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TWO NOVEL DITERPENES FROM BARK OF *CINNAMOMUM CASSIA**

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Key Word Index—*Cinnamomum cassia*; Lauraceae; bark; diterpene; cinncassiol D_4; cinncassiol D_4 glucoside.

Abstract—Two new diterpenes were isolated from the fraction exhibiting anti-allergic activity obtained from the bark of *Cinnamomum cassia*. They have been given the trivial names cinncassiol D_4 and cinncassiol D_4 glucoside and their structures determined on the basis of chemical and spectral evidence.

INTRODUCTION

We have previously reported [1–6] the isolation of a series of diterpenes, Cassia Diterpene, obtained from the fraction exhibiting anti-allergic activity isolated from Cinnamomi cortex (Tōkō Keihi, the dried bark of *Cinnamomum cassia* Blume; one of most widely used crude drugs).

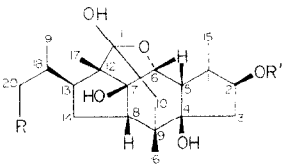
Further studies on this fraction led to the isolation of a new diterpene (**1**) and its glucoside (**2**) which belong to the cinncassiol D series [4, 6] and were named cinncassiol D_4 and cinncassiol D_4 glucoside, respectively.

RESULTS AND DISCUSSION

Cinncassiol D_4 (**1**), an amorphous powder, $[\alpha]_D$ $-16.3°$, possesses a molecular formula $C_{20}H_{32}O_5$, which is one less oxygen atom than that of cinncassiol D_3 (**3**), a novel diterpene with a new skeleton, previously reported by Nohara *et al.* [6]. A comparison of the ^{1}H NMR spectra (Table 1) of cinncassiol D_4 monoacetate (**4**), $[\alpha]_D-26.9°$ and cinncassiol D_3 diacetate (**5**) revealed that the structure of **1** corresponds to the 19-desoxy compound of **3**.

The presence of a hydroxyl group at C-2 was verified by irradiation in turn of H-6, H-5, H-1, H-2 and H-3 in the ^{1}H NMR spectrum of **4** as well as **5**. Furthermore, the configuration of the C-2 hydroxyl was regarded as β because of acetonide formation between the C-2 and C-4 hydroxyl groups; the configuration of the C-4 hydroxyl was also assigned as β in **4** and **5** since the methyl group at C-9 appeared almost in the same range comparing the ^{1}H NMR spectra of both **4** and **5**. Examination of the Dreiding model of **1** showed a restricted conformation and indicated that the distances between H-6 and the C-15 methyl group were *ca* 4.0 and 2.2 Å in the case of the β- and α-configurations, respectively. Since an NOE was observed for H-6 (integration *ca* 8%) when the C-15 methyl group was saturated, the configuration of the C-15 methyl group was represented as α as shown in **1**.

Cinncassiol D_4 glucoside (**2**), an amorphous powder, $[\alpha]_D-12.5°$, showed strong IR absorption at 3400 cm^{-1} due to a hydroxyl group, suggesting **2** to be a glycoside. Enzymatic hydrolysis with crude hesperidinase afforded a diterpene identical with cinncassiol D_4 and D-glucose. Since the field desorption mass spectrum of **2** showed a molecular ion at *m/z* 514, **2** consisted of 1 mol each of **1** and D-glucose. The acetate (**6**) exhibited a terminal peracetylated hexosyl cation at *m/z* 331 in the mass spectrum and four acetyl signals at δ 1.98–2.05 in the ^{1}H NMR spectrum. The above evidence suggested that the glucosyl residue should be bound with the secondary hydroxyl at C-2. As regards the location of this glucosyl bond the glycosidation shift [7, 8] observed in the ^{13}C NMR spectra of **1** and **2** (Table 2) supported



1 R=R'=H
2 R=H, R'=-β-D-glc·pyr
3 R=OH, R'=H
4 R=H, R'=Ac
5 R=OAc, R'=Ac
6 R=H, R'=2',3',4',6'-tetra-*O*-acetyl-β-D-glc·pyr

*Part VII in the series "Studies on the Constituents of Cinnamomi Cortex". For Part VI see Nakano, K., Nohara, T., Tomimatsu, T. and Nishioka, I. (1981) *Yakugaku Zasshi* (in press).

Table 1. ^{1}H NMR spectral data of compounds **4** and **5** in C_5D_5N

	4	**5**
1-Me	1.21 (*d*, *J* = 7 Hz)	1.25 (*d*, *J* = 7 Hz)
2-H	5.62 (*ddd*, *J* = 8, 8 and 8 Hz)	5.60 (*ddd*, *J* = 8, 8 and 8 Hz)
3-H	3.19 (*dd*, *J* = 8 and 14 Hz)	3.20 (*dd*, *J* = 8 and 14 Hz)
6-H	4.38 (*d*, *J* = 2 Hz)	4.39 (*br s*)
9-Me	1.25 (*s*)	1.28 (*s*)
12-Me	1.71 (*s*)	1.70 (*s*)
18-Me	1.03 (*d*, *J* = 6 Hz)	1.31 (*d*, *J* = 6 Hz)
	1.18 (*d*, *J* = 6 Hz)	—
18-CH_2	—	4.04 (*dd*, *J* = 7 and 11 Hz)
	—	4.25 (*dd*, *J* = 7 and 11 Hz)
OAc	2.00 (*s*)	2.00 (*s*)
	—	2.03 (*s*)

its attachment at C-2. Furthermore, the configuration of the glucosyl bond was deduced to be β on the basis of the coupling constant ($J_{H\text{-}1,H\text{-}2}$ = 7 Hz) observed in the ^{1}H NMR spectrum of **2**. Consequently, **2** was represented as cinncassiol D_4 2-O-β-D-glucopyranoside.

