.0 (*d*, C-2′,6′), 111.4 (*s*, C-1′), 113.7 (*d*, C-6), 114.8 (*d*, C-8), 118.9 (*s*, C-4a), 126.1 (*s*, C-5a), 130.4 (*d*, C-5), 131.1 (*s*, C-1), 136.8 (*s*, C-4′), 139.9 (*s*, C-10a), 140.4 (*s*, C-8a), 149.3 (*s*, C-3′,5′), 154.7 (*s*, C-2), 156.4 (*s*, C-7), 157.7 (*s*, C-4). Triacetate: Colourless needles from MeOH, mp 164–165°. MS *m/z* (rel. int.): 576 [M]$^+$ (100), 534 (86), 492 (34), 450 (16), 432 (30), 255 (62); ^{1}H NMR: Table 1.

Compound **2**. White powder, $[\alpha]_D$ −8.3 (MeOH). IR ν_{max} cm^{-1}: 3250, 1600, 1500, 1420; UV λ_{max} nm (log ε): 211 (4.69), 282 (4.34), 310 sh (4.06); MS *m/z* (rel. int.): 420 (100), 402 (51), 265 (13), 237 (16), 137 (9); ^{1}H NMR: Table 1; ^{13}C NMR: δ 28.0 (*t*, C-9), 30.9 (*t*, C-10), 54.7 (*d*, C-12), 56.3 (*q*, 4-OMe), 56.5 (*q*, 3′-OMe), 65.0 (*t*, C-13), 88.8 (*d*, C-11), 94.0 (*d*, C-3), 110.3 (*d*, C-2′), 113.8 (*d*, C-6), 115.0 (*d*, C-8), 116.3 (*d*, C-5′), 116.9 (*s*, C-4a), 118.1 (*s*, C-1), 119.2 (*d*, C-6′), 126.3 (*s*, C-1′), 130.2 (*d*, C-5), 135.8 (*s*, C-10a), 137.6 (*s*, C-5a), 140.3 (*s*, C-8a), 147.3 (*s*, C-4′), 149.1 (*s*, C-3′), 156.2 (*s*, C-7), 159.5 (*s*, C-2), 160.6 (*s*, C-4). Triacetate: Oil. MS *m/z* (rel. int.): 546 [M]$^+$ (100), 504 (33), 444 (32), 402 (31); ^{1}H NMR: Table 1.

Compound **3**. White powder. IR ν_{max} cm^{-1}: 3250, 1590, 1500; UV λ_{max} nm (log ε): 210 (4.58), 280 (3.72); MS *m/z* (rel. int.): 456 (2), 350 (100), 243 (73), 107 (33); ^{1}H NMR: Table 2; ^{13}C NMR: δ 29.8 (*t*, ϕ-C̲H$_2$-CH$_2$-ϕ), 31.6 (*t*, ϕ-CH$_2$-C̲H$_2$-ϕ), 36.2 (*t*, ϕ-CH$_2$-ϕ), 38.6 (*t*, ϕ-CH$_2$-ϕ), 62.2 (*q*, 3-OCH$_3$), 113.6 (*d*, C-6), 113.8 (*d*, C-4′), 115.8 (*d*, C-3″,5″), 116.1 (*d*, C-3‴,5‴), 116.4 (*d*, C-2′), 120.5 (*s*, C-4), 120.8 (*d*, C-6′), 124.4 (*d*, C-2), 130.1 (*d*, C-2″,6″), 130.2 (*d*, C-5′), 130.4 (*d*, C-2‴,6‴), 134.2 (*s*, C-1″,1‴), 141.9 (*s*, C-1′), 144.9 (*s*, C-1), 156.0 (*S*, C-4″), 156.1 (*s*, C-4‴), 156.2 (*s*, C-3′), 158.3 (*s*, C-5), 159.5 (*s*, C-3). Tetraacetate: colourless needles, mp 150–152° (MeOH). MS *m/z* (rel. int.): 624 [M]$^+$ (60), 582 (100), 540 (45), 498 (12), 456 (1), 255 (24), 107 (39); ^{1}H NMR: Table 2.

Compound **4**. Colourless needles, mp 169–171° (MeOH: H$_2$O). IR ν_{max} cm^{-1}: 3250, 1590, 1500; UV λ_{max} nm (log ε): 211 (4.60), 280 (3.78); MS *m/z* (rel. int.): 456 (100), 349 (23), 243 (73), 107 (75); ^{1}H NMR: Table 2; ^{13}C NMR: δ 29.0 (*t*, ϕ-C̲H$_2$-CH$_2$-ϕ), 31.4 (*t*, ϕ-CH$_2$-C̲H$_2$-ϕ), 36.8 (*t*, ϕ-CH$_2$-ϕ), 38.6 (*t*, ϕ-CH$_2$-ϕ), 56.1 (*q*, 3-OCH$_3$), 106.0 (*d*, C-6), 113.8 (*d*, C-4′), 115.8 (*d*, C-3″,5″), 116.1 (*d*, C-3‴,5‴), 116.5 (*d*, C-2′), 120.7 (*s*, C-4), 120.9 (*d*, C-6′), 130.6 (*d*, C-2), 130.1 (*d*, C-2″,6″), 130.2 (*d*, C-5′), 130.3 (*d*, C-2‴,6‴), 133.8 (*s*, C-1″), 133.9 (*s*, C-1‴), 140.9 (*s*, C-1′), 145.0 (*s*, C-1), 156.0 (*s*, C-4″), 156.2 (*s*, C-4‴), 154.8 (*s*, C-3′), 158.3 (*s*, C-5), 157.9 (*s*, C-3). Tetraacetate: Oil. MS *m/z* (rel. int.): 624 [M]$^+$ (73), 582 (100), 540 (61), 498 (20), 468 (7); ^{1}H NMR: Table 2.

REFERENCES

1. Bai, L., Yamaki, M., Yamagata, Y. and Takagi, S., *Phytochemistry*, 1996, **41**, 625.
2. Bai, L., Yamaki, M. and Takagi, S., *Phytochemistry*, 1996, **42**, 853.
3. Bai, L., Masukawa, N., Yamaki, M. and Takagi, S., *Phytochemistry*, 1997, **44**, 1565.
4. Letcher, R. M. and Nhamo, L. R. M., *Journal of the Chemical Society, Perkin Transactions I*, 1972, 2941.
5. Yamaguchi, S., Ito, S., Nakamura, A. and Inoue, N., *Bulletin of the Chemical Society of Japan*, 1965, **38**, 2187.
6. Bai, L., Kato, T., Inoue, K., Yamaki, M. and Takagi, S., *Phytochemistry*, 1993, **33**, 1481.