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

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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 alkaloid[9] [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 ^{1}H- 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.

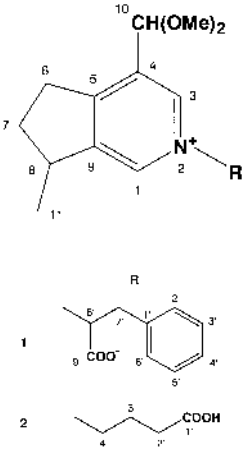
Investigation of the potential pharmacological activity of these compounds in now progress.

Experimental

General Procedures Optical rotations were taken with a JASCO DIP-360 polarimeter. The ^{1}H- 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 F_{254} (0.2 mm, Merck), and spots and bands were viewed by UV light (254 and 366 nm). TLC solvent system was $CHCl_3$–MeOH–H_2O (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–H_2O 8 : 2) and *n*-hexane. The 80% MeOH-sol. part was repeatedly chromatographed over Diaion HP-20 column with H_2O–MeOH (10 : 0—0 : 10), silica gel column with $CHCl_3$–



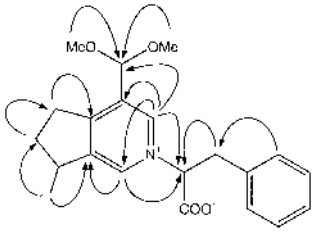


Fig. 1. Selected HMBC Correlations in Delavayine B (**1**)

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MeOH–H_2O (30 : 1 : 0 to 6 : 4 : 1) and chromatorex column with $CHCl_3$–MeOH–NH_3 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 $C_{21}H_{25}NO_4$: 356.1862). ^{1}H-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). ^{13}C-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). ^{1}H-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). ^{13}C-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′).

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