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# FLAVONOIDS FROM *EPIMEDIUM KOREANUM*

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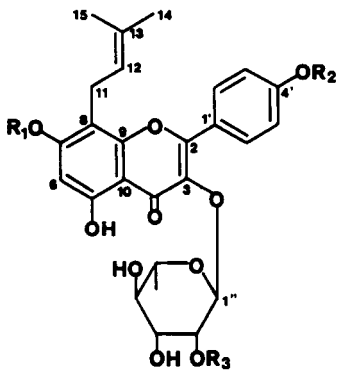
ABSTRACT.—Two new flavonol glycosides, together with epimedoside A [**2**], icariin [**3**], and ikarisoside A [**5**], have been isolated from the underground parts of *Epimedium koreanum* and characterized as 2″-*O*-rhamnosyl ikarisoside A [**1**] and 2″-*O*-rhamnosyl icarisid II [**4**] by chemical and spectral data.

*Epimedium koreanum* N. (Berberidaceae) and other related species belonging to the genus *Epimedium* are important in Chinese herbal medicine. The aerial parts of these plants have been used as a tonic to stimulate hormone secretion to cure impotence and forgetfulness, whereas the underground parts are used for treating asthmatic attacks and menstrual irregularity (1). In our previous papers, the isolation of *n*-alkanes, sterols, sterol glycosides, and flavonoids was reported from *E. koreanum* (2,3). From the underground parts of *E. koreanum*, we have isolated two new prenylated flavonol glycosides **1** and **4** along with epimedoside A [**2**], icariin [**3**], and ikarisoside A [**5**]. This paper describes the isolation and structural characterization of these compounds.

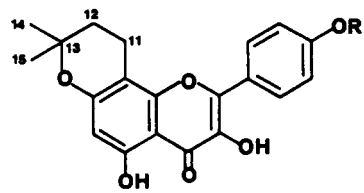
## RESULTS AND DISCUSSION

Repeated cc of the EtOAc-soluble portion of the MeOH extract resulted in the isolation of five compounds, which gave positive Molisch and Shinoda tests.

Compound **1**, mp 177–181°, showed ir absorption bands at 3380 (OH), 1654 (conjugated C=O) and 1070–1030 (glycosidic C-O) $cm^{-1}$ and uv maxima at 271, 318, and 350 nm. Bathochromic shifts on addition of some shift reagents indicated the presence of free hydroxyl groups at C-5, -7, and -4′ of the flavonoid skeleton. In the $^1H$-



**1** $R_1=R_2=H$, $R_3=Rha$
**2** $R_1=Glc$, $R_2=R_3=H$
**3** $R_1=Glc$, $R_2=Me$, $R_3=H$
**4** $R_1=H$, $R_2=Me$, $R_3=Rha$
**5** $R_1=R_2=R_3=H$



**6** R=H
**7** R=Me

Rha = α-L- rhamnopyranosyl
Glc = β-D-glucopyranosyl

nmr spectrum, two one-proton multiplets at δ 3.62 and 3.69, a one-proton broad triplet ($J$ = 6.6 Hz) at δ 5.15, and two methyl singlets at δ 1.62 and 1.68 demonstrated the presence of a prenyl group. Aromatic hydrogen signals were evident at δ 6.30 (1H, s), assignable to the A-ring proton, and a set of *ortho* coupled doublets at δ 6.92 (2H, d, $J$ = 8.7 Hz) and 7.75 (2H, d, $J$ = 8.7 Hz), assignable to the 4′-substituted B-ring protons. The prenyl group was placed at the C-8 position because band I of compound **1** underwent a significant bathochromic shift by 63 nm on addition of $AlCl_3$ (4) and acid hydrolysis of compound **1** gave des-*O*-methyl-β-anhydroicaritin [**6**] (5), mp >300°, as the aglycone and L-rhamnose as the sugar moiety. The $^1$H-nmr spectrum of **1** showed the presence of two moles of L-rhamnose, which was linked at the C-3 hydroxyl group. These findings indicated that compound **1** was des-*O*-methylanhydroicaritin 3-*O*-rhamnobioside. The sequence of the rhamnobiose moiety was determined as follows: the chemical shift of the terminal rhamnose anomeric proton was significantly shifted upfield at δ 4.89, revealing 1→2 linkage (6), which was further supported by the fact that the $^{13}$C-nmr chemical shifts of the anomeric and C-2″ signals were displaced upfield by 1.2 ppm and downfield by 5.1 ppm, respectively, from those of ikarisoside A [**5**]. From the above findings, the structure of **1** was determined to be 3,5,7,4′-tetrahydroxy-8-prenyl flavone 3-*O*-α-L-rhamnopyranosyl (1→2)-α-L-rhamnopyranoside (2″-*O*-rhamnosyl ikarisoside A).

