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Complanadine A, a new dimeric alkaloid from *Lycopodium complanatum*

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Abstract

A new dimeric alkaloid with a lycodine-type skeleton, complanadine A (**1**), has been isolated from the club moss *Lycopodium complanatum*, and the structure including the absolute stereochemistry was elucidated on the basis of spectroscopic data. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Lycopodium complanatum; alkaloid; lycodine; dimer; Lycopodiaceae.

Lycopodium alkaloids[1] are a group of natural products with unique ring systems, which have attracted great interest from biogenetic,[1,2] synthetic,[1,3] and biological[4] points of view. Recently we have isolated serratezomines A, B, and C with a seco-serratinine-type, a serratinine-type, and lycodoline-type skeletons, respectively, from the club moss *Lycopodium serratum* var. *serratum*.[5] Further search for structurally interesting compounds from another club moss resulted in the isolation of a new dimeric alkaloid with a lycodine type skeleton, complanadine A (**1**), from *Lycopodium complanatum* (Lycopodiaceae). Here we describe the isolation and structure elucidation of **1**.

The club moss *L. complanatum*[6] collected in Hokkaido were extracted with MeOH, and the MeOH extract was partitioned between EtOAc and 3% tartaric acid. Water-soluble materials, adjusted at pH 10 with sat. Na_2CO_3, were partitioned with $CHCl_3$. $CHCl_3$-soluble materials were subjected to an amino silica gel column (Hex/EtOAc, 1:0→0:1 and then $CHCl_3$/MeOH, 1:0→0:1), in which a fraction eluted with $CHCl_3$/MeOH (1:1) was purified by a silica gel column ($CHCl_3$/MeOH/EtOAc, 10:1:0.5) to afford complanadine A (**1**, 0.003% yield) together with a known related alkaloid, lycodine (**2**, 0.0005%).[7]

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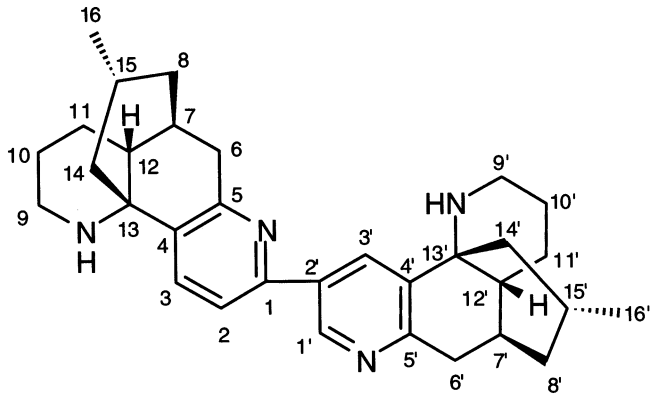
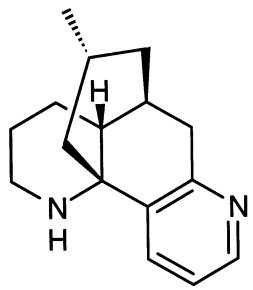


