A new norditerpenoid alkaloid from *Delphinium densiflorum*

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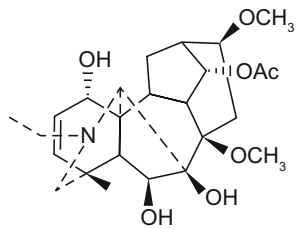
A new norditerpenoid alkaloid delphidenine was isolated from the EtOH extract of the whole plants of *Delphinium densiflorum*. Its structure was established by extensive application of spectroscopic methods, including IR, HR-ESI-MS, 1D and 2D NMR spectroscopy.

Keywords: *Delphinium densiflorum*, Ranunculaceae, norditerpenoid alkaloid

The alkaloids from *Aconitum* and *Delphinium* species are interesting because of their structural diversity and significant bioactivities.[1,2] *Delphinium densiflorum* has been used in traditional Chinese medicine over a long period for the treatment of ringworm, scabies and other skin diseases and inflammations.[3] However the chemical constituents and their biological activities have not been reported previously. In the course of our investigation on the alkaloids from the whole plant, a new norditerpenoid alkaloid was isolated. We now report on its isolation and structural elucidation.

Compound **1** was obtained as a colourless soild. The HR-ESI-MS gave a protonated molecular ion peak ($[M + H]^+$, *m/z* 464.2649, Calcd: 464.2643), suggesting a molecular formula of $C_{25}H_{37}NO_7$ with eight degrees of unsaturation. The IR spectrum indicated the presence of hydroxyl (3478 and 3453 cm^{-1}) and carbonyl (1745 cm^{-1}) groups. The ^1H NMR spectrum (Table 1) displayed a characteristic methyl signal at δ_H 2.06 (s) which, conjunction with ^{13}C NMR data (δ_C 170.0 (C) and 21.4 (Me)), indicated that an AcO group was present in **1**. In addition, the ^1H NMR spectrum displayed a signal for an *N*-ethyl group at δ_H 1.10 (3H, q, 6.8 Hz) and 2.89 (2H, m) and two methoxy groups at δ_H 3.35 and 3.48 (each 3H, s). This characteristic data suggested that **1** was a C_{19}-norditerpenoid alkaloid, most of which have a hydroxy or a methoxy group at C-1, C-14 and C-16.[4] Four fragments: C(1)–C(2)–C(3), C(5)–C(6)–C(7), C(9)–C(14)–C(13)–C(12)–C(10) and C(15)–C(16) were identified from the ^1H–^1H COSY and HSQC spectra. The HMBC experiments showed the following correlations: H-15/C-16, C-7, C-8, C-9 and C-13; H-5/C-18, C-7, C-19, C-11, C-3 and C-4; and H-1/C-11, C-17 and C-5; H-10/C-5 and C-11, which linked the above four fragments. Thus the planar structure of **1** was established as a C_{19}-norditerpenoid alkaloid. In the HMBC experiments correlations between δ_H 3.35, 3.48 and C-16 and C-8, respectively, indicated that the methoxy groups were located at C-16 and C-8. The correlation of H-14 with



Scheme 1

δ_C 170.0, indicated that the AcO group was located at C-14. The stereochemistry of **1** was established from the NOESY experiments which showed correlations between H-6, Ha-15 and H-21; between H-14 and H-13; between H-13, H-10 and Hb-12; between H-1 and H-10; between Ha-12, H-16 and H-19, and between Ha-15 and H-16. Considering a molecular model of **1**, if H-19 were arbitrarily assigned to α-orientation, the relative configation of H-1, H-6, H-14 and H-16 were then β, α, β, and α, respectively. Consequently the structure of **1** was established and named as delphidenine.

Experimental

Optical rotations were measured on a Perkin–Elmer Model 341 polarimeter. IR spectra were recorded on a Nicolet Avatar 360 FT-IR instrument using KBr discs over the range of 400–4000 cm^{-1}. 1D and 2D NMR spectra were obtained on a Varian Mercury-400bb NMR spectrometer with TMS as standard. HRESIMS determinations were run on a Bruker APEX Π FT-MS spectrometer. Analytical and preparative TLC were performed on silica gel plates (GF_{254} 10–40 μm, Qingdao Marine Chemical Factory). Analytical TLC was used to follow the separation and check the purity of isolated compounds. Spots on the plates were observed under UV light and visualised by spraying them with 5% H_2SO_4 in C_2H_5OH (v/v), followed by heating. Column chromatography (CC) was performed on silica gel (200–300 mesh, Qingdao Marine Chemical Factory).