Compound **4**, mp 154–157°, showed ir and uv spectral data closely similar to those of **1**. On acid hydrolysis, **4** yielded the same sugar as in **1**, L-rhamnose, and an aglycone, β-anhydroicaritin [**7**] (7), mp 218°. The $^1$H- and $^{13}$C-nmr spectra of **4** are coincident to those of **1** except for some signals due to the B-ring moiety (Tables 1 and 2). Based on these findings, compound **4** was assumed to be a 4′-*O*-methylated compound **1**. This assumption has been verified by the following synthesis. Permethylation of **1** and **4** prepared separately by the method of Brimacombe *et al.* (8) afforded the same reaction product, octa-*O*-methylether. Methanolysis of this reaction product furnished methyl 2,3,4-tri-*O*-methylrhamnopyranoside and methyl 3,4-di-*O*-methylrhamnopyranoside. In the light of the above observations, the structure of compound **4** was assigned to be 3,5,7-trihydroxy-4′-methoxy-8-prenyl flavone 3-*O*-α-L-rhamnopyranosyl(1→2)-α-L-rhamnopyranoside (2″-*O*-rhamnosyl icarisid II).

TABLE 1. $^1$H-nmr Spectral Data for **1** and **4** in DMSO-$d_6$.[a]

| Proton | Compound | |
|---|---|---|
| | **1** | **4** |
| H-6 | 6.30 s | 6.30 s |
| H-2′, 6′ | 7.75 d (8.7) | 7.85 d (8.8) |
| H-3′, 5′ | 6.92 d (8.7) | 7.11 d (8.8) |
| H-11 | 3.62 m | 3.61 m |
| | 3.69 m | 3.68 m |
| H-12 | 5.15 br t (6.6) | 5.15 br t (6.6) |
| Me-14, 15 | 1.62 s | 1.62 s |
| | 1.68 s | 1.68 s |
| 5-OH | 12.55 | 12.56 |
| OMe | — | 3.85 s |
| anomeric H | 4.89 s | 4.89 s |
| | 5.38 s | 5.38 s |
| rhamnose Me | 0.83 d (5.0) | 0.81 d (5.3) |
| | 1.10 d (6.1) | 1.13 d (6.2) |

[a]Measured with a Bruker AM-300 (300 MHz) spectrometer. Data are δ (ppm), multiplicity, and $J$ (in parentheses) in Hz.

TABLE 2. $^{13}$C-nmr Spectral Data for **1** and **4** in DMSO-$d_6$.[a]

| Carbon | Compound | |
|---|---|---|
| | **1** | **4** |
| C-2 | 156.8 | 156.4 |
| C-3 | 133.9 | 134.3 |
| C-4 | 177.7 | 177.7 |
| C-5 | 162.3 | 162.3 |
| C-6 | 98.5 | 98.4 |
| C-7 | 160.6 | 161.2 |
| C-8 | 105.9 | 105.9 |
| C-9 | 153.7 | 153.7 |
| C-10 | 103.8 | 103.8 |
| C-11 | 21.2 | 21.1 |
| C-12 | 122.4 | 122.3 |
| C-13 | 130.7 | 130.8 |
| C-14 | 25.3 | 25.3 |
| C-15 | 17.4 | 17.4 |
| C-1′ | 120.6 | 122.3 |
| C-2′ | 130.4 | 130.3 |
| C-3′ | 115.3 | 114.0 |
| C-4′ | 158.8 | 158.8 |
| C-5′ | 115.3 | 114.0 |
| C-6′ | 130.4 | 130.3 |
| C-1″ | 100.6 | 100.6 |
| C-2″ | 75.5 | 75.5 |
| C-3″ | 70.5 | 70.5 |
| C-4″ | 71.9 | 71.9 |
| C-5″ | 70.1 | 70.1 |
| C-6″ | 17.7 | 17.7 |
| C-1‴ | 101.5 | 101.5 |
| C-2‴ | 70.2 | 70.2 |
| C-3‴ | 70.5 | 70.5 |
| C-4‴ | 71.3 | 71.3 |
| C-5‴ | 68.7 | 68.7 |
| C-6‴ | 17.4 | 17.4 |
| OMe | | 55.4 |

