Chemistry of Natural Compounds, Vol. 43, No. 6, 2007

TWO NEW COMPONENTS FROM THE ROOTS OF *Angelicae koreana* KITAGAWA

Shin Yoosoo,[1] Lee Ogyu,[2] Park Chungberm,[1] Lee Hakju,[2]
Choi Bongjae,[3] Ham Inhye,[3] Choi Hoyoung,[3]
Seong Naksul,[1] and Cha Sunwoo[1]

UDC 547.972+547.1

Two new compounds, 2-hydroxyethylcarboxylic acid-5,9-eicodien-carboxylate and 1-(2′-propenoxy)undec-2-ene, and two known compounds, 2-hydroxy-4-methyl-acetophenone and β-sitosterol, were isolated from the roots of Angelicae koreana *Kitagawa. The structures of the compounds were established on the basis of MS and NMR spectral data.*

Key words: *Angelicae koreana*, index components, phenolic compound, 2-hydroxyethylcarboxylic acid-5,9-eicodien-carboxylate, 1-(2′-propenoxy)undec-2-ene.

The roots of *Angelica koreana* Kitagawa (Umbelliferae) have been used as a folk medicine for the treatment of paralysis and joint disease in Far East countries such as China, Korea, and Japan. Similar plants of *A. koreana* such as *Notopterygium incisium* Ting and *N. forbesii* Boiss from China and *Ostericum koreana* Max from Korea have been reported, but *A. Koreana* is used mainly in the last decade [1].

Several coumarin derivatives (aesculin, imperatorin, isoimperatorin, oxypeucedanin, isooxypeucedanin, oxypeucedanin-hydrate, -methanolate osthol, prangolarin, xanthotoxol, anomalin, bergapten kahorinin, marmesinin, 4′-*O*-*β*-D-glucopyranosyl-3′-hydroxymarmesin), sesquiterpenes (bisabolangelone, *α*-bisabolol), and essential oils (*α*-pinene, *β*-myrcene, *α*-phellandrene, 3-carene, *p*-cymene, sabinene, cresol, terpinolene, 4-vinyl-2-methoxyphenol, *β*-maaliene, *β*-eudesmol, 4-hydroxy-2-methylacetophenone 1,2-benzenedicarboxylic acid-*bis*(2-ethylhexyl)ester) and others (caffeic acid, cimifugin, feruric acid, uracil, adenosine, *β*-sitosterol) have been isolated from *A. koreana* so far. Among these compounds, coumarin derivatives are reported as the main compounds with biological activity such as cytotoxic activity [2] and TNF-α inhibitory effect [3].

Recently, two species of *A. koreana* Kitagawa, Bukkanghwal (BKH) and Namkanghwal (NKH), were circulated in commercial markets in Korea. But studies on the chemical constituents and index components of the two plants were not reported. We isolated two new compounds and four known compounds from BKH and NKH and investigated the presence of an index component of the two plants.

Two new compounds (**2** and **4**) and two known compounds were isolated from the HEA fraction of the EtOH extract of BKH, and two known compounds from the CME fraction of the EtOH extract of NKH. This paper described the structural determination of the two new compounds (**2** and **4**) on the basis of spectroscopic studies and the presence of 2-hydroxy-4-methyl-acetophenone (**1**) isolated from BKH for the first time.

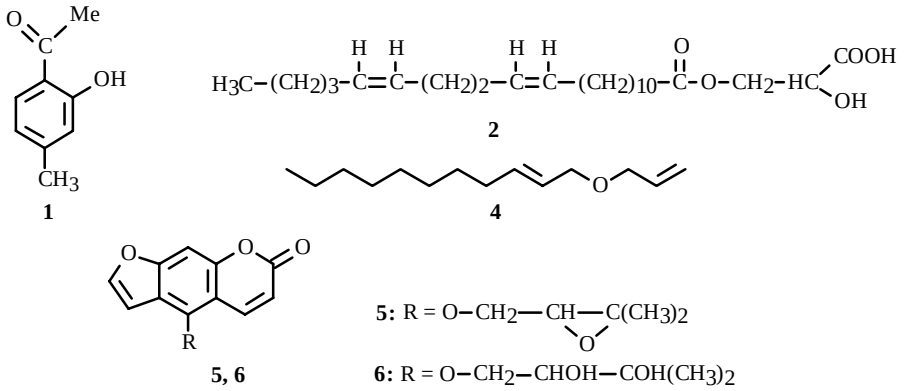
Compounds **1** to **4** from the HEA fraction of the roots of BKH and compound I and II from the CME fraction of the roots of NKH were isolated by repetitive column chromatography with HEA solvent.