1



2

The molecular formula, $C_{32}H_{42}N_4$, of complanadine A (**1**) {$[\alpha]_D^{24}$ +14° (*c* 0.3, MeOH)} was established by HRFABMS [*m*/*z* 483.3477, $(M+H)^+$, Δ −1.1 mmu]. ^1H and ^{13}C NMR data (Table 1) disclosed the existence of ten olefinic carbons, two sp^3 quaternary carbons, six sp^3 methines, twelve sp^3 methylenes, and two secondary methyls. Among them, four olefinic carbons [δ_c 147.15 (d), 154.21 (s), 160.09 (s), and 160.31 (s)] assignable to nitrogen-bearing carbons were elucidated to form two three-substituted pyridine rings together with the remaining six olefinic carbons [δ_c 120.49 (d), 132.70 (s), 133.08 (d), 134.17 (s), 134.78 (s), and 135.25 (d)], while two quaternary carbons (δ_c 60.23 and 60.47) and two methylenes (δ_c 42.05; δ_H 2.73 and 3.05, and δ_c 42.11; δ_H 2.73 and 3.07) were ascribed to those attached to a nitrogen. UV absorptions [290 (ε 14000) and 251 nm (12000)] also supported the presence of the pyridine ring. Since eight out of fourteen unsaturations were accounted for, **1** was inferred to possess six more rings. The gross structure of **1** was elucidated by analyses of 2D NMR data including ^1H–^1H COSY, HOHAHA, HMQC, and HMBC spectra in CD_3OD (Fig. 1). Each pair of these ^1H and ^{13}C NMR signals seemed to be due to each half moiety (parts A and B) of a dimeric compound. In part A, connectivities of C-2 to C-3, C-6 to C-8, C-9 to C-12, C-14 to C-16, C-7 to C-12, and C-8 to C-15 were revealed by the ^1H–^1H COSY and HOHAHA spectra. The presence of a 2,3,6-trisubstituted pyridine ring was elucidated from HMBC correlations of H-3 (δ_H 8.00) to C-1 and C-5, and H-2 (δ_H 7.89) to C-4. HMBC correlations of H-6 to C-5 (δ_C 160.31) and C-4 (δ_C 132.70) suggested the connectivity from C-6 to C-5. The presence of a piperidine ring (N-9 and C-9 to C-13) was deduced from the HMBC correlation of H-9 to C-13 (δ_c 60.23) through a nitrogen atom. HMBC cross-peaks of H-12 to C-4, and H_2-14 to C-13 and C-4 revealed connectivities from C-12 to C-4 and from C-14 to C-4 through C-13, constructing a lycodine-type ring system (part A). On the other hand, the corresponding ^1H–^1H COSY, HOHAHA, and HMBC correlations were also observed for part B (Fig. 1). HMBC correlations of H-1′ to C-2′, C-3′, and C-5′, and H-3′ to C-1′ and C-5′ indicated the existence of a 2,3,5-trisubstituted pyridine ring, constructing another lycodine-type ring system (part B).

The connection between each pyridine ring in parts A and B was provided by the HMBC correlation of H-3′ (δ_H 8.61) to C-1 (δ_c 154.21), thus giving rise to the connectivity of C-1 to C-2′. NOESY correlations of H-2/H-3′ and H-2/H-1′ (Fig. 2) also supported the connectivity

Table 1
^{1}H and ^{13}C NMR data of complanadine A (**1**) in CD_3OD at 300 K

	δ_H	δ_C	HMBC (^{1}H)
1		154.21	3, 3′
2	7.89 (1H, d, 8.2)	120.49	
3	8.00 (1H, d, 8.2)	135.25	
4		132.70	2, 6, 12, 14
5		160.31	3, 6
6a	2.82 (1H, d, 19.2)	35.95	8, 12
6b	3.27 (1H, dd, 7.2, 19.2)		
7	2.26 (1H, m)	34.43	6, 8
8a	1.43 (1H, dt, 3.9, 13.0)	43.61	6, 14, 15, 16
8b	1.86 (1H, brd, 12.7)		
9a	2.73 (1H, m)	42.05	11
9b	3.05 (1H, brd, 13.5)		
10	1.80 (2H, m)	27.08	9, 11
11	1.67 (2H, m)	26.12	9
12	1.96 (1H, brs)	43.76	6, 8, 14
13		60.23	9b, 14
14a	1.53 (1H, dd, 4.8, 12.1)	49.96	8, 16
14b	1.74 (1H, m)		
15	1.29 (1H, m)	25.65	7, 14, 16
16	0.86 (3H, d, 6.4)	22.06	14
1′	9.03 (1H, d, 2.1)	147.15	3′
2′		134.78	1′
3′	8.61 (1H, d, 2.1)	133.08	1′
4′		134.17	6′, 12′, 14′
5′		160.09	1′, 3′, 6′
6a′	2.77 (1H, d, 19.2)	35.46	8′, 12′
6b′	3.24 (1H, dd, 7.2, 19.2)		
7′	2.26 (1H, m)	34.48	6′, 8′
8a′	1.43 (1H, dt, 3.9, 13.0)	43.76	6′, 14′, 15′, 16′
8b′	1.86 (1H, brd, 12.7)		
9a′	2.73 (1H, m)	42.11	11′
9b′	3.07 (1H, brd, 13.5)		
10′	1.80 (2H, m)	27.11	9′, 11′
11′	1.67 (2H, m)	26.23	9′
12′	1.94 (1H, brs)	43.61	6′, 8′, 14′
13′		60.47	9b′, 14′
14a′	1.51 (1H, dd, 4.8, 12.1)	50.13	8′, 16′
14b′	1.74 (1H, m)		
15′	1.29 (1H, m)	25.93	7′, 14′, 16′
16′	0.86 (3H, d, 6.4)	22.06	14′

between parts A and B. Thus the gross structure of complanadine A (**1**) was assigned as a dimer of lycodine (**2**), in which C-1 in part A was connected to C-2′ in part B.