[a]Measured with a Bruker AM-300 (75.5 MHz) spectrometer.

Compounds **2** (epimedoside A), mp 228–230°, **3** (icariin), mp 238°, and **5** (ikarisoside A), mp 149–159°, were identified by comparison of spectral data with reported values and $^{13}$C-nmr spectral data (9–13).

## EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mp's were determined on a Mitamura-Riken apparatus and are uncorrected. Optical rotations were measured on a Rudolph Autopol III automatic polarimeter. Ir spectra were recorded on a Perkin-Elmer 283B spectrophotometer. Elemental analysis was performed on a Perkin-Elmer 240DS instrument. $^1$H-nmr spectra were obtained on a Varian FT-80A (80 MHz) or a Bruker AM-300 (300 MHz) spectrometer using TMS as an internal standard. $^{13}$C-nmr spectra were recorded with a Bruker AM-300 (75.5 MHz) or a Varian FT-80A (20 MHz) instrument. Eims were determined on a Hewlett-Packard 5985B gc/ms system equipped with direct inlet system. Uv spectra were obtained on a Gilford System 2600 spectrophotometer. For tlc, Kieselgel 60 $F_{254}$ sheets (Merck) were used.

PLANT MATERIAL.—Underground parts of *E. koreanum* were collected in Kang Won province of Korea in the summer of 1988 and authenticated by Prof. Chang M. Kim (College of Pharmacy, Kang Won National University). A voucher specimen is deposited at the herbarium of our Institute.

EXTRACTION AND ISOLATION.—Dried plant material (500 g) was refluxed with MeOH for 3 h (4 times). Concentration of the extract under reduced pressure gave a residue which was suspended in distilled $H_2O$ and partitioned with $CHCl_3$, EtOAc, and then *n*-BuOH to yield $CHCl_3$ (2.5 g), EtOAc (5.2 g), and *n*-BuOH (6.2 g) fractions, respectively. The EtOAc fraction was subjected to flash cc over Si gel and eluted with EtOAc, EtOAc-EtOAc saturated with $H_2O$ (4:1, 2:1, 1:1) and EtOAc saturated with $H_2O$ to give 34 fractions. Fraction 4 was rechromatographed over Si gel with $CHCl_3$-MeOH (7:1, 6:1, 5:1), yielding compound **5** (45 mg). Repeated cc of fractions 9 and 10 afforded subfractions A–F. Subfraction E was subjected to Sephadex LH-20 cc eluted with MeOH, followed by cc over Si gel eluted with $CHCl_3$-MeOH-$H_2O$ (26:14:5, lower phase) to yield compounds **4** (25 mg), **2** (360 mg), and **1** (30 mg). Rechromatography of subfraction F over Si gel eluted with $CHCl_3$-MeOH-$H_2O$ (7:3:1) gave compound **3** (120 mg). The physical and spectral data of epimedoside A [**2**], icariin [**3**], and ikarisoside A [**5**] were identical with those previously published (9–13).