Compound **1** had the molecular formula $C_9H_{10}O_2$ as determined from the EI-HR-MS, ^{13}C NMR, and DEPT spectral data and was identified as 2-hydroxy-4-methyl-acetophenone by comparison with literature data. The 4-hydroxy-2-methylacetophenone from the roots of *A. koreana* was reported by Choi et al. [4], but 2-hydroxy-4-methylacetophenone was first isolated as a chemical constituent of BKH.

Compound **3**, **5**, and **6** were identified as *β*-sitosterol, oxypeucedanin, and oxypeucedanin hydrate by comparison with MS and NMR data in the literature [1].

1) National Institute of Crop Science, RDA, Suwon, 441-857, Korea, Fax : 82 31 290 6812, e-mail: totoro@rda.go.kr; 2) Korea Forest Research Institute, Seoul, 130-712, Korea; 3) College of Oriental Medicine, Kyunghee University, Seoul, 130-701, Korea. Published in Khimiya Prirodnykh Soedinenii, No. 6, pp. 540-542, November-December, 2007. Original article submitted October 20, 2006.

 0009-3130/07/4306-0652



Compound **2** had the molecular formula $C_{24}H_{42}O_5$ as determined from the EI-HR-MS, ^{13}C NMR, and DEPT spectral data. In the ^1H NMR spectrum of compound **2**, five multiplets at δ 5.36 to δ 5.40 were assigned to five methine groups. The peaks at δ 4.18, 4.20, 4.35, and 4.37 (dddd) bearing oxygen were assigned to one methylene group. Three multiplets at δ 2.83, 2.35, and 1.67 were assigned to three methylene groups, and one multiplet at δ 2.11 was assigned to two methylene groups. Also, the multiplets of strong intensity at δ 1.37 were assigned to ten methylene groups. One multiplet that appeared at δ 0.95 showed the presence of one methyl group.

In the ^{13}C NMR and DEPT spectra of compound **2**, the signal of one methyl group appeared in δ 14.1, supporting the results obtained by ^1H NMR. Also, the signals appeared in a lower field (δ 34.2, 31.8, 29.6 (×10), 27.4 (×2), 25.8, 25.1, 22.8) than that of methyl groups that contain 17 methylene groups. The two signals at δ 62.2 and 69.2 corresponded to a methylene group and methine group bearing the oxygen or hydroxyl group, respectively. The four signals that appeared in the low field at δ 130.2, 130.0, 128.3, and 128.1 were derived from two double bonds. The two signals at δ 173.1 and 172.7 were assigned to a carboxyl group and ester group. In the ^1H–^1H COSY, the methine proton bearing the oxygen or hydroxyl group at δ 5.36 (m) correlated with the peak of a methylene proton bearing oxygen at δ 4.18, 4.20, 4.35, and 4.37 (dddd). A correlation between the two methine protons at δ 5.36–5.40 (m) and methylene protons δ 2.83 (m) was observed. Correlations between the two methine protons at δ 5.36–5.40 (m) and methylene protons at δ 2.11 (m), and between methylene protons at δ 2.83 (m) and one methylene proton δ 2.11 (m) were also observed. The data showed the partial structure of -CH=CH-$(CH_2)_2$-CH=CH-CH_2- of compound **2**. Correlations between the two methylene protons at δ 2.35 (m) and 1.67 (m), 1.67 (m) and 1.37 (m), and the methyl group at δ 0.95 (m) and methylene protons at δ 1.37 (m) were observed. The data showed the partial structure of -CH_2-CH_2-CH_2-CH_3 of compound **2**.

