SUPPLEMENTARY MATERIAL

A New Lycopodine-type Alkaloid from *Lycopodium japonicum*

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A new lycopodine-type alkaloid, 12 β -hydroxy-acetylfawcettiine *N*-oxide (**1**), together with seven known analogues, acetyllycoposerramine M (**2**), lycopodine (**3**), lycoclavine (**4**), diphaladine A (**5**), lycoposerramine K (**6**), 11 β -hydroxy-12-*epilycodoline* (**7**) and fawcettiine (**8**), were isolated from *Lycopodium japonicum*. Their structures were established by mass spectrometry and 1D and 2D NMR techniques. The isolated alkaloids were assayed for their inhibition activities against acetylcholinesterase, but no inhibitory activities for the compounds were detected.

Keywords: *Lycopodium japonicum*; Lycopodiaceae; lycopodine-type alkaloids

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Supporting information

Spectroscopic data of the known compounds **2-8**.

Figure 2. Selected 2D NMR correlations of **1**.

Original spectra of compound **1**.

ESI-MS

HR-ESI-MS

IR Spectrum

UV Spectrum

^1H NMR Spectrum (500 MHz, CDCl_3)

^{13}C NMR Spectrum (125 MHz, CDCl_3)

^1H - ^1H COSY Spectrum

HSQC Spectrum

HMBC Spectrum

ROESY Spectrum

Spectroscopic data of the known compounds **2-8**.

acetyllycoposerramine M (2)

Colorless solid; C₁₈H₂₇NO₃; MW: 305; ¹H-NMR (500 MHz, CDCl₃) δ_H: 5.12 (m, H-11), 3.26 (m, H-1a), 3.24 (m, H-4), 0.77 (d, *J* = 6.5 Hz, H-16); ¹³C-NMR (125 MHz, CDCl₃) δ_C: 214.0 (s, C-5), 169.9 (s, C-17), 72.1 (d, C-11), 58.7 (s, C-13), 46.9 (t, C-1), 46.5 (d, C-12), 45.2 (d, C-4), 44.2 (t, C-6), 44.1 (t, C-8), 43.6 (t, C-14), 41.9 (t, C-9), 34.9 (d, C-7), 30.8 (t, C-10), 25.2 (d, C-15), 22.6 (q, C-16), 21.7 (q, C-17), 19.8 (t, C-3), 19.1 (t, C-2).

lycopodine (3)

Colorless solid; C₁₆H₂₅NO; MW: 247; ¹H-NMR (500 MHz, CDCl₃) δ_H: 3.30 (td, 14.0, 3.5, H-9a), 3.07 (td, 12.0, 3.0, H-1a), 2.78 (d, 12.0, H-1b), 0.75 (d, 6.5, H-16); ¹³C-NMR (125 MHz, CDCl₃) δ_C: 213.0 (s, C-5), 60.0 (s, C-13), 47.1 (t, C-9), 46.5 (t, C-1), 44.7 (d, C-12), 42.9 (t, C-6), 42.8 (d, C-4), 42.7 (t, C-14), 42.3 (t, C-8), 36.6 (d, C-7), 25.8 (t, C-10), 25.2 (d, C-15), 24.9 (t, C-11), 22.7 (q, C-16), 19.3 (t, C-3), 18.6 (t, C-2).

lycoclavine (4)

Colorless solid; C₁₈H₂₉NO₃; MW: 307; ¹H-NMR (500 MHz, CDCl₃) δ_H: 4.69 (s, H-5), 2.07 (s, H-18), 0.91 (d, 6.5, H-16); ¹³C-NMR (125 MHz, CDCl₃) δ_C: 170.2 (s, C-17), 80.0 (d, C-5), 71.6 (d, C-6), 57.8 (s, C-13), 46.7 (t, C-1), 46.6 (t, C-9), 43.5 (d, C-12), 41.0 (d, C-7), 40.1 (t, C-14), 39.6 (t, C-8), 30.0 (d, C-4), 25.2 (t, C-10), 25.0 (t, C-11), 24.0 (d, C-15), 23.5 (q, C-16), 22.0 (t, C-3), 21.4 (q, C-18), 19.8 (t, C-2).

diphaladine A (5)

Colorless oil; C₁₆H₂₅NO₄; MW: 295; ¹H-NMR (500 MHz, CDCl₃) δ_H: 4.40 (dt, 12.0, 2.0, H-9a), 4.08 (s, H-11a), 3.90 (d, 10.0, H-4), 0.89 (d, 6.5, H-16). ¹³C-NMR (125 MHz, CDCl₃) δ_C: 209.7 (s, C-5), 73.4 (s, C-13), 72.4 (s, C-12), 72.0 (s, C-11), 63.2 (t,

C-1), 55.7 (t, C-9), 48.6 (d, C-4), 45.1 (t, C-6), 40.4 (d, C-7), 36.9 (t, C-8), 30.2 (t, C-14), 26.5 (t, C-10), 24.7 (d, C-15), 22.4 (q, C-16), 21.5 (t, C-2), 17.5 (t, C-3).