*Compound* **1**.—Crystallized from MeOH as yellowish needles: mp 177–181°, $[\alpha]^{15}D$ −69.0° ($c$ = 0.51, MeOH); ir $\nu$ max (KBr) 3380 (OH), 1654 (α,β-unsaturated C=O), 1613, 1508 (C=C), 1070–1030 $cm^{-1}$ (C-O); uv λ max (MeOH) (log ε) 271 (4.79), 318 (4.51), 350 (4.46); (NaOMe) 280 (4.86), 330 (4.54), 395 (4.69); (NaOAc) 277 (4.80), 372 (4.48); (NaOAc + $H_3BO_3$) 271 (4.79), 318 (4.51), 352 (4.47); ($AlCl_3$) 281 (4.74), 310 (4.47), 351 (4.56), 413 (4.38); ($AlCl_3$ + HCl) 281 (4.66), 308 (4.44), 348 (4.51), 397 (4.25); $^1H$ nmr (300 MHz, DMSO-$d_6$) see Table 1; $^{13}C$ nmr (75.5 MHz, DMSO-$d_6$) see Table 2. *Anal.* calcd for $C_{32}H_{38}O_{14}\cdot H_2O$, C 57.83, H 6.02; found C 57.65, H 6.26.

*Compound* **4**.—Crystallized from MeOH as yellowish plates: mp 154–157°, $[\alpha]^{15}D$ −72.4° ($c$ = 0.19, MeOH); ir $\nu$ max (KBr) 3400 (OH), 1654 (α,β-unsaturated C=O), 1612, 1508 (C=C), 1075–1020 (C-O) $cm^{-1}$; uv λ max (MeOH) (log ε) 272 (4.72), 299 (4.45), 350 (4.33); (NaOMe) 282 (4.88), 381 (4.37); (NaOAc) 280 (4.76), 308 (sh, 4.42), 357 (4.29); (NaOAc + $H_3BO_3$) 272 (4.72), 298 (4.46), 348 (4.34); ($AlCl_3$) 281 (4.65), 310 (4.43), 349 (4.47), 413 (4.25); ($AlCl_3$ + HCl) 282 (4.64), 309 (4.45), 345 (4.47), 412 (4.19); $^1H$ nmr (300 MHz, DMSO-$d_6$) see Table 1; $^{13}C$ nmr (75.5 MHz, DMSO-$d_6$) see Table 2. *Anal.* calcd for $C_{33}H_{40}O_{14}\cdot H_2O$, C 58.41, H 6.19; found C 58.42, H 6.27.

ACID HYDROLYSIS OF COMPOUNDS **1–5**.—Acid hydrolysis of compounds **1** (5 mg), **2** (40 mg), **3** (40 mg), **4** (5 mg), and **5** (5 mg) was performed by refluxing each reaction mixture with 25% $H_2SO_4$/dioxane (25 ml) for 5 h. Each reaction mixture was poured onto iced $H_2O$ and filtered. Each filtrate was neutralized with $BaCO_3$ and filtered, and the solution was evaporated to dryness under reduced pressure. L-Rhamnose from **1**, **4**, and **5** and D-glucose and L-rhamnose from **2** and **3** were detected by tlc using a precoated cellulose plate developed in pyridine-EtOAc-HOAc-$H_2O$ (36:36:7:21). The precipitate obtained from **1**, **2**, and **5** afforded the same aglycone, combined and chromatographed on Si gel with $CHCl_3$-MeOH (25:1) to yield **6** which was crystallized from MeOH as a yellowish powder: mp >300° [lit. (5) mp 302–305°]; ir $\nu$ max (KBr) 3300 (OH), 1652 (α,β-unsaturated C=O), 1610, 1505, (C=C), 1368, 1155 $cm^{-1}$; uv λ max (MeOH) (log ε) 259 (sh, 4.13), 273 (4.26), 309 (sh, 3.92), 332 (sh, 3.98), 370 (4.15); (NaOMe) 275 (4.31), 428 (4.32); (NaOAc) 273 (4.25), 336 (sh, 3.85), 383 (4.14); (NaOAc + $H_3BO_3$) 257 (4.15), 274 (4.26), 308 (3.93), 332 (4.00), 372 (4.20); ($AlCl_3$) 277 (4.31), 314 (3.78), 362 (3.92), 428 (4.26); ($AlCl_3$ + HCl) 277 (4.27), 314 (3.81), 361 (3.92), 428 (4.22); $^1H$ nmr (80 MHz, DMSO-$d_6$) 1.32 (6H, s, Me-14, 15), 1.85 (2H, t, $J$ = 6.1 Hz, H-12), 2.84 (2H, t, $J$ = 6.2 Hz, H-11), 6.12 (1H, s, H-6), 6.92 (2H, d, $J$ = 8.8 Hz, H-3′, -5′), 8.07 (2H, d, $J$ = 8.8 Hz, H-2′, -6′), 12.21 (1H, br s, 5-OH); ms *m/z* (rel. int.) $[M]^+$ 354 (69.2), $[M-Me]^+$ 339 (11.5), $[M-CO]^+$ 311 (0.8), $[M-$ isobutenyl$]^+$ 299 (100), $[A_1+H]^+$ 221 (0.2), $[M/2]^+$ 177 (11.9), $[(A_1+H)-2CO]^+$ 165 (24.1), $[B_1]^+$ 134 (6.8), $[B_2]^+$ 121 (48.7), $[B_2-CO]^+$ 93 (13.9).

