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TWO STEROIDAL ALKALOIDS FROM *FRITILLARIA HARELINII*

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Key Word Index—*Fritillaria harelinii*; Liliaceae; steroidal alkaloids; peiminine; harepermine; hareperminside.

Abstract—Two new steroidal alkaloids named harepermine and hareperminside have been isolated from the bulbs of *Fritillaria harelinii* together with a known alkaloid, peiminine. The structure of the alkaloids were found to be 3β,6β-dihydroxy-5α,14α,17β-cevanine and its 3-*O*-glucoside on the basis of spectroscopic evidence.

INTRODUCTION

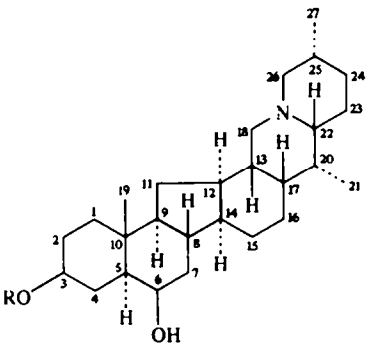
The bulbs of *Fritillaria harelinii* (Fish.) Regel ex Baker, which grows on sandbanks at Xinjiang in China [1], have been used as one of the traditional drugs for the treatment of coughs. This communication deals with new alkaloids to add to the interesting group of C-nor-D-homosteroidal alkaloids in *Fritillaria* species [2–5, 7].

RESULTS AND DISCUSSION

Compound **1**, mp 213–214°, $[\alpha]_D$ −18.1° (*c* 2.0; $CHCl_3$), $C_{27}H_{43}NO_3$ was obtained as colourless crystals. The IR spectrum suggest that it contained hydroxyl groups (3450 cm^{-1}), a carbonyl group (1700 cm^{-1}) and a *trans*-quinolizidine moiety (2750 cm^{-1}). The mass spectrum showed fragments at *m/z* 429$[M]^+$ and 112 (base peak). In the ^1H NMR spectrum, signals at δ0.75 (3H, *s*), 1.03 (3H, *s*), 1.11 (3H, *d*, *J* = 6 Hz) and 3.56 (1H, *m*, $W_{1/2}$ = 24 Hz) are assignable to the methyl protons at C-19, C-21, C-27 and a proton at C-3, respectively. Each methylene carbon in the ^{13}C NMR spectrum is clearly assignable by the INEPT/R method (see Table 1). From the above spectral data, **1** was identified as peiminine [6]. The structure was further confirmed by comparison with an authentic sample.

Compound **2**, mp 157–160°, $[\alpha]_D$ −18.7° (*c* 0.8; $CHCl_3$), $C_{27}H_{45}NO_2$ was obtained as colourless crystals. The presence of hydroxy groups and a *trans*-quinolizidine moiety were supported by the IR spectrum (3400 and 2780 cm^{-1}). The mass spectrum showed fragments at *m/z* 415$[M]^+$, 397$[M-18]^+$, 344$[M-71]^+$, 112, 111 (base peak) and 98. These spectral data indicate that **2** has a C-nor-D-homo-steroidal skeleton and that the hydroxyl group is not located at C-21. The ^1H NMR spectrum shows signals at 1.02 (Me-19), 3.67 ($W_{1/2}$ = 25 Hz, H-3) and 3.86 (*J* = 8 Hz, H-6), respectively. These data are very similar to those obtained for isoverticine [7]. In the ^{13}C NMR spectrum, the carbons at δ71.3 (C-3) and 72.5 (C-6) show a downfield shift in comparison with those found in isoverticine. Therefore, the two β-hydroxyl groups should be located at C-3 and C-6, respectively. The signal at C-19 (δ1.02) in the ^1H NMR spectrum suggest that **2** possess *trans*-junctures between A/B, C/D, E/F and a *cis*-juncture between D/E [8]. In comparison with the ^{13}C NMR spectrum of shinonomenine and isoverticine [9], the ^{13}C-signals of C-16, C-17, C-18 and C-22 of **2** are shifted to highfield. The signals of delavine [10] isolated from *F. delavayi* are in agreement with those of **2**. Therefore **2** has a *cis*-juncture between E/F and the H-17 of **2** has the β-configuration. The structure of **2** can therefore be assigned as 3β,6β-dihydroxy-5α,14α,17β-cevanine.