Table 1 ^1H (400 MHz) and ^{13}C (100 MHz) NMR spectroscopic data for compound **1** ($CDC1_3$, δ in ppm, *J* in Hz)

No.	δ_H	δ_C	No.	δ_H	δ_C
1	3.73 (d, *J* = 4.4)	71.1 (d)	14	4.81 (t, 4.8)	75.0 (d)
2	5.79 (dd, 9.6, 4.4)	130.1 (d)	15	2.52 (m)/1.86 (m)	26.3 (t)
3	5.66 (d, 9.6)	137.6 (d)	16	3.35 (d, *J* = 4.8)	82.8 (d)
4	–	33.3 (s)	17	2.78 (s)	65.5 (d)
5	1.69 (br s)	57.8 (d)	18	1.09 (s)	23.6 (q)
6	4.31 (br s)	82.0 (d)	19	2.42 (m)	56.6 (t)
7	–	89.2 (s)	21	2.89 (m)	50.4 (t)
8	–	85.0 (s)	22	1.10 (q, *J* = 6.8)	13.5 (q)
9	2.16 (m)	44.5 (d)	OMe-(8)	3.48 (s)	57.5 (q)
10	2.58 (m)	37.2 (d)	OMe-(16)	3.35 (s)	56.4 (q)
11	–	49.6 (s)	OAc	2.06 (s)	21.4 (q)
12	2.28 (m)/2.23 (m)	27.7 (t)			170.0 (s)
13	3.25 (m)	42.5 (d)			

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Plant material

Delphinium densiflorum was collected in Tianzhu county (Gansu Province, China) in September 2007 and was identified by Prof. Yin-Fe Wang (College of Life Science, Northwest Normal University, Lanzhou, P.R. China). A voucher specimen (No. 200709DD) is deposited in Gansu Centre for Disease Control and Prevention, Lanzhou, China.

Extraction and isolation

The air-dried and powdered herb (4.5 kg) was extracted with 95% EtOH (15 L) three times (each time for 7 days) at room temperature to give a syrup 520g, which was dissolved in water and defatted with petroleum ether. The defatted aqueous extract was then extracted with EtOAc at two pH levels: pH 4–5 and 9–10, which was adjested by the addition of H_2SO_4 (2%) and NaOH (2%) solution, respectively. The fraction (pH 9–10, 39.0 g) was subjected to column chromatography (6 × 100 cm) on silica gel eluting with $CHCl_3$-CH_3OH (99 : 1, 50:1, 30 : 1, 10 : 1, 5 : 1) to afford five fractions (1–5). Fraction 2 (5.2 g) was further chromatographed on silica gel CC (2 × 50 cm) and eluted with petroleum ether-EtOAc (16 : 1, 8 : 1, 2 : 1) gradient to give **1** (5 mg).

Delphidenine (**1**); Colourless solid; $[\alpha]^{20}_{D}$: +68 (*c* 0.4 $CHCl_3$); IR (KBr) ν_{max} = 3478, 3453 and 1745 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) and ^{13}C NMR ($CDCl_3$, 100 MHz) see Table 1; HR-ESI-MS: *m/z* = 464.2649 (Calcd for $[M + H]^+$: 464.2643).

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References

1 Atta-ur-Rahman and M.I Choudhary, *Nat. Prod. Rep.*, 1997, **14**, 191.
2 Atta-ur-Rahman and M.I. Choudhary, *Nat. Prod. Rep.*, 1999, **16**, 619.
3 The Flora Committee of Chinese Academy of Sciences, *The Chinese flora*, Vol. 27 Science Press, Beijing, 1979, p.365.
4 D.R. Gardner, G.D. Manners, K.E. Panter, S.T. Lee and J.A. Pfister, *J. Nat. Prod.*, 2000, **63**, 1127.