The precipitates from **3** and **4** yielded the same aglycone and were combined, followed by chromatography over Si gel with $CHCl_3$ as eluent, to yield **7** as yellowish needles from MeOH: mp 218° [lit. (7) mp 221–223°]; ir $\nu$ max (KBr) 3300 (OH), 1657 (α,β-unsaturated C=O), 1600, 1510 (C=C), 1365, 1165 $cm^{-1}$; uv λ max (MeOH) (log ε) 262 (sh, 4.65), 274 (4.77), 310 (4.44), 330 (4.46), 371 (4.59); (NaOMe) 264 (4.82), 276 (sh, 4.81), 340 (4.41), 418 (4.68); (NaOAc) 274 (4.77), 328 (4.34), 385 (4.57), 408 (sh, 4.51); (NaOAc + $H_3BO_3$) 256 (sh, 4.72), 274 (3.83), 313 (sh, 4.58), 330 (4.61), 371 (4.73); ($AlCl_3$) 277 (4.81), 314 (4.32), 359 (4.45), 428 (4.74); ($AlCl_3$ + HCl) 277 (4.70), 314 (4.31), 363 (4.43), 428 (4.63); $^1H$ nmr (80 MHz, DMSO-$d_6$) 1.33 (6H, s, Me-14, -15), 1.86 (2H, t, $J$ = 6.6 Hz, H-12), 2.85 (2H, t, $J$ = 6.5 Hz, H-11), 3.84 (3H, s, OMe), 6.13 (1H, s, H-6), 7.12 (2H, d, $J$ = 9.0 Hz, H-3′, -5′), 8.17 (2H, d, $J$ = 9.0 Hz, H-2′, -6′), 12.25 (1H, br s, 5-OH); ms *m/z* (rel. int.) $[M]^+$ 368 (49.6), $[M-Me]^+$ 353 (9.5), $[M-(Me+CO)]^+$ 325 (1.0), $[M-$ isobutenyl$]^+$ 313 (100), $[A_1+H]^+$ 221 (0.3), $[M/2]^+$ 184 (2.5), $[(A_1+H)-2CO]^+$ 165 (20.1), $[B_1]^+$ 148 (6.0), $[B_2]^+$ 135 (37.1), $[B_2-CO]^+$ 107 (8.0).

PERMETHYLATION OF **1** AND **4**.—NaH powder (20 mg) was added to **1** in 2 ml of DMF, and the mixture was stirred for 20 min. To the reaction mixture was then added 0.5 ml of MeI, and the mixture was