In the HMBC spectrum, the correlations between the methine proton at δ 5.36 and two carbon (δ 172.7 and 62.2), methylene protons at δ 4.18, 4.20, 4.35, 4.37 (dddd) and three carbon (δ 62.2, 69.2 and 173.1) and methylene protons (δ 2.35 and 1.67) and three carbon (δ 173.1, 34.2, 29.6), showed the partial structure of –CH_2-CH_2-COO-CH_2-CHOH-COOH. On the basis of the data, we concluded that compound **2** is 2-hydroxyethylcarboxylic acid-5,9-eicodien-carboxylate.

Compound **4** exhibited a molecular ion peak at *m/z* 208 in EI-MS and its formula is $C_{14}H_{24}O$ by EI-HR-MS. In the ^1H NMR spectrum of compound **4**, three multiplets at methine protons at δ 5.53, 5.60, and 5.93 were assigned to three methine groups. The peaks at δ 5.26 and 5.48 were assigned to one methylene group of the double bond. Also, the two peaks at 4.93 (d, J = 5.5) and 5.20 (tt) bearing oxygen were assigned to two methylene groups. Six multiplets at δ 1.28 to 2.11 were assigned to seven methylene groups, and one multiplet at δ 2.11 (m) was assigned to one methylene group. Also one multiplet that appeared at δ 0.89 showed the presence of one methyl group.

In the ^1H–^1H COSY, the two methylene protons bearing oxygen at δ 5.20 (tt) and 4.93 (d, J = 5.5) correlated with the peak of a methine proton at δ 5.53 (m) and 5.93 (m), respectively. Also, two correlations between the peak at δ 5.53 (m) and 5.60 (m), and at δ 5.93 (m) and 5.26, 5.48 (tt) of methylene protons of the double bond were observed. The methine proton at δ 5.60 (m) correlated with the methylene protons at δ 2.11 (m), and the methylene protons at δ 2.11 (m) correlated with the methylene protons at δ 1.28 (m). A correlation between the methylene protons at δ 1.28 (m) and methyl protons at δ 0.89 (t) was observed.

The data showed the partial structure of propenoxy at C-1 and undecane bearing a double bond at C-2 of compound **2**. The HMBC spectrum of compound **4** proved the above results of the COSY spectrum. On the basis of the data, we concluded that compound **4** is 1-(2′-propenoxy)undec-2-ene.

EXPERIMENTAL

General Experimental Procedures. The NMR spectra were measured on a Varian FT-NMR 500 MHz using deuterated chloroform ($CDCl_3$) as solvent and tetramethylsilane (TMS) as an internal standard. Two-dimensional (2D) NMR was performed with ^1H-^1H COSY, DEPT, HQMC, and HMBC. EI-MS and EI-HR-MS spectra were obtained using a JEOL JMS-AX500 mass spectrometer. Thin-layer chromatography (TLC) was perfomed on silica gel 60 F_{254}.

Plant Material. The roots of two species of *A. koreana* Kitagawa, which is called Bukkanghwal (BKH) and Namganghwal(NKH), were collected in May 2005 in the Jeonseong-goon area, Gangwon-do, Korea and dried at room temperature for 1 month. The material was confirmed taxonomically by Professor H. Y. Choi at the College of Oriental Medicine, Kyunghee University, in Seoul, Republic of Korea. A voucher specimen has been deposited at Kyunghee University and the National Institute of Crop Science, RDA, Korea.

Isolation. The powdered roots of two species of *A. koreana* Kitagawa (BKH 870g and NKH 840g) were extracted five times with 95% EtOH at room temperature for 24 h. The EtOH extracts were obtained and concentrated under reduced pressure. The concentrated extracts (BKH 56.1 g and NGH 53.5 g) were successively separated by silica gel column chromatography (CC, Wakogel C-200) with developing solvents of *n*-hexane, *n*-hexane–EtOAc (HEA, 10:1, 4:1, 1:1, v/v), EtOAc, $CHCl_3$–MeOH (CME, 10:1, 4:1, 1:1, v/v), and MeOH, successively. By monitoring with TLC using the developing solvent, (SGIII, toluene–formic acid–ethyl formate, 5:1:4, v/v and CMH, $CHCl_3$–MeOH–H_2O, 14:6:1, v/v), the extractives were separated into several fractions. In BKH, crude 2-hydroxy-4-methylacetophenone (**1**) and compound **2** were obtained from Fraction 3 using a silica gel column (Wakogel C-200) with HEA (10:1, v/v) solvent. Finally, the purified 2-hydroxy-4-methylacetophenone (**1**) (25 mg) and compound **2** (45 mg) were isolated. β-Sitosterol (**3**) (22 mg) was isolated from Fractions 4 and 5 and purified by slilica gel column chromatography using HEA (4:1, v/v) solvent. Compound **4** was isolated from Fraction 7 by silica gel column chromatography using HEA (1:1, v/v) solvent. The yield of compound **4** was 34 mg. In NKH, crude oxypeucedanin (**5**) and oxypeucedanin hydrate (**6**) were obtained from the CME fraction using a silica gel column (Wakogel C-200) with CHA ($CHCl_3$–acetone, 9:1, v/v). The yields of the isolated compounds oxypeucedanin and oxypeucedanin hydrate (**6**) were 5 and 9 mg, respectively.