lycoposerramine K (6)

Colorless solid; $C_{16}H_{23}NO_2$; MW: 261; 1H -NMR (500 MHz, $CDCl_3$) δ_H : 5.73 (s, H-11), 3.85 (d, 2.5, H-6), 0.86 (d, 6.0, H-16); ^{13}C -NMR (125 MHz, $CDCl_3$) δ_C : 210.9 (s, C-5), 139.2 (s, C-12), 120.9 (d, C-11), 78.1 (d, C-6), 59.8 (s, C-13), 49.5 (d, C-4), 47.9 (d, C-7), 47.7 (t, C-1), 45.0 (t, C-9), 39.1 (t, C-8), 36.9 (t, C-14), 26.3 (t, C-10), 25.5 (d, C-15), 22.7 (t, C-2), 22.7 (q, C-16), 19.3 (t, C-3).

11 β -hydroxy-12-epilycodoline (7)

Colorless solid; $C_{16}H_{26}NO_3$; MW: 280; 1H -NMR (500 MHz, $CDCl_3$) δ_H : 2.86 (t, 12.5, H-4), 0.82 (d, 6.0, H-16). ^{13}C -NMR (125 MHz, $CDCl_3$) δ_C : 212.2 (s, C-5), 73.6 (d, C-11), 70.7 (s, C-12), 62.1 (s, C-13), 53.4 (d, C-4), 48.3 (t, C-1), 45.6 (t, C-6), 43.4 (t, C-9), 40.5 (d, C-7), 37.3 (t, C-8), 31.4 (t, C-10), 25.0 (t, C-14), 24.6 (t, C-2), 23.8 (d, C-15), 22.2 (q, C-16), 17.9 (t, C-3).

fawcettiine (8)

Colorless solid; $C_{18}H_{29}NO_3$; MW: 307; 1H -NMR (500 MHz, $CDCl_3$) δ_H : 5.11 (s, H-5), 3.21 (t, 12, H-8), 1.43 (m, H-18), 1.08 (d, 6.5, H-16). ^{13}C -NMR (125 MHz, $CDCl_3$) δ_C : 170.4 (s, C-17), 78.7 (d, C-8), 69.5 (d, C-5), 55.0 (s, C-13), 47.4 (t, C-1), 46.7 (t, C-9), 43.2 (d, C-12), 41.3 (d, C-7), 41.0 (t, C-14), 31.9 (d, C-15), 31.2 (d, C-4), 25.9 (t, C-10), 24.3 (t, C-6), 24.2 (t, C-11), 22.6 (t, C-3), 21.5 (q, C-18), 20.8 (q, C-16), 19.8 (t, C-2).

Figure 2. Selected 2D NMR correlations of **1**.

Original spectra of compound 1.

ESI-MS:

Sample Name	NYQ-1	Position	P1-A3	Instrument Name	Instrument 1	User Name	
Inj Vol	0.1	InjPosition		SampleType	Sample	IRM Calibration Status	Success
Data Filename	NYQ-1.d	ACQ Method	SIBU.m	Comment		Acquired Time	9/14/2015 3:52:17 PM

Original spectra of compound 1.

HR-ESI-MS:

Qualitative Analysis Report

Data Filename	NYQ-1.d	Sample Name	NYQ-1
Sample Type	Sample	Position	P1-A3
Instrument Name	Instrument 1	User Name	
Acq Method	SIBU.m	Acquired Time	9/14/2015 3:52:17 PM
IRM Calibration Status	Success	DA Method	ESI+.m
Comment			

Sample Group		Info.
Acquisition SW	6200 series TOF/6500 series	
Version	Q-TOF B.05.01 (B5125.2)	

User Spectra

Peak List

m/z	z	Abund	Formula	Ion
296.1856	1	59844.51		
382.2227	1	4291236	C20 H31 N O6	(M+H)+
383.226	1	901494.5	C20 H31 N O6	(M+H)+
384.2283	1	119785.92	C20 H31 N O6	(M+H)+
404.2041	1	100472.8		
763.438	1	897267		
764.4415	1	370063		
765.4441	1	95267.63		

Formula Calculator Element Limits

Element	Min	Max
C	3	60
H	0	120
O	0	30
N	0	30

Formula Calculator Results

Formula	CalculatedMass	CalculatedMz	Mz	Diff. (mDa)	Diff. (ppm)	DBE
C20 H31 N O6	381.2151	382.2224	382.2227	-0.3	-0.7	6.0000

--- End Of Report ---

IR Spectrum

Sample : NYQ-1		Frequency Range : 399.246 - 3996.32		Measured on : 21/09/2015	
Technique : KBr压片	Resolution : 4	Instrument : Tensor27		Sample Scans : 16	
Customer : 150921IR0	Zerofilling : 2	Acquisition : Double Sided,Forw			

UV Spectrum

¹H NMR:

¹³C NMR:

H-H COSY:

HSQC:

HMBC:

ROESY: