Diterpenoid and Norditerpenoid Alkaloids from the Roots of *Aconitum yesoense* var. *macroyesoense*

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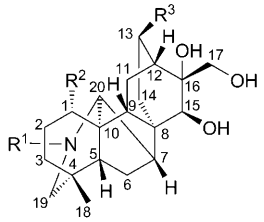
A new diterpenoid alkaloid, macroyesoenline (**1**), and a new norditerpenoid alkaloid, 9-hydroxyvirescenine (**2**), have been isolated from the roots of *Aconitum yesoense* var. *macroyesoense*. Their structures were elucidated on the basis of spectral data (1D- and 2D-NMR experiments) and mass spectrometry. The known norditerpenoid and diterpenoid alkaloids delphinifoline (**3**) and cochlearenine (**13**) were isolated for the first time from this plant.

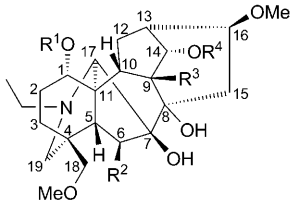
Introduction. – We have investigated the alkaloidal components of *Aconitum yesoense* var. *macroyesoense* (NAKAI) TAMURA [1], a plant native to Hokkaido island, Japan. Pharmacological studies demonstrated that diterpenoid alkaloid components and their derivatives show cytotoxicity against A172 human malignant glioma cells [2]. By conventional chromatographic procedures, new diterpenoid and norditerpenoid alkaloids were isolated and named macroyesoenline (**1**) and 9-hydroxyvirescenine (**2**). Additionally, five known C_{19}-norditerpenoid alkaloids, delphinifoline (**3**) [3], 14-acetylbrowniine (**4**) [4], 14-acetyldelcosine (**5**) [4], browniine (**6**) [5] and delcosine (**7**) [4], and six known C_{20}-diterpenoid alkaloids, kobusine (**8**) [6], pseudokobusine (**9**) [6], yesonine (**10**) [7], lucidusculine (**11**) [6], dehydrolucidusculine (**12**) [8], and cochlearenine (**13**) [9] were found. Delphinifoline (**3**) and cochlearenine (**13**) have not been reported previously from *Aconitum yesoense* var. *macroyesoense*. In this report, the structure elucidation of two new minor alkaloids by applying 2D-NMR techniques is described. Since *Pelletier et al.* [10] previously reported only partial ^{13}C-NMR data for delphinifoline (**3**), the complete ^{13}C-NMR assignments are reported.

Results and Discussion. – The pulverized, air-dried roots of *A. yesoense* var. *macroyesoense* were extracted with 90% MeOH. The residue of the extract was dissolved in 5% HCl and extracted with hexane. The acidic fraction was basified at pH 10 with aqueous ammonia, and then extracted with $CHCl_3$. The hexane fraction was extracted with 5% HCl and the acidic fraction was basified at pH 10 with aqueous ammonia, and then extracted with $CHCl_3$. The combined $CHCl_3$ extract was concentrated to afford a crude alkaloidal mixture. Extensive purification by repeated chromatography finally afforded compounds **1** and **2**, as well as eleven known alkaloids.

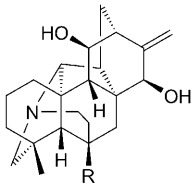
Compound **1** was isolated as a colorless amorphous solid. The molecular formula of **1** was determined to be $C_{23}H_{35}NO_6$ by HR-EI-MS (*m*/*z* 421.2485 (M^+)). The assignments of the ^{1}H- and ^{13}C-NMR signals (*Table 1*) of **1** were accomplished by a



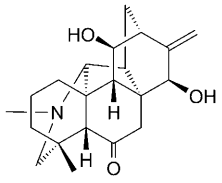
1 R^1 = Me, R^2 = AcO, R^3 = OH
13 R^1 = Et, R^2 = OH, R^3 = H
14 R^1 = Me, R^2 = R^3 = H



2 R^1 = R^2 = R^4 = H, R^3 = OH
3 R^1 = R^3 = R^4 = H, R^2 = OH
4 R^1 = Me, R^2 = MeO, R^3 = H, R^4 = Ac
5 R^1 = R^3 = H, R^2 = MeO, R^4 = Ac
6 R^1 = Me, R^2 = MeO, R^3 = R^4 = H
7 R^1 = R^3 = R^4 = H, R^2 = MeO
15 R^1 = R^2 = R^3 = R^4 = H