Compound **3**, mp 168–170°, $[\alpha]_D$ −27° (*c* 0.5; MeOH), $C_{33}H_{55}NO_7$ was obtained as colourless crystals, which gave a positive Molish test. The IR spectrum showed hydroxyl absorption and a *trans*-quinolizidine moiety at 3425 and 2750 cm^{-1}. In the mass spectrum, fragments at 577 $[M]^+$, 415 (aglycone) and 111 (base peak) were observed. The above spectral data show that **3** is a C-nor-D-homosteroidal alkaloid glycoside. The chemical shifts of the carbon in the ^{13}C NMR spectrum are similar to **2** with the signal at C-3 (δ78.7) shifted downfield by 7.4 ppm. The glycosylation shift shows that the sugar moiety is attached to C-3. The sugar moiety was determined as β-glucopyranose by ^{13}C NMR (C-1′, δ102.3; $J_{^1H-^{13}C}$ = 142 Hz; measured by INEPT/NON). Consequently, **3** is the 3-*O*-β-D-glucopyranoside of **2**, 6β-hydroxy-5α,14α,17β-cevanine-3β-*O*-glucopyranoside.



2 R = H
3 R = β-D-glucoside

Table 1. ^{13}C NMR spectral data of 1, 2 and 3 (pyridine-d_5)

Carbon no.	1	2	3
1	37.1	37.1	39.6
2	32.6	32.6	32.2
3	71.3	71.3	78.7
4	31.5	34.0	33.9
5	56.5	49.1	48.7
6	210.5	72.5	72.5
7	46.2	40.1	40.3
8	42.0	39.3	39.4
9	56.8	58.7	58.7
10	38.3	36.5	36.7
11	29.4	31.4	30.1
12	41.4	40.4	40.1
13	39.4	40.2	41.1
14	44.4	42.0	41.8
15	24.9	30.6	28.9
16	21.0	18.2	18.1
17	49.2	42.0	42.1
18	62.7	59.2	59.0
19	12.4	15.3	15.9
20	70.1	40.3	40.0
21	21.8	16.1	15.0
22	70.3	61.8	62.0
23	19.6	26.3	27.1
24	30.0	31.5	31.5
25	28.2	31.5	31.1
26	62.4	61.7	61.2
27	17.6	19.9	19.7
1′			102.3
2′			75.4
3′			78.2
4′			72.2
5′			78.6
6′			63.3

EXPERIMENTAL

Extraction and isolation. Dried bulbs (10 kg) were percolated with EtOH and the percolate concd to give a brown residue. The residue was dissolved in 3% HCl, the acidic soln filtered and adjusted to pH 10 with NH_4OH. The aq. soln was extracted with $CHCl_3$ and the extract chromatographed by rotary TLC on silica gel (satd with NH_3 gas for 5 min) with $CHCl_3$–MeOH (9:1) to give compounds **1** (30 mg) and **2** (40 mg). The EtOAc extract obtained was treated in the same manner as the $CHCl_3$ extract. The solvent system used for rotary TLC was $CHCl_3$–MeOH–H_2O (13:7:2, lower phase). From the EtOAc extract, compound **3** was obtained (120 mg).

Compound **1** (*peiminine*). Colourless powder, mp 213–214°, $[\alpha]_D$ −18.1° (*c* 0.2; EtOH); M_r 429, $C_{27}H_{43}NO_3$. UV λ_{max}^{EtOH} nm (log ε): 287 (1.88). IR ν_{max}^{KBr} cm^{-1}: 3450, 2950, 2750, 1705, 1463, 1054. ^1H NMR ($CDCl_3$, 90 MHz): δ0.75 (3H, *s*, Me-19), 1.03 (3H, *s*, 21-Me), 1.11 (3H, *d*, *J* = 7 Hz, 27-Me), 3.56 (1H, *m*, $W_{1/2}$ = 24 Hz, H-3α). MS (*m/z*): 429 $[M]^+$, 414, 411, 386, 384, 372, 358, 238, 234, 217, 180, 164, 156, 155, 154, 140, 125, 112 (base), 111, 98.

