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CONIFERYL AND SINAPYL ALCOHOL DERIVATIVES FROM *LIGULARIA DUCIFORMIS*

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Key Word Index—*Ligularia duciformis*; Compositae; roots; coniferyl alcohol derivatives; sinapyl alcohol derivatives.

Abstract—Investigation of *Ligularia duciformis* roots afforded four novel coniferyl alcohol derivatives and two new sinapyl alcohol derivatives: 4-*O*-[6-hydroxy-7(9)-dehydro-6,7-dihydrogeranyl]-coniferyl alcohol, 4-*O*-[7-hydroxy-5,6*E*-dehydro-6,7-dihydrogeranyl]-coniferyl alcohol, 4-*O*-[6-hydroperoxy-7(9)-dehydro-6,7-dihydrogeranyl]-coniferyl alcohol, 4-*O*-[7-hydroperoxy-5,6*E*-dehydro-6,7-dihydrogeranyl]-coniferyl alcohol, 4-*O*-[6-hydroxy-7(9)-dehydro-6,7-dihydrogeranyl]-sinapyl alcohol and 4-*O*-[7-hydroxy-5,6*E*-dehydro-6,7-dihydrogeranyl]-sinapyl alcohol. © 1997 Elsevier Science Ltd

INTRODUCTION

The genus *Ligularia* has been studied by our group for several years. Sesquiterpenes, especially eremophilane compounds appear to be characteristic of this genus [1–6]. The constituents of *L. Duciformis*, grown in mid-China, which is often used in folk medicine for reducing inflammation and for curing apoplexy [7] are described in the present paper.

RESULTS AND DISCUSSION

Ten compounds were isolated by repeated column chromatography and preparative TLC from the petrol (60–90°)–diethyl ether–methanol (1:1:1) extract of the roots of *L. duciformis*. They were identified as β-sitosterol, stigmasterol, 4-*O*-geranyl-coniferyl alcohol (**1**) and 4-*O*-geranyl-sinapyl alcohol (**6**) [8], as well as six new coniferyl and sinapyl alcohol derivatives related to geranyl (**2**–**5**, **7** and **8**).

The ^{1}H NMR spectrum of **2** (Table 1) showed signals for a coniferyl alcohol moiety. The EI-mass spectrum exhibited a significant base peak at m/z 180 due to a coniferyl alcohol fragment and the $[M]^+$ at m/z 332, revealing its molecular formula as $C_{20}H_{28}O_4$, which was confirmed by ^{13}C NMR and DEPT spectra (Table 2). Furthermore, comparison between the ^{1}H NMR spectrum of **2** with that of **1** [8] showed that they were similar in structure, only differing in the side-chain. Instead of the dimethylethenyl signals in **1**, signals for a isopropenyl and an oxygenated methine CHOR were observed in **2**. Thus, **2** was identified as 4-*O*-[6-hydroxy-7(9)-dehydro-6,7-dihydrogeranyl]-coniferyl alcohol, which has an identical side-chain to 6-hydroxy-7(9)-dehydro-6,7-dihydrogeranyl acetate [9]. This conclusion was confirmed by 2D ^{1}H-^{1}H COSY, HMQC and HMBC spectra (Table 3), which were used to confirm spectral assignments and connectivities.

Compound **3** had the same $[M]^+$ (m/z 332) and base peak (m/z 180) in the EI mass spectrum as those of **2**, corresponding to the same molecular formula $C_{20}H_{28}O_4$. Their ^{1}H and ^{13}C NMR spectra (Tables 1 and 2) showed an identical coniferyl alcohol moiety, but there were some differences in the side-chains, indicating the presence of a *trans*-CH=CHC$(CH_3)_2$OH terminal in **3**. Thus, compound **3** was 4-*O*-[7-hydroxy-5,6*E*-dehydro-6,7-dihydrogeranyl]-coniferyl alcohol.