8 R = H
9 R = OH



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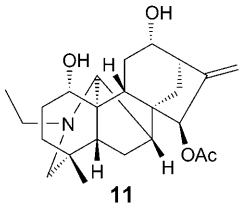

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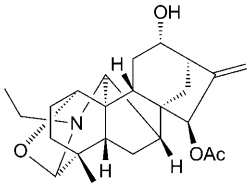

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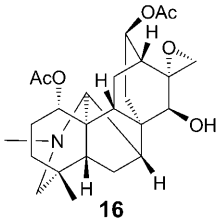

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combination of DEPT, HMBC (*Fig. 1* and *Table 1*), and NOESY (*Fig. 2* and *Table 1*) data, which allowed to elucidate the structure of **1**.

The IR spectrum of **1** indicated the presence of a OH group (3350 cm^{-1}) and an ester function (1720 cm^{-1}). The ^{1}H-NMR spectrum revealed the presence of a Me

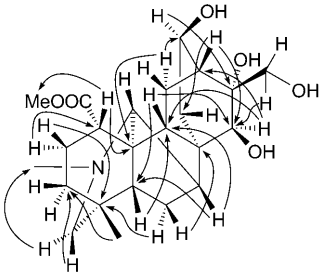

Fig. 1. *Key HMBC of* **1**

Table 1. *^{1}H-NMR* (400 MHz) *and ^{13}C-NMR* (100 MHz, DEPT) *Data*[a]) *for* **1**, *and ^{13}C-NMR* (100 MHz, DEPT) *Data for* **13** *in $CDCl_3$*. δ in ppm, J in Hz.

Position	**1**				**13**
	δ(C)	δ(H)	HMBC (C → H)	NOESY	δ(C)
1	74.3 (*d*)	5.02 (*dd*, J = 10.7, 6.3)	H_α–C(2), H–C(3)	H_β–C(2)	70.9 (*d*)
2	26.5 (*t*)	2.19–2.32 (*m*, H_α) 1.83–1.89 (*m*, H_β)	H–C(1)	H_a–C(19), H–C(20)	31.6 (*t*)
3	38.0 (*t*)	1.29–1.39 (*m*) 1.57–1.63 (*m*)	Me(18), CH_2(19)	Me(18)	38.4 (*t*)
4	33.6 (*s*)		H_β–C((2), H–C(5), CH_2(6), Me(18), CH_2(19)		33.5 (*s*)
5	53.0 (*d*)	1.38 (*d*, J = 7.5)	H–C(3), CH_2(6), H–C(7), H–C(9), Me(18), H_a–C(19), H–C(20)	H_β–C(6), H–C(9), Me(18)	53.1 (*d*)
6	23.2 (*t*)	1.21–1.27 (*m*, H_α) 2.76 (*dd*, J = 13.6, 7.8, H_β)	H–C(20)	Me(18), H_b–C(19) H–C(5)	23.8 (*t*)
7	41.7 (*d*)	2.22–2.24 (*m*)	H–C(5), CH_2(6)	H–C(15)	36.8 (*d*)
8	43.4 (*s*)		CH_2(6), H–C(9), H–C(14), H–C(15)		42.6 (*s*)
9	38.5 (*d*)	1.79–1.83 (*m*)	H–C(5), H–C(7), CH_2(11), H–C(12), H_β–C((14), H–C(15)	H–C(5), H_b–C(17)	50.8 (*d*)
10	48.0 (*s*)		H–C(1), H–C(5), H–C(7), H–C(9), H_α–C(11), H–C(20)		48.5 (*s*)
11	22.9 (*t*)	1.06 (*t*, J = 12.2, H_α) 1.62–1.72 (*m*, H_β)	H–C(9), H–C(12)	H–C(13)	21.5 (*t*)
12	40.1 (*d*)	1.79–1.86 (*m*)	CH_2(11), H–C(15)	H–C(13), H_a–C(17)	42.4 (*d*)
13	71.5 (*d*)	3.88–3.94 (*m*)	CH_2(11), H–C(12), H_β–C(14)	H_α–C(11), H–C(12), H_α–C(14)	23.7 (*t*)
14	40.0 (*t*)	2.44–2.52 (*m*, H_α)	H–C(9), H–C(12), H–C(15)	H–C(13), H–C(20)	26.9 (*t*)
		1.21–1.27 (*m*, H_β)		H_α–C(15)	
15	86.5 (*d*)	4.16 (*s*)	H–C(9), H–C(12), CH_2(14), CH_2(17)	H–C(7), H_β–C(14)	87.5 (*d*)
16	80.5 (*s*)		H_α–C(11), H–C(12), H–C(13), H–C(15), CH_2(17)		78.8 (*s*)
17	67.1 (*t*)	3.55 (*AB*, J = 11.4, H_a) 4.01 (*AB*, J = 11.4, H_b)		H–C(12) H–C(9)	67.9 (*t*)
18	25.8 (*q*)	0.71 (*s*)	H–C(5), H_b–C(19), MeN	H–C(3), H–C(5), H_α–C(6), CH_2(19)	26.8 (*q*)
19	59.1 (*t*)	2.44 (*AB*, J = 11.2, H_a)	H–C(3), H–C(5), Me(18), H–C(20), MeN	H_α–C(6), Me(18)	57.0 (*t*)
		2.30 (*AB*, J = 11.2, H_b)		H_α–C(2)	
20	69.5 (*d*)	3.50 (*s*)	H–C(1), H–C(5), H–C(6), H–C(9), H_a–C(19), MeN	H_α–C(2), H_α–C(14)	67.2 (*d*)
N–*CH_2*Me	–	–	–	–	51.1 (*t*)
MeN or N–CH_2*Me*	43.8 (*q*)	2.30 (*s*)	H_b–C(19)		13.6 (*q*)
CO	170.9 (*s*)		H–C(1), Me		
Me	21.9 (*q*)	2.04 (*s*)			

