Two Novel Actinidine-Type Monoterpene Alkaloids from *Incarvillea delavayi*

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Received June 28, 2000; accepted August 2, 2000

Two new actinidine-type monoterpene alkaloids, delavayines B (1) and C (2), were isolated from the MeOH extract of the aerial parts of *Incarvillea delavayi*, a close species of which, *I. sinensis*, is used as an analgesic for rheumatic pain in China, and the structures have been elucidated on the basis of spectroscopic evidence.

Key words *Incarvillea delavayi*; actinidine-type monoterpene alkaloid; delavayine B; delavayine C

We previously reported several new monoterpene alkaloids from the Chinese medical plant *Incarvillea sinensis*,^{1–5} including incarvillateine, which showed antinociceptive and anti-inflammatory activity.^{6,7} Subsequently, we obtained a related antinociceptive monoterpene alkaloid from *Incarvillea delavayi* Bureau & Franchet.⁸ Here, we report the isolation and structure determination of two additional new actinidine-type monoterpene alkaloids, delavayines B (1) and C (2) from *I. delavayi*.

The isolation of the monoterpene alkaloids 1 and 2 was performed by extracting the dried aerial parts of *I. delavayi* with MeOH, followed by fractionation of the crude extract by a variety of column chromatographic procedures (Diaion HP-20, Chromatorex and Si gel) to afford 1 (0.00080%, dry weight) and 2 (0.0011%, dry wt).

Compound 1 showed an $[M+H]^+$ ion peak at m/z 356 in the positive-mode FAB-MS, and its molecular formula was established as $C_{21}H_{25}NO_4$ by HR-FAB-MS. The ^{13}C -NMR signals of 1 were resembled to those of an actinidine-type alkaloid⁹ [δ 139.2 (d), 140.9 (d), 133.0 (s), 161.8 (s), 148.8 (s), 31.1 (t), 33.7 (t), 37.8 (d), 19.2 (q)] and a 2-substituted dihydrocinnamate group [δ 136.6 (s), 128.5 (d), 128.7 (d), 126.9 (s), 41.0 (t), 78.6 (d), 167.5 (s)]. The HMBC data confirmed the actinidine skeleton and allowed the placement of the dihydrocinnamate group at N-2 [connectivities between the C-8' carbon (δ 78.6 d) and the H-1, H-3, H-7a' and H-7b' protons (δ 8.42 s, 8.48 s, 3.37 m and 4.04 m, respectively)]. The two methoxy groups (δ_H 3.17 and δ_H 3.29 correlated with C-10 (δ 98.5), indicating the presence of a dimethyl acetal functionality (Fig. 1). Therefore, the structure

of delavayine B was established as 1, although the configurations at C-8, 8' were not determined. The MS and NMR data indicated that 1 was isolated as a zwitterion, as shown.¹⁰

The 1H - and ^{13}C -NMR spectra of 2 were similar to those of 1 for the actinidine portion of the molecule, but indicated a different substituent at N-2 [δ_H 2.55 (2H, m), 3.00 (1H, m), 3.17 (1H, m) and 5.20 (2H, m); δ_C 174.7 (s), 31.8 (t), 27.8 (t) and 60.7 (t)]. These signals were assignable to a γ -substituted butyrate moiety. The HMBC spectrum showed correlations between the H-4' and C-1, C-3, C-3' and C-2' signals; thus, the location of the butyrate side chain was assigned to N-2. Therefore, the structure of 2 was established, although again the stereochemistry at C-8 was not determined.

Investigation of the potential pharmacological activity of these compounds is now in progress.

Experimental

General Procedures Optical rotations were taken with a JASCO DIP-360 polarimeter. The 1H - and ^{13}C -NMR were recorded in pyridine- d_5 solution on JEOL JNM-GX-400 and JEOL α -500 spectrometers, and chemical shifts are given on a δ (ppm) scale with tetramethylsilane (TMS) as an internal standard. ^{13}C -NMR assignments were determined from HMQC and HMBC spectra. FAB-MS and HR-FAB-MS were recorded on a JEOL DX-303HF mass spectrometer. Column chromatography was carried out on silica gel 60 (spherical, 40–100 mesh, Kanto Chemicals), Chromatorex Chromatography Silica gel (DM-1020, 100–200 mesh, FUJI Silylia) and Diaion HP-20 (Mitsubishi Chemicals). TLC was performed on precoated Kieselgel 60 F₂₅₄ (0.2 mm, Merck), and spots and bands were viewed by UV light (254 and 366 nm). TLC solvent system was $CHCl_3$ –MeOH–H₂O (7 : 3 : 0.5).