The ^{1}H and ^{13}C NMR spectra of compounds **4** and **5** (Tables 1 and 2), which could not be separated, indicated that they were also coniferyl alcohol derivatives, since their spectra were close to those of **2** and **3**. However, two broadened singlets at δ 8.18 and 8.75 in the ^{1}H NMR spectrum indicated two hydroperoxide moieties, which was confirmed by two remarkably downfield-shifted signals at δ 88.80 (CH) and 81.86 (C) in the ^{13}C NMR spectrum and reduction with triphenylphosphine [10]. The R_f values of the reductive products were identical with those of **2** and **3**, respectively. The ^{1}H and ^{13}C NMR spectra could be assigned by comparison with those of **2** and **3**, as

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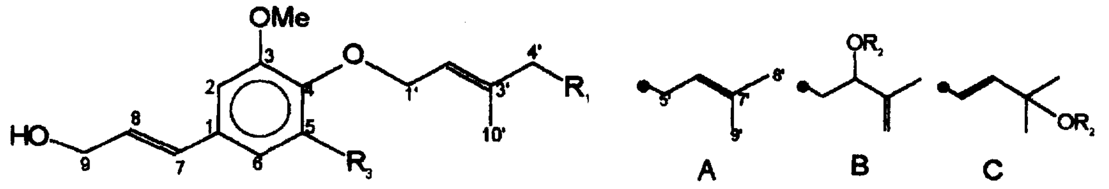
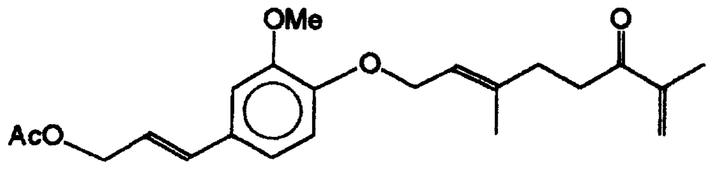


	1/6	2/7	3/8	4/9	5/10
R_1	A	B	C	B	C
R_2	-	H	H	OH	OH
R_3	1 - 5: H;		6 - 10: OMe		



4a

Table 1. ^{1}H NMR spectral data for compounds **2**–**5**, **7** and **8** (400 MHz, $CDCl_3$, TMS as internal standard)

H	**2**	**3**	**4**	**4a**	**5**	**7**	**8**
2	6.95 *d*	6.95 *d*	6.93 *d*	6.93 *d*	6.93 *d*	6.59 *br s*	6.60 *br s*
5	6.82 *d*	6.83 *d*	6.81 *d*	6.82 *d*	6.81 *d*	—	—
6	6.90 *dd*	6.91 *dd*	6.89 *dd*	6.90 *dd*	6.89 *dd*	6.59 *br s*	6.60 *br s*
7	6.55 *dt*	6.55 *dt*	6.53 *dt*	6.61 *dt*	6.53 *dt*	6.52 *dt*	6.54 *dt*
8	6.28 *dt*	6.25 *dt*	6.23 *dt*	6.18 *dt*	6.23 *dt*	6.28 *dt*	6.30 *dt*
9	4.31 *dd*	4.31 *dd*	4.30 *dd*	4.71 *dd*	4.30 *dd*	4.32 *dd*	4.32 *dd*
1′	4.63 *br d*	4.62 *br d*	4.60 *br d*	4.62 *br d*	4.60 *br d*	4.54 *br d*	4.55 *br d*
2′	5.53 *br t*	5.54 *br t*	5.55 *br t*	5.53 *br t*	5.55 *br t*	5.58 *br t*	5.57 *br t*
4′	2.10 *m*	2.75 *br d*	2.09 *m*	2.31 *br t*	2.78 *br d*	2.06 *m*	2.73 *br d*
5′	1.68 *m*	5.61 *dt*	1.59 *m*	2.78 *br t*	5.67 *dt*	1.69 *m*	5.59 *dt*
6′	4.04 *br t*	5.65 *br d*	4.25 *br t*	—	5.53 *br d*	4.02 *br t*	5.63 *br d*
8′	1.72 *br s*	1.31 *s*	1.71 *br s*	1.86 *br s*	1.33 *s*	1.72 *br s*	1.32 *s*
9′	4.93 *br s*	1.31 *s*	4.99 *br s*	5.94 *br s*	1.33 *s*	4.92 *br s*	1.32 *s*
	4.84 *br s*	—	4.97 *br s*	5.75 *br s*	—	4.82 *br s*	—
10′	1.74 *br s*	1.71 *br s*	1.73 *br s*	1.72 *br s*	1.72 *br s*	1.64 *br s*	1.64 *br s*
OOH	—	—	8.75 *br s*	—	8.18 *br s*	—	—
OMe	3.89 *s*	3.89 *s*	3.88 *s*	3.85 *s*	3.88 *s*	3.87 *s*	3.87 *s*
OAc	—	—	—	2.10 *s*	—	—	—