[a]) Chemical shifts were assigned on the basis of ^{1}H,^{1}H-COSY, HETCOR, and HMBC experiments.

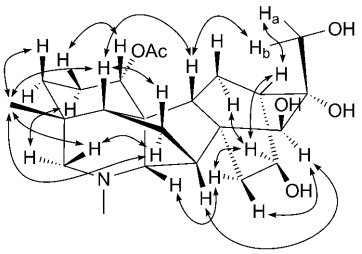

Fig. 2. *Key NOESY correlations of* **1**

group (δ(H) 0.71, *s*, 3 H), a MeN group (δ(H) 2.30, *s*, 3 H), and an Ac ester unit (δ(H) 2.04, *s*, 3 H). The ^{13}C-NMR spectrum suggested that this compound had one AcO group and a total of five oxygenated C-atoms (δ(C) 67.1, 71.5, 74.3, 80.5, and 86.5). The ^{1}H- and ^{13}C-NMR spectra indicated that macroyesoenline was a C_{20}-diterpenoid alkaloid with an atisine-denudatine skeleton [6]. Formation of the fragment ion *m*/*z* 390 corresponded to loss of a CH_2OH unit present as the result of the conversion of the exomethylene group at the C(16)–C(17) position, typical of a C_{20}-diterpenoid alkaloid, to a vicinal diol system, as in cochlearenine (**13**) [9] and dictysine (**14**) [11].

After assigning all non-quaternary C-atoms by H,C-COSY experiments (*Table 1*), placement and configuration of the functional groups were deduced by two- or three-bond heteronuclear correlations from HMBC experiments (*Fig. 1*) and the NOESY spectrum (*Fig. 2*) as described as follows. A signal at δ(H) 3.88–3.94 (*m*, 1 H) connected to δ(C) 80.5 (C(16)) indicated the presence of a secondary OH group at C(13). Correlations of the signal at δ(H) 4.16 (*s*, H–C(15)) with δ(C) 80.5 (C(16)), 38.5 (C(9)), and 41.7 (C(7)) were observed. Furthermore, intense interaction between this signal (δ(H) 4.16) and H–C(14) in the NOESY spectrum demonstrated that macroyesoenline contains a β-orientated OH group on C(15) as do most of the C_{20}-diterpenoid alkaloids. Correlations of the signal at δ(C) 74.3 (C(1)) with δ(H) 1.57–1.63 (H–C(3)) and the coupling constants of a H-atom (δ(H) 5.02, *dd*, $J = 10.7$, 6.3, 1 H) showed the presence of an α-AcO group at C(1). Signals at δ(H) 3.55 and 4.01 (each $J = 11.4$) with a coupling pattern of *AB* type showed that C(17) contains a primary alcohol group. The NOE cross-peak between H–C(17) and H–C(9) established the relative configuration at the C(16) position as illustrated in *Fig. 2*. Other key two- and three-bond heteronuclear correlations also substantiated the structure of **1** (*Table 1*).

