DITERPENOID ALKALOIDS FROM DELPHINIUM PEREGRINUM. THE STRUCTURE OF PEREGRINE

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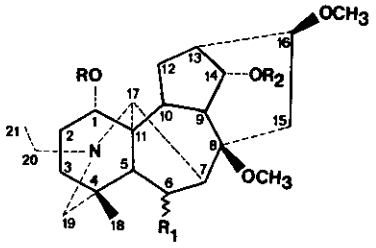
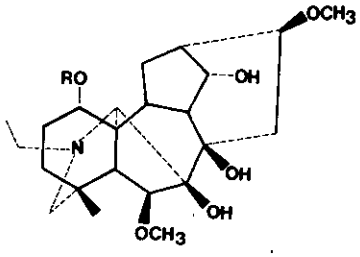
Abstract - From Delphinium peregrinum var. elongatum Boiss., bicoloridine, dihydrogadesine, nudicaulidine, 13-acetylhetisinone, and a new C-19 diterpenoid alkaloid peregrine were isolated. The structure of the new base was determined from chemical and spectral evidence.

Delphinium peregrinum var. elongatum Boiss. (Ranunculaceae) is an annual taxon found growing in sandy soils in coastal areas of Algeria and Morocco.[1] The dried ethanolic extract (673 g) of the aerial parts of plants collected in Medina Plage[2] (Rabat), during the flowering period, was treated with 5% sulphuric acid. After extraction with chloroform, the acid solution was adjusted to pH 9 with sodium carbonate, extracted with chloroform, and the solvent removed. The crude alkaloid mixture (4.78 g) was chromatographed over basic alumina (activity II-III) in mixtures of hexane-ethyl acetate and ethyl acetate-methanol to furnish the purified alkaloids.





1 R = CH_3; R_1 = αOAc; R_2 = H
2 R = CH_3; R_1 = αOAc; R_2 = Ac
3 R = CH_3; R_1 = αOH; R_2 = H
4 R = H; R_1 = βOAc; R_2 = H
5 R = H; R_1 = βOH; R_2 = H

8 R = H
9 R = CH_3

The new base peregrine (1) had mp 124-125°C, crystallized from ethyl acetate-hexane, $|\alpha|_D$+12° (c 0.34, EtOH). Its molecular formula was obtained by high resolution mass spectroscopy; M^+, m/z 463.2958 for $C_{26}H_{41}NO_6$, Δ -2.6 mmu. Ir (KBr), 3500 and 3450 (br) (OH), 1730 and 1250 (acetate), and 1090 cm^{-1} (C-O).

The ms (70 eV) presented the distinctive fragmentation of a C-19 diterpenoid alkaloid with a lycoctonine skeleton, providing ions at m/z, 463 (2%) M^+, 448 (2%) M^+-CH_3, 432 (100%) M^+-OCH_3, 430 (17%) $|M^+-CH_3|-H_2O$, 420 (6%) M^+-Ac, 404 (5%) M^+-OAc, 402 (5%) $|M^+-Ac|-H_2O$, 400 (11%) $|M^+-OCH_3|-CH_3OH$, 388 (13%) $|M^+-Ac|-CH_3OH$, and 372 (19%) $|M^+-OCH_3|$-AcOH.

The ^{1}H-nmr spectrum of peregrine (200 MHz, $CDCl_3$) also exhibited characteristic signals of a C-19 diterpenoid alkaloid[3,4] at δ0.82 (3H, s, CH_3), 1.04 (3H, t, J = 7.0 Hz, N-CH_2-$\underline{CH}_3$), 2.04 (3H, s, OAc), 3.07, 3.25 and 3.34 (3H each, s, three OCH_3), 3.63 (1H, d, J = 6.5 Hz, OH), and 3.97 (1H, q, J = 5.4 Hz). When D_2O was added, the quartet at δ3.97 became a triplet J = 4.8 Hz with the concomitant disappearance of the doublet at δ3.63. The coupling between these two signals was also established by double resonance experiments. Consequently, these resonances were assigned to C-14βH and C-14αOH, respectively. In this case, the strength of the intramolecular hydrogen bond between O-14H and O-16 or O-8, in axial configuration, restricts the free rotation of the hydroxyl group and inhibits the rapid proton exchange in $CDCl_3$, so the three-bond J_{HCOH} is observable.[5-7]

The ^{1}H-nmr spectrum moreover displayed an AX system at δ2.71 and 5.22 (J = 7.3 Hz). The one-proton doublet at δ2.71 was ascribed to C-5βH because of its one-bond correlation, observed in a SFSD experiment, with C-5 carbon resonance at δ42.4. As a result, the other proton doublet at δ5.22 was assigned to C-6βH (αOAc), considering the near 0° and 90° dihedral angles, measured on a Dreiding molecular model, for C-5βH and C-6βH, and C-6βH and C-7H, respectively. In bicoloridine (4), see below, the C-6αH (d, J = 7.4 Hz) is coupled with C-7H (0° dihedral angle), and not with C-5βH (90° dihedral angle).

Treated with Ac_2O-Py, peregrine provided the diacetate (2) as a resin, M^+ 505 (2%). Its ^{1}H-nmr spectrum presented signals at δ1.98 and 1.97 (3H each, s, two OAc), and a one-proton triplet at 4.69 (J = 4.8 Hz), indicating that the C-14αOH group was acetylated.