J(Hz): compounds **2**–**5**: 2,6 = 1.5; 5,6 = 8.1; compounds **2**–**5**, **7** and **8**: 7,8 = 15.8; 8,9 = 6.0; 7,9 = 1.0; 1′,2′ = 6.5; compounds **2**, **4** and **7**: 5′,6′ = 6.0; compound **4a**: 4′,5′ = 7.5; compounds **3**, **5** and **8**: 4′,5′ = 5.8; 5′,6′ = 16.0.

well as the known compounds, 6-peroxy-7(9)-dehydro-6,7-dihydrogeranyl acetate, 7-peroxy-5,6*E*-dehydro-6,7-dihydro-geranyl acetate [9] and (*E*)-7-(6-hydroperoxy-3,7-dimethyl-2,7-dienyloxy)-coumarin [11]. Further evidence for this assignment was obtained by acetylation of the mixture which gave **4a**, as it is known that, after acetylation, only a hydroperoxy group could be turned into a keto function by terminal elimination of acetic acid [12, 13].

The ^{1}H NMR spectra of compounds **7**–**10** suggested the presence of a sinapyl alcohol moiety, with the signals for the side chains corresponding with those

Table 2. ^{13}C NMR spectral data of compounds **2–5**, **7** and **8** (400 MHz, $CDCl_3$)

C	**2**	**3**	**4**	**4a**	**5**	**7**	**8**
1	129.9	129.9	123.0	129.5	130.0	132.3	132.3
2	109.3	109.2	109.3	109.3	109.3	103.3	103.5
3	149.6	149.5	149.5	149.5	149.5	153.6	153.7
4	148.2	148.1	148.0	148.1	148.0	136.3	136.6
5	113.3	113.3	113.2	113.3	113.4	153.6	153.7
6	119.6	119.5	119.5	119.5	119.5	103.3	103.5
7	131.2	131.1	131.0	134.1	131.0	131.1	131.2
8	126.6	126.5	126.6	122.0	126.6	127.8	127.9
9	63.8	63.8	63.6	64.9	63.6	63.5	63.6
1′	65.9	65.8	65.7	65.7	65.8	69.2	69.3
2′	120.1	120.6	120.1	121.2	120.7	120.6	121.2
3′	140.4	139.3	140.0	140.2	139.3	141.0	140.0
4′	35.5	42.1	35.5	35.8	42.3	35.4	42.2
5′	32.8	124.1	28.7	33.7	128.3	32.5	124.5
6′	75.4	140.1	88.8	201.3	135.3	75.2	139.8
7′	147.4	70.6	143.8	144.4	81.9	147.2	70.8
8′	17.6	29.7	17.0	17.5	24.2	17.4	29.7
9′	111.1	29.7	113.9	120.5	24.2	111.0	29.7
10′	16.6	16.6	16.6	16.6	16.4	16.1	16.3
OMe	55.9	55.8	55.8	55.7	55.8	56.0	56.1
OAc	—	—	—	*	—	—	—

* δ 170.75 (C=O), 20.71 (Me).

Table 3. HMBC correlations of compound **2**

	HMBC
C-1	H-7, H-8
C-3	H-5, OC$\underline{H}_3$
C-4	H-2, H-1′
C-3′	H-10′, H-4′
C-7′	H-8′, H-6′
C-2′	H-10′, H-1′
C-5′	H-4′
C-9′	H-8′

of **2–5**. Among them, the structure of **8** had been reported [14], but it was not consistent with its NMR spectra. Based on the NMR data, compound **4** [14] needs to be revised to **9**. In addition, compound **1** [14] must also be revised to **10** [15].

EXPERIMENTAL

General. ^1H and ^{13}C NMR spectra were recorded on a Bruker AM 400FT-NMR spectrometer with TMS as int. standard. MS data were obtained at 70 eV. Silica gel (200–300 mesh) was used for CC and silica gel GF_{254} for TLC. Spots were detected on TLC under UV or by heating after spraying with 5% H_2SO_4.