Compound **2** (*harepermine*). Colourless powder, mp 157–160° (EtOH), M_r 415, $C_{27}H_{45}NO_2$, $[\alpha]_D$ −18.7° (*c* 0.8; $CHCl_3$). IR ν_{max}^{KBr} cm^{-1}: 3400, 2950, 2780, 1460, 1380, 1130, 1058, 1035, 1010. ^1H NMR ($CDCl_3$, 90 MHz): δ0.81 (3H, *d*, *J* = 6.2 Hz, 21-Me), 0.82 (3H, *d*, *J* = 6 Hz, 27-Me), 1.02 (3H, *s*, 19-Me), 3.67 (1H, *m*, $W_{1/2}$ = 25 Hz, H-3α), 3.86 (1H, *br d*, *J* = 8 Hz, H-6α). MS *m/z* (rel. int.): 415 $[M]^+$ (31), 414 (25), 297 (5), 358 (13), 344 (2), 214 (25), 178 (3), 164 (8), 150 (4), 139 (5), 125 (5), 124 (6), 112 (53), 111 (100), 98 (53).

Compound **3** (*harepermside*). Colourless powder, mp 168–170°, $[\alpha]_D$ −27° (*c* 0.5; MeOH). M_r 577, $C_{33}H_{55}NO_7$. IR ν_{max}^{KBr} cm^{-1}: 3425, 2950, 2750, 1650, 1485, 1370, 1080, 1030, 750. ^1H NMR (C_6D_5N, 270 MHz): δ0.80 (3H each, *d*, *J* = 5.0 Hz, 21-Me, 27-Me), 1.08 (3H, *s*, 19-Me). MS *m/z* (rel. int.): 577 $[M]^+$ (47) 576 (19), 562 (7), 559 (3), 520 (8), 415 (6), 218 (2), 178 (1), 164 (5), 150 (2), 139 (4), 125 (5), 124 (2), 112 (50), 111 (100), 98 (24).

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REFERENCES

1. Institute of Botany, Academia Sinica (1976) *Iconographia Cormophytorum Sinicorum*, Tomus V, pp. 459–460.
2. Qing-Han Liu, Xiao-Guang Jia, Yong-Fang Ren Muhatal and Xeao-Tian Liaug (1984) *Acta Pharm. Sin.* **19**, 894.
3. Dong-Min Xu, Ben Ahang, Huan-Rong Li and Mao-Li Xu (1982) *Acta Pharm. Sinica*, **17**, 555.
4. Kaneko, K., Naruse, N., Tanaka, M., Yoshida, N. and Mitsuhashi, H., (1980) *Chem. Pharm. Bull.* **28**, 3711.
5. Kitajiam, J., Noda, N., Ida, Y., Miyahara, K., and Kawasaki, T. (1981) *Heterocycles* **15**, 781.
6. Jin-Hou Wu (1982) *Chinese Traditional and Herbal Drugs* **13**, 3.
7. Kaneko, K., Naruse, N., Haruki, K. and Mitsuhashi, H. (1980) *Chem. Pharm. Bull.* **28**, 1345.
8. Shakirov, R. and Yumsov, Y. (1980) *Chem. Nat. Compds* 6.
9. Kaneko, K., Tanak, M., Haruki, K., Naruse, N. and Mistuhashi, H. (1979) *Tetrahedron Letters* 3757.
10. Kaneko, K., Satsuhara, T., Ooka, T., Mitsuhashi, H., Hsu, H. and Chen, Y. (1985) Announcement at 105th Annual Meeting of The Pharmaceutical Society of Japan, Kanazawa.