The alkaline hydrolysis of peregrine with 5% methanolic KOH afforded the amino-alcohol (3), mp 142-144°C, crystallized from ethyl acetate-hexane, M^+ 421 (3%); ^{1}H-nmr, δ0.95 (3H, s, CH_3), 1.05 (3H, t, J = 7.1 Hz, N-CH_2-$\underline{CH}_3$), 3.25, 3.31 and

^{13}C-nmr chemical shifts and assignments for peregrine (1), its amino-alcohol (3), 14-acetylperegrine (2), bicoloridine (4), its amino-alcohol (5), 6-acetylheteratisine (6), and heteratisine (7).

Carbon	1	2	3	4	5	6	7
1	84.7	85.6	84.2	72.6	72.9	82.2	83.5
2	26.5	26.5	27.1	29.7	30.3	26.7	26.9
3	33.0	33.1	35.8	31.6	31.8	36.4	36.8
4	34.5	34.6	34.2	32.8	32.6	34.7	34.7
5	42.4	45.9	42.0	42.3	43.5	49.7	50.9
6	73.4	73.0	73.1	72.3	72.5	74.0	72.9
7	56.4	58.9	56.5	52.8	54.8	48.6	49.3
8	79.1	80.9	78.5	79.9	81.8	75.3	75.4
9	46.2	46.3	46.0	44.4	45.5	57.4	57.8
10	38.6	37.7	39.1	40.0	39.9	42.8	42.8
11	48.2	48.3	48.5	48.9	48.9	49.7	49.3
12	28.6	28.5	28.6	30.6	29.6	35.7	33.1
13	44.6	43.8	41.2	44.4	44.4	75.8	75.8
14	75.5	75.2	76.3	75.9	75.8	173.8	176.0
15	37.1	37.5	37.2	37.8	37.4	28.9	29.1
16	82.5	82.4	83.6	83.3	83.0	29.4	29.2
17	64.7	64.3	64.0	65.5	65.1	62.4	62.2
18	25.9	26.0	26.1	27.3	27.5	25.9	26.2
19	57.6	58.1	57.5	61.6	62.2	54.8	58.3
20	49.3	49.6	48.5	48.3	48.6	48.7	49.0
21	13.6	13.8	13.6	12.9	13.1	13.4	13.5
1′	56.0	56.3	56.0	48.0	--	55.5	55.2
8′	48.3	48.6	48.0	--	48.5	--	--
16′	56.4	56.5	56.5	56.4	--	--	--
CO	170.2	--	171.5	170.9	--	170.8	--
CH_3	21.7	--	21.7	21.5	--	21.7	--
CO	--	--	171.5	--	--	--	--
CH_3	--	--	21.4	--	--	--	--

Chemical shifts in ppm downfield from TMS.

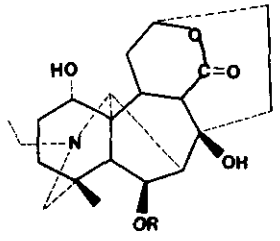
Solvent deuterochloroform.

3.37 (3H each, s, three OCH_3), 4.03 (1H, m, $W_{\frac{1}{2}}$ = 15 Hz; after D_2O, t, J = 4.8 Hz, C-14βH), and 4.30 (1H, d, J = 7.2 Hz, C-6βH).

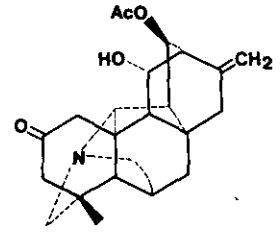
Bicoloridine (alkaloid A) (4)[8] was also isolated from this plant and identified by comparison with an authentic sample; ^{1}H-nmr, δ0.97 (3H, s, CH_3), 1.11 (3H, t, J = 7.2 Hz, N-CH_2-$C\underline{H}_3$), 2.03 (3H, s, OAc), 2.72 (1H, d, J = 7.4 Hz, C-7H), 3.10 and 3.38 (3H each, s, two OCH_3), 3.78 (1H, m, $W_{\frac{1}{2}}$ = 6 Hz, C-1βH), 4.12 (1H, m, $W_{\frac{1}{2}}$ = 13 Hz; after D_2O, t, J = 4.8 Hz, C-14 H), and 5.35 (1H, d, J = 7.4 Hz, C-6αH). By hydrolysis with 5% methanolic KOH bicoloridine (4) gave the amino-alcohol (5), the ^{1}H-nmr spectrum of which displayed signals at δ1.04 (3H, s, CH_3), 1.10 (3H, t, N-CH_2-$C\underline{H}_3$), 3.34 and 3.39 (3H each, s, two OCH_3), 3.74 (1H, m, $W_{\frac{1}{2}}$ = 7.3 Hz, C-1βH), 4.14 (1H, t, J = 4.5 Hz, C-14βH), and 4.39 (1H, d, J = 7.2 Hz, C-6αH).

The ^{1}H-nmr spectra of (4) and (5) are close to those of peregrine (1) and its amino-alcohol (3), respectively, revealing their structural similarities.

The BBD and DEPT ^{13}C-nmr spectra of peregrine (1) and its derivatives (2) and (3), compared with those of bicoloridine (4),[9] its amino-alcohol (5), heteratisine (7),[9] and 6-acetylheteratisine (6),[9] allowed us to make clear the structure of peregrine (1).



6 R = Ac
7 R = H



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The published assignments for C-7 and C-8 in bicoloridine (4) have been reversed, because its DEPT ^{13}C-nmr spectrum clearly showed a methine and a quaternary carbon resonance at 52.8 and 48.0 ppm, respectively, the latter in agreement with the expected value for a tertiary methoxyl group.[10,11]

We have also isolated dihydrogadesine (8),[12] nudicaulidine (9),[13] and 13-acetyl-hetisinone (10),[14] identified by comparison with authentic samples.

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