EXPERIMENTAL

Mps were uncorr. The ^{1}H NMR spectra were recorded at 100 MHz; the ^{13}C NMR spectra were determined at 22.5 MHz; the MS spectra were recorded under the following conditions: 70 eV, 4.5 kV, 300 μA, 220°; FD 60–70°, 3×10^{-7} Torr.

Table 2. ^{13}C NMR spectral data of compounds **1** and **2** in C_5D_5N

Carbon	**1**	**2**
1	44.0	42.0
2	78.4	88.4
3	46.9	45.7
4	89.0	88.9
5	53.7	53.1
6	76.3	76.3
7	83.7	83.8
8	48.6	48.6
9	42.3	42.3
10	27.3	27.3
11	107.6	107.6
12	57.8	57.8
13	46.9	47.0
14	44.2	44.3
15	13.1	13.1
16	22.9	22.8
17	10.3	10.2
18	29.3	29.3
19	24.2	24.2
20	19.2	19.2
1′	—	105.5
2′	—	75.3
3′	—	77.6
4′	—	71.4
5′	—	78.3
6′	—	62.9

Isolation. Further separation of fractions 8 + 9 and fraction 17 [3] obtained from the H_2O extractive of the dried bark (10 kg) of *C. cassia* gave cinncassiol D_4 (40 mg) and cinncassiol D_4 glucoside (50 mg), respectively.

Cinncassiol D_4 (**1**). An amorphous powder; $[\alpha]_D^{26}$ − 16.3° (MeOH; *c* 0.49); EIMS *m/z*: 352 $[M]^+$, 334, 316, 301, 291, 275, 257, 214, 151; ^{1}H NMR (C_5D_5N): δ 1.03 (3 H, *d*, *J* = 7 Hz, 18(19)-Me), 1.19 (3H, *d*, *J* = 6 Hz, 18(20)-Me), 1.30 (3H, *s*, 9-Me), 1.41 (3H, *d*, *J* = 6 Hz, 1-Me), 1.71 (3H, *s*, 12-Me), 4.43 (1H, *d*, *J* = 2 Hz, 6-H); (CD_3OD): δ 0.94 (6 H, *d*, *J* = 7 Hz, 18-Me_2), 0.95 (3H, *s*, 9-Me), 1.08 (3H, *d*, *J* = 6 Hz, 1-Me), 1.09 (3 H, *s*, 12-Me), 2.52 (1 H, *dd*, *J* = 8, 14 Hz, 3-H), 3.77 (1H, *d*, *J* = 2 Hz, 6-H), 4.10 (1H, *ddd*, *J* = 8, 8 and 8 Hz, 2-H). (Found: C, 68.33; H, 9.09. $C_{20}H_{32}O_5$ requires: C, 68.15; H, 9.15%.)

Cinncassiol D_4 2-O-monoacetate (**4**). A mixture of **1** (18 mg), Ac_2O (2 ml) and pyridine (1 ml) was left to stand for 30 min at room temp. to give a monoacetate **4** (16 mg). An amorphous powder; $[\alpha]_D^{26}$ − 26.9° (MeOH; *c* 1.15); EIMS *m/z*: 394 $[M]^+$, 376, 358, 316, 299, 283, 273, 257, 249, 213; ^{1}H NMR ($CDCl_3$): δ 0.93 (6H, *d*, *J* = 7 Hz, 18-Me_2), 0.98 (3H, *s*, 9-Me), 1.08 (3H, *d*, *J* = 7 Hz, 1-Me), 1.16 (3H, *s*, 12-Me), 2.04 (3H, *s*, –OAc), 3.77 (1H, *d*, *J* = 2 Hz, 6-H); ^{13}C NMR (C_5D_5N): δ 10.2, 12.5, 19.1, 22.7, 24.2, 29.1, 29.2, 42.5(×2), 44.4, 45.4, 47.0, 48.5, 53.0, 57.8, 75.0 (C–O–), 81.3 (C–O–), 83.8 (C–O–), 88.8 (C–O–), 107.7 (). (Found: C, 66.71; H, 8.72. $C_{22}H_{34}O_6$ requires: C, 66.98; H, 8.69%.)

2, 4-*Acetonide of* **1**. A soln of **1** (8 mg), 2, 2-dimethoxypropane (2 ml) and trace of *p*-TsOH was stirred at room temp. for 3 hr to give the acetonide (3 mg), an amorphous powder, FDMS *m/z*: 392 $[M]^+$.