Plant material. Roots of *L. duciformis* were collected in Shengnongja, Hubai Province, in 1994, and identified by Prof. Z. N. Zhao of the Wuhan Institute of Botany, Chinese Academy of Sciences. A voucher specimen (949601) is deposited in the Chemistry Department of Lanzhou University.

Extraction and isolation. Air-dried roots (2.5 kg) were pulverized and extracted at room temp. with petrol (60–90°)–Et_2O–MeOH (1:1:1) (7 days × 3). The resultant extract was concd to a residue (60 g), which was sepd by CC over 900 g silica gel (200–300 mesh) with a gradient of petrol–EtOAc (50:1–0:1). Six frs were collected and from the second, β-sitosterol (100 mg) and stigmasterol (40 mg) were obtained. The third fr. (petrol–EtOAc, 8:1) was isolated by repeated CC on silica gel with first CH_2Cl_2–EtO (40:1–20:1), then C_6H_6–Me_2CO (8:1–2:1). Finally, **1** (30 mg) was purified by prep. TLC (silica gel GF_{254}) with C_6H_6–Me_2CO (5:1) and **2** (20 mg) and **3** (25 mg) sepd by prep. TLC with C_6H_6–Me_2CO (3:1). The mixt. of **4** and **5** (25 mg) was obtained by prep. TLC with C_6H_6–EtOAc (3:1). Fr. 4 (petrol–EtOAc, 5:1) gave **6** (28 mg), **7** (15 mg) and **8** (18 mg) by repeated CC on silica gel with C_6H_6–Me_2CO (15:1–2:1) and prep. TLC (× 2) on silica gel GF_{254} with petrol–Me_2CO (3:1).

4-O-[6-*Hydroxy*-7(9)-*dehydro*-6,7-*dihydrogeranyl*]-*coniferyl alcohol* (**2**). Colourless oil. IR ν_{max}^{Film} cm^{-1}: 3354, 2940, 2867, 1651, 1512, 1454, 1417, 1377, 1261, 1221, 1137, 1054, 1032, 1016, 902, 858. MS m/z (rel. int.): 332 $[M]^+$ (0.5), 180 $[M-C_{10}H_{15}OH]^+$ (100), 152 (15), 137 (55), 124 (46), 107 (12), 93 (19), 91 (18), 81 (16), 67 (20), 55 (22). UV λ_{max}^{MeOH} nm (log ε): 218 (3.56), 262 (2.81). ^1H and ^{13}C NMR: Tables 1 and 2.

4-O-[7-*Hydroxy*-5,6E-*dehydro*-6,7-*dihydrogeranyl*]-*coniferyl alcohol* (**3**). Colourless oil. IR ν_{max}^{Film} cm^{-1}: 3388, 3312, 2946, 2835, 1650, 1511, 1454, 1416, 1262, 1229, 1118, 1032, 898, 840. MS m/z (rel. int.): 332 $[M]^+$

(0.4), 314 $[332-H_2O]^+$ (0.4), 180 $[M-C_{10}H_{15}OH]^+$ (100), 152 (18), 137 (60), 124 (46), 109 (16), 107 (16), 93 (23), 91 (25), 81 (39), 67 (18), 55 (28). UV λ_{max}^{MeOH} nm (log ε): 218 (3.74), 265 (3.00). ^1H and ^{13}C NMR: Tables 1 and 2.

