Pergamon

Phytochemistry, Vol. 49, No. 1, pp. 211–213, 1998

Printed in Great Britain
0031–9422/98/$–see front matter

PII: S0031–9422(97)00951–5

A POLYACETYLENIC ACETATE AND A COUMARIN FROM *ANGELICA PUBESCENS F. BISERRATA*

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(*Received in revised form* 20 *August* 1997)

Key Word Index—*Angelica pubescens f. biserrata*; Umbelliferae; roots; columbianetin propionate; falcarindiol, bisabolangelone; 11(*S*),16(*R*)-dihydroxy-octadeca-9*Z*,17-dien-12,14-diyn-1-yl acetate.

Abstract—11(*S*),16(*R*)-dihydroxy-octadeca-9*Z*,17-dien-12,14-diyn-1-yl acetate and a new coumarin, columbianetin propionate, were isolated from the dichloromethane extract of the roots of *Angelica pubescens f. biserrta*, along with falcarindiol and bisabolangelone. © 1998 Published by Elsevier Science Ltd. All rights reserved

INTRODUCTION

Duhuo (Radix Angelicae pubescentis) has been used in traditional Chinese medicine as a remedy for arthritic disease. We have previously reported ca 30 coumarins and other compounds isolated from the roots of *Angelica pubescens f. biserrata* [1–4]. Some coumarins in this plant were found to be active as inhibitors of thromboxane formation in platelets and phosphoinositide breakdown, and to exert antiproliferatory and relaxant effect on the trachealis [5–9]. We now present the isolation and structural elucidation of further constituents from this source.

RESULTS AND DISCUSSION

The dichloromethane extract of the roots of *A. pubescens* afforded compounds **1–4**. Compound **1** was isolated as colourless needles, showing a blue-violet fluorescence at 365 nm. In the mass spectrum, the ion peak at *m/z* 302 supported the molecular formula $C_{17}H_{18}O_5$. UV and IR spectra exhibited similar patterns to columbianetin derivatives which we had previously isolated [1, 4]. The ^{1}H NMR spectrum presented signals for a 7,8-disubstituted coumarin. Two methyl signals and signals of an ABX-system indicated an angular dihydrofuranocoumarin [10]. Signals at δ 2.25 (2H, *q*, J = 7.50 Hz) and 1.05 (3H, *t*, J = 7.50 Hz) showed the existence of a propionate group. This, compound **1** was presumed to be columbianetin propionate, which is a new natural product. Signals in the ^{13}C NMR and fragments in EI mass spectrum confirmed this assignment.

Compound **4** was obtained as a colourless viscous oil. The ion peak at *m/z* 350 $[M+NH_4]^+$ in the CI mass spectrum supported the molecular formula $C_{20}H_{28}O_4$. The ^{1}H and ^{13}C NMR spectra exhibited signals which resembled those of falcarindiol, except for the signals due to C-1 and the acetate group (Table 1). The configuration of **4** was established by comparison of its ORD spectra with that of falcarindiol; both of them showed a positive curve. Therefore, its

Table 1. ^{1}H (500 MHz) and ^{13}C NMR (125 MHz) data of compound **4** (in $CDCl_3$)

No.	δ_C	δ_H
18	117.3	5.26 (1H, *br d*, J = 10.09 Hz, H-18a), 5.47 (1H, *br d*, J = 17.02 Hz, H-18b)
17	135.8	5.95 (1H, *ddd*, J = 17.02 Hz; 10.09 Hz; 5.04 Hz)
16	63.4	4.94 (1H, *d*, J = 5.04 Hz)
11	58.6	5.21 (1H, *d*, J = 8.20 Hz)
10	127.8	5.52 (1H, *dd*, J = 8.20 Hz; 10.72 Hz)
9	134.5	5.61 (1H, *dt*, J = 10.72 Hz; 7.57 Hz)
8	27.6	2.11 (2H, *dt*, J = 7.57 Hz; 6.93 Hz)
7	29.1	1.39 (2H, *m*)
2	28.6	1.62 (2H, *q*, J = 6.94 Hz)
1	64.7	4.06 (2H, *t*, J = 6.94 Hz)
15–12	79.8 (C-15)	70.2 (C-14) 68.7 (C-13) 78.4 (C-12)
6–3	29.2 (C-6)	29.1 (C-5) 38.9 (C-4) 25.8 (C-3)
$COCH_3$	171.5	
	21.1	2.05 (3H, *s*)