Cinncassiol D_4 glucoside (**2**). An amorphous powder; $[\alpha]_D^{26}$ − 12.5° (MeOH; *c* 0.88); FDMS *m/z*: 514 $[M]^+$; IR ν_{max}^{KBr} cm^{-1}: 3400 (OH); ^{1}H NMR (C_5D_5N): δ 1.01 (3H, *d*, *J* = 6 Hz, 18(19)-Me), 1.19 (3H, *d*, *J* = 8 Hz, 18(20)-Me), 1.23 (3H, *s*, 9-Me), 1.43 (3H, *d*, *J* = 7 Hz, 1-Me), 1.69 (3H, *s*, 12-Me), 3.21 (1H, *dd*, *J* = 9, 15 Hz, 3-H), 4.40 (1H, *d*, *J* = 2 Hz, 6-H), 4.74 (1H, *d*, *J* = 7 Hz, 1′-H). (Found: C, 60.48; H, 8.31. $C_{26}H_{42}O_{10}$ requires: C, 60.68; H, 8.23%.)

Enzymatic hydrolysis of **2**. A soln of **2** (15 mg), crude hesperidinase (10 mg, Tanabe Co., Ltd.) in H_2O (3 ml) was incubated for 5 hr at 37°. The products were separated on a cellulose column ($CHCl_3$–MeOH–H_2O, 9:2:0.2 to 7:3:0.5) to give cinncassiol D_4 (**1**) (5 mg) and D-glucose (4 mg), $[\alpha]_D^{18}$

+69.2° (H_2O, *c* 0.39), R_f 0.39 (Si gel; $CHCl_3$–MeOH–Me_2CO–H_2O, 3:3:3:1).

2',3',4',6'-Tetraacetyl cinncassiol D_4 glucoside (**6**). Compound **2** (15 mg) was acetylated with Ac_2O (2 ml) and pyridine (3 ml) for 30 min at room temp. to give the acetate (**6**) (11 mg). An amorphous powder; $[\alpha]_D^{21}$ −5.7° (MeOH; *c* 1.05), EIMS *m/z*: 682 $[M]^+$, 664, 646, 603, 587, 331, 316, 298, 257, 255, 239, 176, 169, 157, 115, 109, ^1H NMR (C_5D_5N): δ 1.04, 1.21 (each 3H, *d*, *J* = 7 Hz, 18-Me_2), 1.28 (3H, *d*, *J* = 7 Hz, 1-Me), 1.31 (3H, *s*, 9-Me), 1.72 (3H, *s*, 12-Me), 1.98, 2.00, 2.05 (12H, all *s*, 4× –OAc), 4.38 (1H, *br s*, 6-H); ($CDCl_3$): δ 0.94 (6H, *d*, *J* = 7 Hz, 18-Me_2), 0.98 (3H, *s*, 9-Me), 1.02 (3H, *d*, *J* = 6 Hz, 1-Me), 2.00, 2.04, 2.06 (12H, all *s*, 4× –OAc), 3.75 (1H, *br s*, 6-H), 4.51 (1H, *d*, *J* = 7 Hz, 1'-H).

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DITERPENES FROM *BALLOTA* SPECIES

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Key Word Index—*Ballota andreuzziana*; *B. pseudodictamnus*; Labiatae; diterpenes; hispanolone; ballonigrin; 18-hydroxyballonigrin; marrubenol; 5-hydroxy-7, 4'-dimethoxyflavone.

Abstract—Hispanolone was isolated from *Ballota andreuzziana*; *B. pseudodictamnus* contains ballonigrin, 18-hydroxyballonigrin, marrubenol, and the flavone 7, 4'-di-*O*-methylapigenin.

During a chemotaxonomic investigation of the genus *Ballota*, we have reported several new furanoid diterpenes in *B. nigra* [1–3], *B. rupestris* [2, 4], *B. hispanica* [5, 6], *B. lanata* [7] and *B. acetabulosa* [8]. Continuing this work, we have now extracted the species *B. andreuzziana* Pampan. and *B. pseudodictamnus* (L.) Bentham.

Usual chromatographic work-up of the acetone extract of the aerial part of *B. andreuzziana*, collected in Cyrenaica (Libya), gave only one diterpene, identified as the known hispanolone **1** occurring in *B. hispanica* [5].

Examination of a sample of *B. pseudodictamnus*, collected in Cyrenaica (Libya), yielded three known diterpenes; ballonigrin **2**, also occurring in *B. nigra* [2], *B. rupestris* [2], *B. lanata* [7]; 18-hydroxyballonigrin **3**, isolated from *B. acetabulosa* [8]; marrubenol **4** (in traces), previously found in *Marrubium vulgare* (Labiatae) [9]. From the same source, we isolated the known 7, 4'-di-*O*-methylapigenin (5-hydroxy-7, 4'-dimethoxyflavone) [10]. Another sample of *B. pseudodictamnus*, collected in Greece near Athens, contained the same products, but marrubenol occurred in rather richer amounts.