Pergamon

0031-9422(94)00926-0

Phytochemistry, Vol. 39, No. 1, pp. 231–233, 1995

0031-9422/95 $9.50 + 0.00

A 9,10-DIHYDROPHENANTHRENE DERIVATE FROM *EPIMEDIUM KOREANUM*

WEN-KUI LI, JING-QI PAN,* MU-JIAN LÜ, RU-YI ZHANG† and PEI-GEN XIAO

Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences and Peking Union Medical College, Haidian District, Dong Beiwang, Beijing 100094, China; *Department of Chemistry, Peking University, Beijing 100871, China; †Department of Phytochemistry, School of Pharmaceutical Sciences, Beijing Medical University, Beijing 100083, China

(*Received* 12 *September* 1994)

Key Word Index—*Epimedium koreanum*; Berberidaceae; 9,10-dihydrophenanthrene derivate; epimedoicarisoside A; flavonoids; icariin; epimedoside C; icarisid I; baohuoside I; diphylloside A; baohuoside II; 2″-*O*-rhamnosylicarisid II, sagittatoside A; hyperoside; icaritin-3-*O*-rhamnopyranoside.

Abstract—A new 9,10-dihydrophenanthrene derivative named epimedoicarisoside A was isolated from the aerial parts of *Epimedium koreanum* along with ten known flavonoids. The structure of this compound was determined on the basis of spectral analysis (FAB-Mass spectrometry, ^1H–^1H COSY, ^1H–^{13}C COSY, DEPT, and ^1H–^{13}C long range COSY, etc.) as 2-hydroxy-3,4,6,7-tetramethoxy-9,10-dihydrophenanthrene-2-*O*-β-D-glucopyranoside. The known compounds were identified as icariin, epimedoside C, icarisid I, baohuoside I, diphylloside A, baohuoside II, 2″-*O*-rhamnosylicarisid II, sagittatoside A, hyperoside and icaritin-3-*O*-rhamnopyranoside.

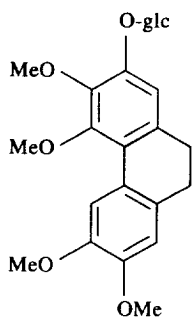
INTRODUCTION

In our previous paper, the isolation and structural determination of some new flavonoids in *Epimedium koreanum* were described [1–3]. In continuation of our study on the chemotaxonomy of the genus *Epimedium* (Berberidaceae), we isolated a new 9,10-dihydrophenanthrene derivate together with ten known compounds from the aerial parts of *E. koreanum*. We now deal with the structural elucidation of the new compound (**1**).

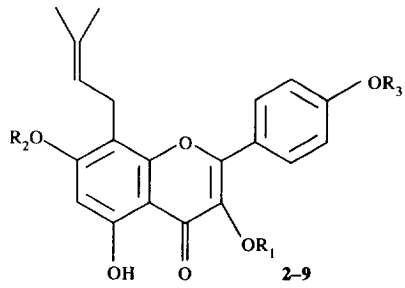
RESULTS AND DISCUSSION

Compounds **1**–**11** were isolated after repeated polyamide and silica gel chromatography of the EtOAc-soluble portion of a 95% EtOH extract and final purification by Sephadex LH-20 chromatography or recrystallization.

Epimedoicarisoside A (**1**), mp 205–206°, was obtained as an amorphous powder. The UV spectrum showed a maxima at 280, 304 and 316 nm. The ^1H NMR spectrum exhibited a broad singlet due to benzylic methylene protons at δ2.60 (4H), four singlets due to methoxyl protons at δ3.83, 3.78, 3.77 and 3.71 (each 3H), a doublet due to an anomeric proton at δ4.92 (1H, J = 7.5 Hz) and three singlets due to aromatic protons at δ7.85, 6.90 and 6.88 (each 1H). From these data, **1** was assumed to be a 9,10-dihydrophenanthrene derivate having four methoxyl groups and a glycosyl residue [4]. The FAB mass spectrum of **1** gave $[M + H]^+$ at m/z 479 and $[aglycone + H]^+$ at m/z 317. The sugar was identified as glucose by co-TLC with authentic specimen after acid hydrolysis [5]. The ^{13}C NMR spectrum of **1** exhibited four methoxyl carbon signals at δ55.6, 55.5, 60.4 and 60.9, the latter two signals might be due to ortho-disubstituted methoxyl groups because of the downfield shifts [6]. In the ^1H–^{13}C long range coupling spectrum obtained by HMBC (heteronuclear multiple bond correlation), the methylene protons at δ2.60 showed cross peaks with the carbons at δ130.5 (C-8_a), 133.5 (C-10_a), 111.6 (C-1), 111.6 (C-8), 124.3 (C-4_b) and 121.1 (C-4_a) (shown in Fig. 2); the proton at δ6.90 (H-1) showed cross peaks with the carbons at δ149.5 (C-2), 141.7 (C-3), 121.1 (C-4_a) and 29.9 (C-10); the proton at δ7.85 (H-5) showed cross peaks with the carbons at δ147.4 (C-7), 146.9 (C-6), 124.3 (C-4_b),