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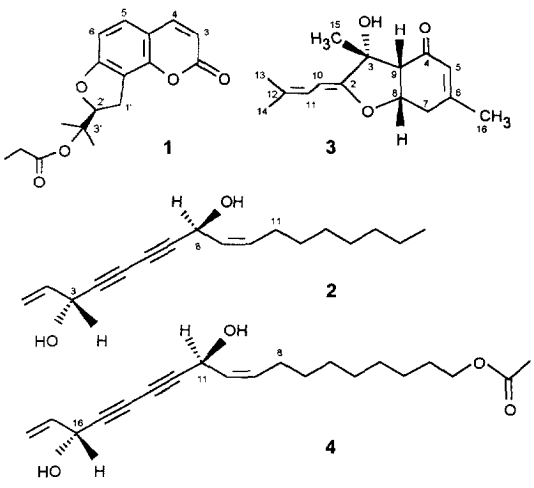

structure was suggested to be 11(*S*),16(*R*)-dihydroxyoctadeca-9*Z*,17-dien-12,14-diyn-1-yl acetate. It is a new compound and exhibits inhibitory activity on 5-lipoxygenase (5-LO) and cyclooxygenase (COX-1) with IC_{50} values of 24 μM and 73 μM, respectively.

Compound **2** was isolated as a viscous oil and was identified by comparison of its R_f value in TLC and R_t in HPLC with that of a reference of falcarindiol in two different solvent systems. Falcarindiol is reported to be an anaesthetic and antifungal principle in medicinal plants [12]. It also exhibits prominent inhibitory effect on 5-LO with an IC_{50} value of 9.4 μM and moderate inhibitory activity on COX-1 with an IC_{50} value of 66 μM.

Compound **3** was obtained as yellow needles. It was identified as bisabolangelone, which has already been reported in *A. silvestris* [13] and *A. koreana* [14], according to the UV, ^{1}H and ^{13}C NMR, ^{1}H-^{1}H and ^{1}H-^{13}C COSY, DEPT and HMBC spectra. The ^{1}H NMR signals corresponded with those for bisabolangelone [13], except for the ABX system at δ 2.78 (1H, *dd*, $J_{7a,7b}$ = 18.29 Hz, $J_{7a,8}$ = 5.05 Hz, H-7a), 2.70 (1H, *dd*, $J_{7a,7b}$ = 18.29 Hz, $J_{7b,8}$ = 5.67 Hz, H-7b) and 4.87 (1H, *ddd*, $J_{8,9}$ = 6.94 Hz, $J_{7,8}$ = 5.05 Hz, $J_{7b,8}$ = 5.67 Hz, H-8), which were originally assigned as 2.74 (2H, *dd*, $J_{8,7\alpha} = J_{8,7\beta}$ = 5.5 Hz, $J_{7\alpha,7\beta}$ = 5.0 Hz) and 4.87 (1H, *dt*, $J_{8,9}$ = 6.7 Hz, $J_{8,7\alpha} = J_{8,7\beta}$ = 5.5 Hz). Signals in ^{13}C NMR were now assigned unambiguously on the basis of ^{1}H-^{13}C COSY and HMBC spectra. Bisabolangelone has been isolated for the first time from *A. pubescens* and is reported to have strong antifeeding properties against insects but is unstable in both basic and acidic media [15].

EXPERIMENTAL

General

Mps are uncorr. NMR spectra were recorded in $CDCl_3$ at 500 MHz (^{1}H) and 125 MHz (^{13}C) with TMS as int. standard. CC was carried out on silica gel (230–400 mesh, Merck). MPLC columns were filled with RP-18 silica gel (25–40 μm, Merck).

Plant material

Roots of *A. pubescens* Maxim *f. biserrata* Shan *et* Yuan were purchased from Shenyang and identified by Prof. Tingguo Kang of Liaoning College of Traditional Chinese Medicine. A voucher specimen is deposited there.

Extraction and isolation

Roots (900 g) were powdered and extracted with 4.51 CH_2Cl_2 in a Soxhlet for 10 h. The solvent was evapd to obtain 46.6 g extract; 15 g of this extract ($\times$ 3) was separated by flash CC (150 g silica gel) with a gradient of petrol–EtOAc (100:0; 95:5; 90:10;... 50:50; MeOH), the elution vol. of each gradient being 300 ml. The 8th fr. (6.1 g) was rechromatographed on a silica gel column with *n*-hexane–EtOAc (17:3) to yield 8 subfrs, and the 8th subfr. was again subjected to MPLC with a gradient of H_2O–MeOH (3:2 → 0:100 within 100 min) to yield compound **1** (6.4; mg). Yellow crystals of compound **3** (32.4 mg) were obtained from the 9th fr. (1.1 g). The 10th fr. was further sepd with *n*-hexane–EtOAc (17:3) to give 12 subfrs. The 5th subfr. was rechromatographed by MPLC using gradient elution with H_2O–MeOH (100.0 → 0:100 within 100 min) to give compound **2** (5.1 mg). The 11th subfr. was purified by MPLC using gradient elution with H_2O–MeOH (7:3 → 0:100 within 100 min) to yield compound **4** (34.7 mg).

