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Lycopladine A, a new $C_{16}N$ alkaloid from *Lycopodium complanatum*

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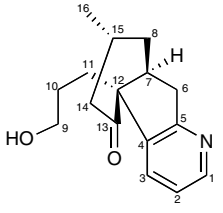
Abstract—A new $C_{16}N$ type alkaloid, lycopladine A (**1**), has been isolated from the club moss *Lycopodium complanatum*, and the structure and relative stereochemistry of **1** were elucidated on the basis of spectral data.

Club moss (Lycopodiaceae) is known to be a rich source of *Lycopodium* alkaloids possessing unique heterocyclic ring systems such as $C_{16}N$, $C_{16}N_3$, and $C_{27}N_3$, which have attracted great interest from biogenetic, synthetic, and biological points of view.[1] In our continuing efforts to find new *Lycopodium* alkaloids,[2] a new $C_{16}N$ type alkaloid, lycopladine A (**1**),[3] was isolated from the club moss *Lycopodium complanatum*. In this letter, we describe the isolation and structure elucidation of **1**.



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The club moss *L. complanatum* collected at Nayoro in Hokkaido was extracted with MeOH, and the MeOH extract was partitioned between EtOAc and 3% tartaric acid. Water-soluble materials, which were adjusted at pH 10 with saturated Na_2CO_3, were partitioned with $CHCl_3$. $CHCl_3$-soluble materials were subjected to an amino silica gel column (hexane/EtOAc, 50:1→1:1 and then $CHCl_3$/MeOH, 1:0→0:1), in which a fraction eluted with $CHCl_3$/MeOH (1:0 and 50:1) was purified by a silica gel column ($CHCl_3$/MeOH, 1:0→4:1) to afford lycopladine A (**1**, 0.0001% yield).

Lycopladine A (**1**) showed the pseudomolecular ion peak at *m/z* 260 $(M+H)^+$ in the ESIMS, and the molecular formula, $C_{16}H_{21}NO_2$, was established by HRESIMS [*m/z* 260.1653, $(M+H)^+$, Δ +0.2 mmu]. IR absorptions implied the presence of hydroxyl group (3380 cm^{-1}) and ketone carbonyl (1700 cm^{-1}). ^{13}C NMR data of **1** (Table 1) revealed 16 carbon signals due to one carbonyl carbon, two sp^2 quaternary carbons, three sp^2 methines, one sp^3 quaternary carbon, two sp^3 methines, six sp^3 methylenes, and one methyl group. Among them, two olefinic carbons [δ_C 148.8 (d), 164.3 (s)] assignable to nitrogen-bearing carbons were elucidated to form a disubstituted pyridine ring together with the remaining three olefinic carbons [δ_C 123.0 (d), 136.1 (d), 140.0 (s)]. The UV absorption [270 nm (ε 2800)] also supported the presence of the pyridine ring. Since five out of seven unsaturations were accounted for, **1** was inferred to possess two 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). The ^1H–^1H COSY and HOHAHA spectra of **1** revealed three structural units **a** (C-1–C-3), **b** (C-6–C-8, C-8–C-15, and C-14–C-16), and **c** (C-9–C-11). It was elucidated that unit **a** constituted a 2,3-disubstituted pyridine ring by HMBC correlations of H-1 and H-3 (δ_H 8.30, 7.67, respectively) to C-5, and H-2 (δ_H 7.24) to C-4. HMBC correlations of H-11a (δ_H 2.06) to C-4, C-7,

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Table 1. ^{1}H and ^{13}C NMR data of Lycopladine A (**1**) in CD_3OD

Position	δ_H	δ_C
1	8.30 (1H, dd, 5.0, 1.4)	148.8 d
2	7.24 (1H, dd, 7.6, 5.1)	123.0 d
3	7.67 (1H, dd, 7.7, 1.4)	136.1 d
4		140.0 s
5		164.3 s
6a	3.09 (1H, dd, 16.5, 8.2)	38.6 t
6b	2.83 (1H, dd, 16.5, 9.1)	
7	2.97 (1H, m)	43.5 d
8a	1.90 (1H, m)	34.8 t
8b	1.83 (1H, m)	
9	3.53 (2H, m)	62.8 t
10a	1.56 (1H, m)	29.1 t
10b	1.35 (1H, m)	
11a	2.06 (1H, ddd, 13.6, 13.6, 4.6)	33.4 t
11b	1.88 (1H, m)	
12		62.7 s
13		214.6 s
14	2.29 (2H, m)	47.7 t
15	2.12 (1H, m)	29.5 d
16	1.08 (3H, d, 6.5)	22.0 q

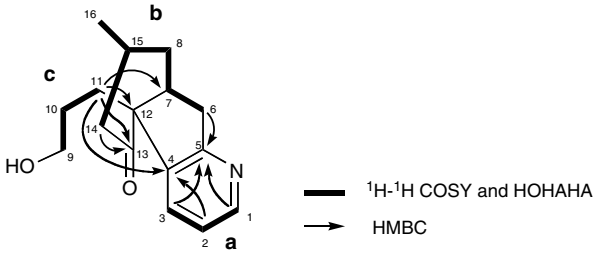


Figure 1. Selected 2D NMR correlations for lycopladine A (**1**).