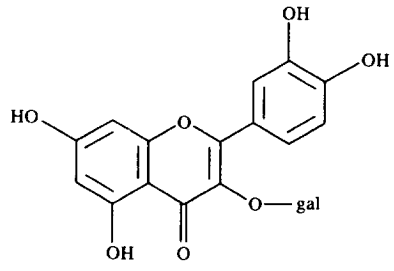


1

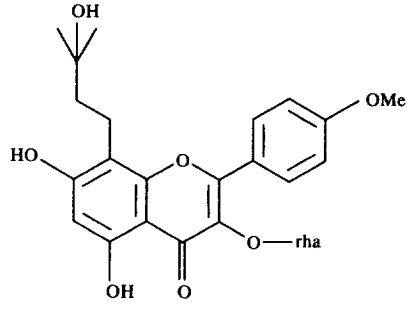


2–9

	R_1	R_2	R_3
2	rha	glc	Me
3	H	glc	H
4	H	glc	Me
5	rha	H	Me
6	rha²glc	glc	H
7	rha	H	H
8	rha²rha	H	Me
9	rha²glc	H	Me



10



11

Fig. 1. Structure of 1–11.

121.1 (C-4_a) and 130.5 (C-8_a); the proton at δ6.88 (H-8) showed cross peaks with the carbons at δ28.6 (C-9), 124.3 (C-4_b) and 146.9 (C-6); the anomeric proton (δ4.92) showed cross peak with the carbon at δ149.5 (C-2) indicating that the glucose was attached by a β-glycosidic linkage to the phenoxyl group at C-2. On the other hand, the methoxyl protons at δ3.83, 3.78, 3.77 and 3.71, which gave cross peaks with the carbons at δ60.9, 55.5, 55.6 and 60.4, respectively in the ^1H–^{13}C coupling spectrum, showed cross peaks with the carbons at δ141.7 (C-3), 147.4 (C-7), 146.9 (C-6) and 150.8 (C-4), respectively in the ^1H–^{13}C long range coupling spectrum. From the spectral data described, the structure of **1** was elucidated to be 2-hydroxy-3,4,6,7-tetramethoxy-9,10-dihydrophenanthrene-2-*O*-β-D-glucopyranoside and named as epimedoicarisoside A.

Compound **2** (mp 223–225°), **3** (mp 290–294°), **4** (mp 252–254°), **5** (mp 207–208°), **6** (mp 203–205°), **7** (mp 154–156°), **8** (mp 158–160°), **9** (mp 170–171°), **10** (mp 233–235°) and **11** (mp 239–241°) were obtained as yellow amorphous powders or needles. The UV, IR, mass, ^1H and ^{13}C NMR spectral data suggested that they were the known compounds icariin, epimedoside C [7], icarisid I [8], baohuoside I [9], diphylloside A [10], baohuoside II [7], 2″-*O*-rhamnosylicarisid II [11], sagittatoside A [12], hyperoside [7] and icaritin-3-*O*-rhamnopyranoside [13].

EXPERIMENTAL

All mps are uncorr. Ms were obtained with ZAB-HS and AEI MS-50. ^1H NMR and ^{13}C NMR were recorded with Bruker ARX-400 or Varian VXR-300 in DMSO-d_6; chemical shifts are given in ppm value with TMS as int. standard.

Extraction and isolation of compounds. Fr. 3 of the EtOAc-soluble portion described in detail in a previous paper [1] was chromatographed on a silica gel column with $CHCl_3$–MeOH (in gradient) as the eluent to give 11 subfrs. Subfr. 9 was rechromatographed on a silica gel column with $CHCl_3$–MeOH (20:1) as the eluent to yield subfraction A-O. Subfraction G was subjected to a Sephadex LH-20 column eluted with MeOH to give **1** (50 mg). Compound **2** (1100 mg) from fr. 9; **3** (400 mg) from fr. 11; **4** (370 mg) from fr. 4; **5** (1800 mg) from fr. 6; **6** (100 mg) from fr. 13; **7** (35 mg), **8** (21 mg), **9** (18 mg) and **11** (32 mg) from fr. 7 and **10** (4000 mg) from fr. 15 were obtained with silica gel and polyamide chromatography