allowed to stand at room temperature for 3 h. Distilled $H_2O$ was added and the mixture extracted twice with $CHCl_3$. The extract was dried and evaporated to give 6 mg of the crude product, which was purified by Si gel tlc [$CHCl_3$-MeOH (50:1)] to give the pure permethylated product: $^1H$ nmr (80 MHz, $CDCl_3$) 0.98 (3H, d, $J$ = 5.9 Hz, Rha-Me), 1.28 (3H, d, $J$ = 5.4 Hz, Rha-Me), 1.66, 1.70 (3H each, s, Me-14, 15), 3.45 (3H, s, OMe), 3.48 (6H, s, 2 × OMe), 3.49 (6H, s, 2 × OMe), 3.86 (3H, s, OMe), 3.93 (6H, s, 2 × OMe), 4.66 (1H, d, $J$ = 1.5 Hz, anomeric H), 5.16 (1H, d, $J$ = 1.6 Hz, anomeric H), 5.27 (1H, m, H-12), 6.38 (1H, s, H-6), 6.98 (2H, d, $J$ = 8.8 Hz, H-3′, -5′), 7.83 (2H, d, $J$ = 8.8 Hz, H-2′, -6′); ms (30 eV) *m/z* (rel. int.) $[M-(3MeOH+MeO)]^+$ 631 (1.4), 601 (2.9), 585 (3.4), $[M-Rha(OMe)_3]^+$ 569 (1.1), $[601-isobutenyl]^+$ 546 (2.5), 545 (8.7), 456 (6.0), 442 (17.7), 427 (10.8), 412 (44.6), 410 (26.4), $[aglycone(OMe)_3]^+$ 396 (46.3), $[396-isobutenyl]^+$ 341 (17.0), $[A_1+H]^+$ 249 (1.1), $[Rha(Ome)_3]^+$ 189 (100), $[189-MeOH]^+$ 157 (48.3), $[B_2]^+$ 135 (25.9), $[189-2MeOH]^+$ 125 (24.2), 101 (60.7).

The permethylether of **4** (3 mg) was prepared as described above and identified as octa-*O*-methylether by direct comparison with an authentic sample (tlc, co-tlc and ms).

METHANOLYSIS OF OCTA-*O*-METHYLETHER.—The octa-*O*-methyl ether was refluxed with 2% MeOH/HCl (10 ml) for 3 h. The reaction mixture was concentrated to half volume, added to iced $H_2O$, and extracted with $CHCl_3$. The $CHCl_3$ layer was dried, concentrated, and subjected to glc [column 10% DEGS on chromosorb WHP 100-120 mesh, 2.3 mm × 6 ft, column temperature 170°; flow rate ($N_2$) 45.9 ml/min; chart speed 1 cm/min]. Methyl 2,3,4-tri-*O*-methylrhamnopyranoside (Rt 2.78) and methyl 3,4-di-*O*-methylrhamnopyranoside (Rt 6.41) were identified by comparison with authentic samples (14).

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## LITERATURE CITED

1. L.M. Perry, "Medicinal Plants of East and Southeast Asia: Attributed Properties and Uses," MIT Press, Cambridge, 1980, p. 55.
2. S.S. Kang, K.H. Shin, S.G. Chung, and E.H. Cho, *Kor. J. Pharmacogn.*, **19**, 93 (1988).
3. S.S. Kang, J.S. Kim, Y.-J. Kang, and H.K. Han, *Kor. J. Pharmacogn.*, **21**, 56 (1990).
4. E.A. Sherif, R.K. Gupta, and M. Krishnamurti, *Tetrahedron Lett.*, **21**, 641 (1980).
5. Y. Tokuoka, K. Daigo, and T. Takemoto, *Yakugaku Zasshi*, **95**, 698 (1975).
6. G.G.S. Dutton, E.H. Merrifield, C. Laffite, F. Pratviel-Sosa, and R. Wylde, *Org. Magn. Reson.*, **20**, 154 (1982).
7. Y. Tokuoka, K. Daigo, and T. Takemoto, *Yakugaku Zasshi*, **95**, 825 (1975).
8. J.S. Brimacombe, B.D. Jones, M. Stacey, and J.J. Willard, *Carbohydr. Res.*, **2**, 167 (1966).
9. M. Mizuno, M. Iinuma, T. Tanaka, N. Sakakibara, T. Fujikawa, S. Hanioka, Y. Ishida, X.-S. Liu, and H. Murata, *Phytochemistry*, **27**, 3645 (1988).
10. M. Mizuno, S. Hanioka, N. Suzuki, M. Iinuma, T. Tanaka, X.-S. Liu, and Z.-D. Min, *Phytochemistry*, **26**, 861 (1987).
11. T. Fukai and T. Nomura, *Phytochemistry*, **27**, 259 (1988).
12. F. Li and Y.L. Liu, *Acta Pharm. Sin.*, **23**, 672 (1988).
13. F. Li and Y.L. Liu, *Acta Pharm. Sin.*, **23**, 739 (1988).
14. S.S. Kang, J.S. Kim, W.-J. Kwak, and K.-H. Kim, *Arch. Pharm. Res.*, **13**, 207 (1990).

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