Columbianetin propionate (**1**). Colourless needles, mp 117–118°. $C_{17}H_{18}O_5$, $[\alpha]_D^{20}$ = 155.8° (*c* 0.52, $CHCl_3$). UV λ_{max} nm: 325, 260, 251. IR ν (KBr) cm^{-1}: 1730, 1615, 1580, 1275, 840. EI-MS (70 eV) *m/z* (rel. int.): 302 $[M]^+$, 246, 228, 213 (100), 187, 176, 131, 115, 102, 77, 57, 43. ^{1}H NMR: δ 7.64 (1H, *d*, *J* = 9.5 Hz, H-4), 6.22 (1H, *d*, *J* = 9.5 Hz, H-3), 7.31 (1H, *d*, *J* = 8.2 Hz, H-5), 6.75 (1H, *d*, *J* = 8.2 Hz, H-6), 3.38 (1H, *dd*, *J* = 16.4 Hz; 10.1 Hz, H-1′a), 3.30 (1H, *dd*, *J* = 16.4; 7.5 Hz, H-1′b), 5.14 (1H, *dd*, *J* = 7.5 Hz; 10.1 Hz, H-1′), 1.58 (3H, *s*, H-4′), 1.52 (3H, *s*, H-5′), 2.25 (2H, *q*, *J* = 7.5 Hz, $COC\underline{H}_2CH_3$), 1.05 (3H, *t*, *J* = 7.5 Hz, $COCH_2C\underline{H}_3$). ^{13}C NMR: δ 163.9 (C-2), 112.2 (C-3), 144.0 (C-4), 128.8 (C-5), 106.7 (C-6), 161.06 (C-7), 113.0 (C-8), 151.3 (C-9), 113.5 (C-10), 27.6 (C-1′), 88.9 (C-2′), 81.9 (C-3′), 21.0 (C-4′), 22.1 (C-5′), 29.7 (CH_2), 9.1 (CH_3), 173.6 (C=O).

Falcarindiol (**2**). Viscous oil. ORD (*c* = 0.022, MeCN), $[\alpha]_D^{20}$ (nm): +321.4° (589), +321.4° (578), +178.6° (546), +392.9° (436), +678.6° (365). UV λ_{max} nm: 228, 244, 257, 267, 282.

Bisabolangelone (**3**). Yellow needles, mp 147–148°. $[\alpha]_D^{20}$ 198° (*c* = 0.064, EtOH). UV λ_{max} nm: 252. ^{1}H NMR: δ 6.0 (2H, *m*, $J_{10,11}$ = 11.35 Hz, H-5, H-11), 5.37 (1H, *d*, *J* = 11.35 Hz, H-10), 4.87 (1H, *ddd*, $J_{8,9}$ = 6.94 Hz, $J_{7a,8}$ = 5.05 Hz, $J_{7b,8}$ = 5.67 Hz, H-8),

2.78 (1H, *dd*, $J_{7a,7b}$ = 18.29 Hz, $J_{7a,8}$ = 5.05 Hz, H-7a), 2.70 (1H, *dd*, $J_{7a,7b}$ = 18.29 Hz, $J_{7b,8}$ = 5.67 Hz, H-7b), 2.65 (1H, *d*, J = 6.94, H-9), 2.02 (3H, *s*, 6-CH_3), 1.79 (3H, *s*, 12-CH_3), 1.72 (3H, *s*, 12-CH_3), 1.62 (3H, *s*, 3-CH_3). ^{13}C NMR: δ 196.8 (C-4), 160.0 (C-6), 158.2 (C-2), 132.6 (C-12), 127.2 (C-5), 117.7 (C-11), 94.4 (C-10), 78.6 (C-3), 76.1 (C-8), 53.6 (C-9), 34.9 (C-7), 27.4 (C-15), 26.0 (C-13), 24.6 (C-16), 18.2 (C-14).

11(*S*),16(*R*)-*dihydroxy-octadeca-9Z,17-dien-12,14-diyn-1-yl acetate* (**4**). Colourless viscous oil. $C_{20}H_{28}O_4$. ORD (*c* 0.028, MeCN), $[\alpha]_D^{20}$ (nm): +318.2° (589), +272.7° (578), +227.3° (546), +545.5° (436), +818.2° (365). UV λ_{max} nm: 228, 245, 258, 280, 320. CI MS *m/z*: 350 $[M+NH_4]^+$ 332 $[M]^+$, 314, 198, 262, 256, 244. ^{1}H and ^{13}C NMR: Table 1.

Acknowledgements—J.-H.L. is grateful to the Alexander von Humboldt Foundation for a fellowship. We thank Dr Matthiesen, Central Institute of Clinical Chemical Laboratory Diagnostics and the NMR Centre of the Institute of Inorganic Chemistry, both University of Düsseldorf, for recording MS and NMR, respectively.

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