Plant Material The bulbs of *Incarvillea delavayi* Bureau & Franchet (Bignoniaceae) were purchased from Heiwa-en Ltd., Tenri, Japan. The plants were grown at the Botanical Garden of the Faculty of Pharmaceutical Sciences, Kumamoto University, and collected in June 1998. A voucher specimen is kept at the same garden.

Extraction and Separation Dried aerial parts of *Incarvillea delavayi* (1.5 kg) were extracted with MeOH at room temperature. The MeOH extract was concentrated under reduced pressure to a syrup (yield 128 g), and was partitioned with 80% MeOH (MeOH–H₂O 8 : 2) and *n*-hexane. The 80% MeOH-sol. part was repeatedly chromatographed over Diaion HP-20 column with H₂O–MeOH (10 : 0–0 : 10), silica gel column with $CHCl_3$ –

Fig. 1. Selected HMBC Correlations in Delavayine B (1)

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MeOH-H₂O (30:1:0 to 6:4:1) and chromatorex column with CHCl₃-MeOH-NH₃ soln. (9:1:0.1 to 6:4:1) to afford compounds delavayine B (1, 12 mg) and C (2, 17 mg).

Compound 1: A off-white powder, $[\alpha]_D^{24}$ -6.5° ($c=1.8$, pyridine). Positive FAB-MS m/z : 356 $[M+H]^+$, negative FAB-MS m/z : 354 $[M-H]^-$. HR-FAB-MS m/z : 356.1861 $[M+H]^+$ (Calcd for C₂₁H₂₅NO₄: 356.1862). ¹H-NMR (pyridine-*d*₅) δ : 1.25 (3H, d, $J=8.0$ Hz, H-11), 1.73 (1H, m, H-7a), 2.45 (1H, m, H-7b), 3.00 (1H, m, H-6a), 3.13 (1H, m, H-6b), 3.17 (3H, s, OMe), 3.29 (3H, s, OMe), 3.33 (1H, m, H-8), 3.37 (1H, m, H-7'a), 4.04 (1H, m, H-7'b), 5.29 (1H, br d, $J=4.7$ Hz, H-8'), 5.43 (1H, s, H-10), 7.01 (2H, d, $J=7.0$ Hz, H-2'), 7.12 (2H, t, $J=7.0$ Hz, H-3'), 7.15 (1H, t, $J=7.0$ Hz, H-4'), 8.42 (1H, s, H-1), 8.48 (1H, s, H-3). ¹³C-NMR (pyridine-*d*₅) δ : 19.2 (C-11), 31.1 (C-6), 33.7 (C-7), 37.8 (C-8), 41.0 (C-7'), 52.4 (OMe), 53.1 (OMe), 78.6 (C-8'), 98.5 (C-10), 126.9 (C-4'), 128.5 (C-2', 6'), 128.7 (C-3', 5'), 133.0 (C-4), 136.6 (C-1'), 139.2 (C-1), 140.9 (C-3), 148.8 (C-9), 161.8 (C-5), 167.5 (C-9').

Compound 2: A off-white powder, $[\alpha]_D^{24}$ -72.3° ($c=0.5$, MeOH). ¹H-NMR (pyridine-*d*₅) δ : 1.37 (3H, d, $J=7.0$ Hz, H-11), 1.56 (1H, m, H-7a), 2.26 (1H, m, H-7b), 2.55 (2H, m, H-3'), 2.73 (2H, m, H-6), 3.00—3.17 (2H, m, H-2'), 3.36 (3H, s, OMe), 3.38 (1H, m, H-8), 3.40 (3H, s, OMe), 5.20 (2H, m, H-4'), 5.72 (1H, s, H-10), 9.28 (1H, s, H-3), 9.80 (1H, s, H-1). ¹³C-NMR (pyridine-*d*₅) δ : 19.3 (C-11), 27.8 (C-3'), 31.6 (C-6), 31.8 (C-2'), 34.0 (C-7), 38.4 (C-8), 53.5 (OMe), 54.0 (OMe), 60.7 (C-4'), 99.9 (C-10), 134.8 (C-4), 141.1 (C-3), 141.5 (C-1), 150.5 (C-9), 163.0 (C-5), 174.7 (C-1').

Acknowledgments We are grateful to Mr. I. Hori and Mr. K. Kitaoka of our faculty for cultivating the crude plants in the Botanical Garden, and to

Mr. K. Takeda and Mr. T. Iriguchi of Kumamoto University for measurement of the NMR and mass spectra.

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