The phase sensitive NOESY spectrum of **1** showed cross-peaks as shown in computer-generated 3D drawing (Fig. 2). The relative configurations at C-7, C-12, C-13, and C-15 in part A were based on NOESY correlations of H_b-6/H-15, H-12/H_b-10 and H-3/H_b-14, while the

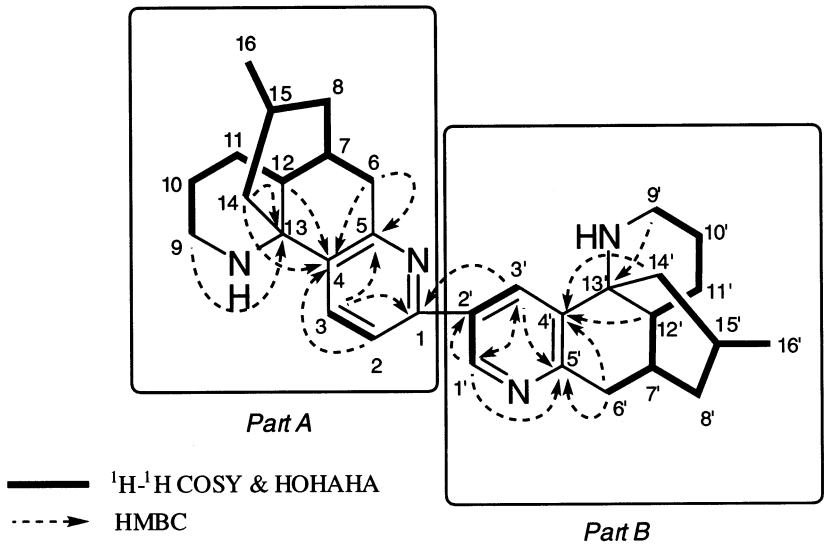


Figure 1. Selected 2D NMR correlations of complanadine A (**1**)

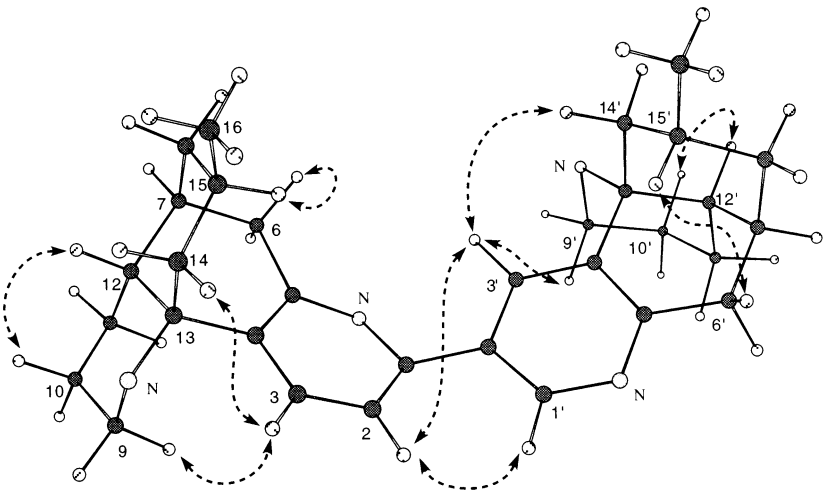


Figure 2. Selected NOESY correlations (dotted arrows) and relative stereochemistry for complanadine A (**1**)

piperidine and cyclohexane (C-7, C-8, and C-12 to C-15) rings adopted both chair conformations. On the other hand, the corresponding NOESY correlations were also observed for part B. Thus, the relative stereostructure of complanadine A (**1**) was elucidated to be that shown in Fig. 2. The CD spectrum of **1** in MeOH showed similar CD curves [λ_{max} 260 (θ 4500), 285 (1500), 295 (2000), and 315 (3000) nm] to those [λ_{max} 250 (θ 5000), 280 (3000), and 325 (3000) nm] of lycodine (**2**), indicating the same absolute stereochemistry for parts A and B of **1** as that of lycodine (**2**).[7]

Complanadine A (**1**) is the first dimeric alkaloid containing a lycodine-type $C_{16}N_2$ skeleton among many *Lycopodium* alkaloids reported so far.[1] Complanadine A (**1**) was cytotoxic against murine leukemia L1210 cells (IC_{50}, 5.6 μg/ml) in vitro.

Acknowledgements

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