4-O-[6-*Hydroperoxy*-7(9)-*dehydro*-6,7-*dihydrogeranyl*]-*coniferyl alcohol and* 4-O-[7-*hydroperoxy*-5,6E-*dehydro*-6,7-*dihydro-geranyl*]*coniferyl alcohol* (**4** *and* **5**). Colourless oil, which could not be resolved. IR ν_{max}^{Film} cm^{-1}: 3435, 3288, 2956, 2843, 1646, 1511, 1454, 1415, 1379, 1263, 1220, 1120, 1018, 890. MS *m/z* (rel. int.): 348 $[M]^+$ (1.2), 314 $[M-HOOH]^+$ (0.4), 180 $[M-C_{10}H_{15}OOH]^+$ (100), 152 (15), 124 (40), 109 (15), 93 (22), 81 (35), 67 (14), 55 (25). UV λ_{max}^{MeOH} nm (log ε): 214 (2.87), 263 (1.52). ^1H and ^{13}C NMR: Tables 1 and 2. Reaction of the mixt. (5 mg) with triphenylphosphine in $CDCl_3$ afforded **2** and **3**, identical with the natural compounds (TLC). The mixt. (15 mg) on heating with 2.5 ml Ac_2O (2 hr, 70°) gave 10 mg 4-*O*-[6-oxo-7(9)-dehydro-6,7-dihydro-geranyl]-coniferyl alcohol (**4a**). Colourless oil. ^1H and ^{13}C NMR: Tables 1 and 2.

4-O-[6-*Hydroxy*-7(9)-*dehydro*-6,7-*dihydrogeranyl*]-*sinapyl alcohol* (**7**). Colourless oil. IR ν_{max}^{Film} cm^{-1}: 3384, 2941, 2857, 1651, 1511, 1454, 1410, 1370, 1258, 1220, 1137, 1052, 1034, 1016, 886. MS *m/z* (rel. int.): 362 $[M]^+$ (0.5), 210 $[M-C_{10}H_{15}OH]^+$ (100), 182 (23), 167 (50), 154 (46), 137 (12), 121 (18), 67 (20). ^1H and ^{13}C NMR: Tables 1 and 2.

4-O-[7-*Hydroxy*-5,6E-*dehydro*-6,7-*dihydrogeranyl*]-*sinapyl alcohol* (**8**). Colourless oil. IR ν_{max}^{Film} cm^{-1}: 3408, 2966, 2865, 1655, 1501, 1457, 1418, 1262, 1230, 1128, 1032, 966, 840. MS *m/z* (rel. int.): 362 $[M]^+$ (0.6), 210 $[M-C_{10}H_{15}OH]^+$ (100), 182 (18), 167 (56), 154 (41), 139 (16), 121 (20), 67 (18). ^1H and ^{13}C NMR: Tables 1 and 2.

REFERENCES

1. Jia, Z. J. and Chen, H. M., *Phytochemistry*, 1991, **30**, 3132.
2. Chen, H. M., Jia, Z. J. and Yang, L., *Phytochemistry*, 1992, **31**, 2146.
3. Zhao, Y., Jia, Z. J., Tan, R. X. and Yang, L., *Phytochemistry*, 1992, **31**, 2785.
4. Jia, Z. J., Zhao, Y. and Tan, R. X., *Planta Medica*, 1992, **58**, 365.
5. Jia, Z. J., Zhao, Y. and Tan, R. X., *Journal of Natural Products*, 1993, **56**, 494.
6. Zhao, Y., Jia, Z. J. and Peng, H. R., *Journal of Natural Products*, 1995, **58**, 1358.
7. Beijing Institute of Botany, Chinese Academy of Sciences, *Iconographia Cormophytorum Sinicorum Tomus* IV. Science Press of China, 1975, p. 579.
8. Zdero, C., Jakupovic, J. and Bohlmann, F., *Phytochemistry*, 1990, **29**, 1231.
9. Zdero, C., Bohlmann, F., King, R. M. and Robinson, H., *Phytochemistry*, 1986, **25**, 509.
10. Bohlmann, F., Jakupovic, J., Ahmed, M. and Schuster, A., *Phytochemistry*, 1983, **22**, 1623.
11. Rashid, M. A., Gray, A. I. and Waterman, P. G., *Journal of Natural Products*, 1992, **55**, 851.
12. Bohlmann, F., Adler, A., Jakupovic, J., King, R. M. and Robinson, H., *Phytochemistry*, 1982, **21**, 1349.
13. Sevil, O., Roberto, R. G., Heebyung, C., John, M. P., Geoffrey, A. C. and Ayhan, U., *Plant Medica*, 1994, **60**, 594.
14. Zhao, Y., Peng, H. R. and Jia, Z. J., *Chinese Chemical Letters*, 1995, **6**, 387.
15. Ma, B., Gao, K., Shi, Y. P. and Jia, Z. J., *Phytochemistry*, (accepted for publication).