Compound 1: yellowish oil. EI-MS; *m/z* 150, HR-EI-MS; $C_9H_{10}O_2$, ^1H NMR ($CDCl_3$, δ, J/Hz); 12.28 (s, 2-OH), 7.60 (d, J = 8, 6-H), 6.70 (d, J = 8, 5-H), 6.79 (s, 3H), 2.59 (s, 1′-Me), 2.35 (s, 4′-Me). ^{13}C-NMR ($CDCl_3$); 203.8 (-CO-), 162.7 (C-2), 148.0 (C-4), 130.6 (C-6), 120.2 (C-5), 118.5 (C-3), 117.7 (C-1), 26.4 (C-1′-Me), 21.9 (C-4-Me).

Compound 2: yellowish oil, EI-MS; *m/z* 410, HR-EI-MS; $C_{24}H_{42}O_5$, ^1H NMR ($CDCl_3$, δ); 5.42 (m, 4H), 5.33 (m, -CHOH), 4.18, 4.20, 4.35, 4.37 (dddd, -CH_2O-), 2.35 (m, -CH_2), 2.11 (m, -2CH_2), 2.83 (m, -CH_2), 1.67 (m, -CH_2), 1.37 (m, -10CH_2), 0.95 (t, -CH_3). ^{13}C NMR($CDCl_3$); 173.1 (COOH), 172.7 (COO-), 130.2 (=CH-), 130.0 (=CH-), 128.3 (=CH-), 128.1 (=CH-), 69.2 (-HCOH-), 62.2 (-CH_2O-), 34.2 (-CH_2-), 27.4 (-2CH_2-), 25.8 (-CH_2-), 25.1 (-CH_2-), 29.2 (-10CH_2), 14.1 (CH_3).

Compound 4: yellowish oil, EI-MS; *m/z* 208, HR-EI-MS; $C_{14}H_{24}O$, ^1H NMR ($CDCl_3$, δ): 5.93(ddd, 2′-H), 5.60 (m, 3-H), 4.53 (m, 2-H), 5.26, 5.48 (tt, 3′-2H), 5.20 (tt, 1-H), 4.93(d, J = 5.5, 1′-H), 2.11 (m, -CH_2), 1.28 (m, -6CH_2), 0.89 (t, -CH_3). ^{13}C NMR ($CDCl_3$): 135.8 (2′-C), 134.5 (3-C), 127.6 (2-C), 117.3 (3′-C), 63.4 (1′-C), 58.5 (1-C), 31.7 (-CH_2-), 29.3 (4-CH_2-), 27.6 (-CH_2-), 22.6 (-CH_2-), 14.0 (CH_3).

REFERENCES

1. Kwon Yongsoo, Woo Eunran, and Kim Changmin, *Kor. J. Pharmacogn.*, **22**, 156 (2000).
2. Bae Kihwan, Ji Jongmyung, Kang Jongseong, and Ahn Byungzun, *Arch. Pharm. Res.*, **17**, 45 (1994).
3. Cho Jaeyoul, Lee Jongsoo, Park Jisoo, and Park Myunghwan, *Yakhak Hoeji*, **42**, 125 (1998).
4. H. Y. Choi, Y. B. Suh, and I. H. Ham, *Kor. J. Herbol.*, **19**, 169 (2004).