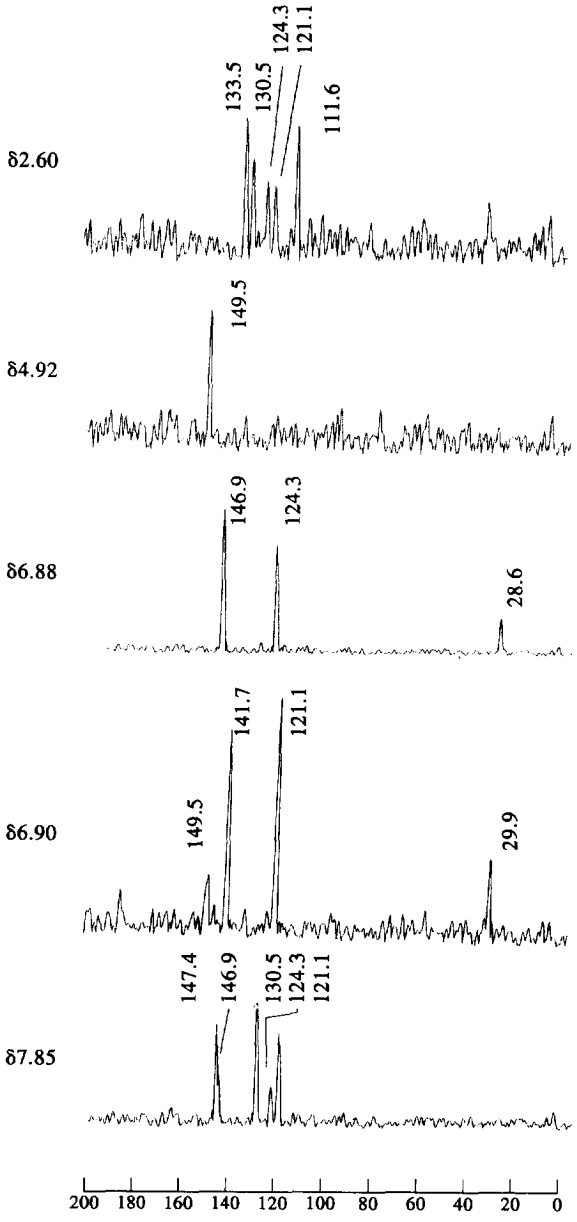


Fig. 2. Slices through protons in $^{1}H-^{13}C$ long range COSY of **1**.

and final purification with Sephadex LH-20 chromatography or recrystallization.

Compound **1** (*epimedoicarisoside A*). An amorphous powder (MeOH). Mp 205–206°. $C_{24}H_{30}O_{10}$. UV λ_{max}^{MeOH} nm: 280, 304, 316. IR ν_{max}^{KBr} cm^{-1}: 3500, 3400, 1610, 1600, 1520, 1460, 1400. ^{13}C NMR (DMSO-d_6, 100 MHz) δ: 111.6 (C-1), 149.5 (C-2), 141.7 (C-3), 150.8 (C-4), 111.2 (C-5), 146.9 (C-6), 147.4 (C-7), 111.6 (C-8), 28.6 (C-9), 29.9 (C-10), 121.1 ($C-4_a$), 124.3 ($C-4_b$), 130.5 ($C-8_a$), 133.5 ($C-10_a$), 60.9 (OMe-3), 60.4 (OMe-4), 55.6 (OMe-6), 55.5 (OMe-7), 100.8 (G-1), 73.5 (G-2), 76.9 (G-3), 69.9 (G-4), 77.2 (G-5), 60.8 (G-6).

Compounds **2**–**11**. Properties and spectral data identical to those reported data.

REFERENCES

1. Li, W. K., Xiao, P. G., Tu, G. Z., Ma, L. B. and Zhang, R. Y. (1995) *Phytochemistry* **38**, 263.
2. Li, W. K., Xiao, P. G. and Zhang, R. Y. (1994) *Phytochemistry* **38**, 807.
3. Li, W. K., Xiao, P. G., Pan, J. Q., Lü, M. J. and Zhang, R. Y. (1994) *Chinese Chemical Letters* **5**, 1033.
4. Letcher, R. M. and Nhamo, L. R. (1971) *J. Chem. Soc.* (*C*) 3070.
5. Zhao, P. P., Li, B. M. and He, L. Y. (1987) *Acta Pharm. Sin.* **22**, 70.
6. Levy, G. C. (1976) *Topics in Carbon-13 NMR Spectroscopy*, Vol. 2, p. 108, John Wiley and Sons. Inc., New York.
7. Li, F. and Liu, Y. R. (1988) *Acta Pharm. Sin.* **23**, 672.
8. Li, Y. S. and Liu, Y. R. (1992) *Chinese Trad. Herbs and Drugs* **23**, 8.
9. Li, F. and Liu, Y. R. (1988) *Acta Pharm. Sin.* **23**, 739.
10. Fukai, T. and Nomura, T. (1988) *Phytochemistry* **27**, 259.
11. Kang, S. S., Kang, Y. J. and Lee, M. W. (1991) *J. Nat. Prod.* **54**, 542.
12. Mizuno, M., Sakakibara, N., Hanioka, S., Iinuma, M., Tanaka, T., Liu, X. S. and Shi, D. W. (1988) *Phytochemistry* **27**, 3641.
13. Mizuno, M., Hanioka, S., Suzuki, N., Iinuma, M., Tanaka, T., Liu, X. S. and Min, Z. D. (1987) *Phytochemistry* **26**, 861.