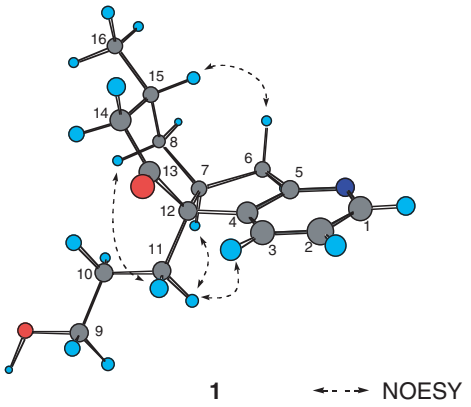
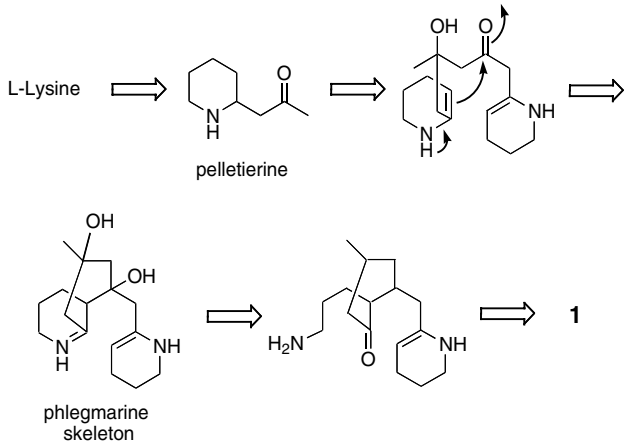


Figure 2. Selected NOESY correlations and relative stereochemistry for lycopladine A (**1**).



Scheme 1. Plausible biogenetic path of lycopladine A (**1**).

and C-12 revealed the connectivities from C-11 to C-4 and C-7 through C-12. The HMBC correlations of H_2-11 and H_2-14 (δ_H 2.29) to C-13 indicated the connectivities from C-11 to C-14 through C-12 and C-13. HMBC cross-peaks of H_2-6 (δ_H 3.09, 2.83) to C-5 suggested the connectivity from C-6 to C-5. The remaining C-9 (δ_H 3.53, δ_C 62.8) was elucidated to be connected with a hydroxyl group. Thus, the gross structure of lycopladine A was assigned as **1**.

The NOESY spectrum of **1** showed cross-peaks as shown in computer-generated 3D drawing (Fig. 2). The relative configurations of C-7, C-12, and C-15 in the cyclohexanone ring (C-7, C-8, and C-12 to C-15) were deduced from NOESY correlations of H-3/H_2-11, H-6b/H-15, H-7/H_2-11, H-8b/H_2-11, and H_2-14/H_2-11. Thus, the relative configurations of lycopladine A (**1**) were elucidated as shown in Figure 2.

Lycopladine A (**1**) possesses an unprecedented skeleton different from known $C_{16}N$-type alkaloids. A plausible biogenetic path of lycopladine A (**1**) was proposed as shown in Scheme 1. Biogenetically, lycopladine A (**1**) might be derived from L-lysine via pelletierine and then plegmarine skeleton.[4,5] Lycopladine A (**1**) showed weak cytotoxicity against murine lymphoma L1210 cells (IC_{50}, 7 μg/mL) in vitro, while **1** did not show such activity against human epidermoid carcinoma KB cells ($IC_{50} > 10$ μg/mL).

Acknowledgements

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References and notes

1. For reviews of *Lycopodium* alkaloids, see: (a) Kobayashi, J.; Morita, H. In *The Alkaloids*; Cordell, G. A., Ed.; Academic: New York, 2005; Vol. 61, p 1; (b) Ayer, W. A.; Trifonov, L. S. In *The Alkaloids*; Cordell, G. A., Brossi, A., Eds.; Academic: New York, 1994; Vol. 45, p 233.
2. Hirasawa, Y.; Kobayashi, J.; Morita, H. *Org. Lett.* **2006**, *8*, 123–126, and references cited therein.

3. *Lycopladine A* (**1**). A colorless solid; $[\alpha]_D^{23}$ +102 (*c* 1.0, MeOH); IR (neat) ν_{max} 3380 and 1700 cm^{-1}; UV (MeOH) λ_{max} 270 nm (ε 2800); ^1H and ^{13}C NMR data (Table 1); ESIMS *m/z* 260 $(M+H)^+$; HRESIMS *m/z* 260.1653 (M+H; calcd for $C_{16}H_{22}NO_2$, 260.1651).
4. Hemscheidt, T.; Spenser, I. D. *J. Am. Chem. Soc.* **1996**, *118*, 1799–1800.
5. Nyembo, L.; Goffin, A.; Hootele, C.; Braekman, J.-C. *Can. J. Chem.* **1978**, *56*, 851–856.