Compound **2** was obtained as colorless needles and had the molecular formula $C_{23}H_{37}NO_7$, as deduced from the HR-EI-MS (*m*/*z* 439.2552 (M^+)). From the ^{1}H- and ^{13}C-NMR, DEPT, HMBC (*Table 2*), and NOESY (*Table 2*, *Fig. 3*) data, the structure of **2** was established.

The IR spectrum of **2** indicated the presence of OH groups (3320 cm^{-1}) but was devoid of absorptions attributable to CO or olefinic functionalities. The ^{1}H-NMR spectrum revealed the presence of an Et unit (δ(H) 1.04, *t*, $J = 7.3$, N–CH_2*Me*; 2.90, *q*, $J = 7.3$, N–C*H*$_2$Me) and two MeO groups (δ(H) 3.24 and 3.28), while the ^{13}C-NMR spectrum suggested that this alkaloid had seven O-bearing C-atoms (δ(C) 72.7, 77.9, 78.0, 79.0, 81.1, 82.4, and 85.6). This evidence indicated that **2** is a C_{19}-norditerpenoid alkaloid with a lycoctonine skeleton [12].

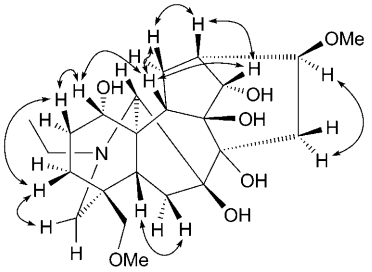

Fig. 3. *Key NOESY correlations of* **2**

The ^{13}C-NMR spectra revealed resonances for 23 C-atoms, in accordance with the molecular composition deduced from the mass spectral evidence. The ^{13}C-NMR resonances were very similar to the resonances of virescenine (**15**) [4][5][13], except for the resonances of C(8), C(9), C(10), C(12), C(14), and C(15) (*Table 2*), all this suggested that a H–C(9) group was replaced by a HO–C(9) group. Thus, the ^{13}C-NMR spectrum contained three signals for O-bearing tertiary C-atoms at δ(C) 77.9, 78.0, and 85.6. The relative configuration of HO–C(9) was deduced from the NOESY spectrum, with the observed NOEs H–C(10)/H_β–C(12), H_β–C(12)/H–C(13), H–C(13)/H_β–C(14), and H_β–C(14)/H–C(10), which indicated that the OH group is placed at C(9). In HMBC experiments, the *doublet* at δ(H) 1.39 (H_α–C(6)) correlated to δ(C) 37.6 (C(4)), 40.7 (C(5)), 78.0, and 85.6, and the signal the δ(C) 78.0 correlated to δ(H) 1.89 (H–C(10)), and was thus assigned to C(8). In addition, correlations of the signal at δ(H) 1.73 (*dd*, $J = 6.8, 15.6$, H_β–C(15)) with δ(C) 78.0 and 85.6 suggested that the signal at δ(C) 85.6 could be assigned to C(7). A *doublet* signal at δ(H) 3.85 ($J = 5.3$, H–C(14)) was coupled to a CH group at δ(H) 2.20 (*dd*, $J = 5.3, 7.8$, H–C(13)) in ^{1}H,^{1}H-COSY experiments. The remaining O-bearing tertiary C-atom was assigned through correlations of the resonance at δ(C) 77.9 (C(9)) to H–C(14) (δ(H) 3.85) and H_α–C(15) (δ(H) 3.04).

Delphinifoline (**3**) was isolated and identified from this plant for the first time. *Pelletier et al.* [10] previously reported only partial ^{13}C-NMR data for delphinifoline (**3**); here, we report the complete ^{13}C-NMR assignments (*Table 3*).

The ^{1}H-NMR spectrum revealed the presence of an Et unit (δ(H) 3.04–3.11 and 3.22–3.28, 2*m*, N–*CH*$_2$Me, 1.13, *t*, $J = 7.0$, N–CH$_2$*Me*) and two MeO units (δ(H) 3.16 and 3.34). The ^{13}C-NMR spectra indicated resonances for 23 C-atoms, in accordance with the molecular composition deduced from the mass spectral evidence. The ^{13}C-NMR spectrum suggested that this alkaloid had seven O-bearing C-atoms (δ(C) 73.3, 76.2, 78.9, 79.0, 80.7, 83.7, and 88.6) (*Table 3*). Furthermore, the ^{13}C-NMR spectrum showed two signals for O-bearing tertiary C-atoms at δ(C) 79.0 and 88.6. In COLOC experiments, the signal at δ(C) 79.0 correlated to δ(H) 2.43–2.51 (H–C(15)), 3.58 (H–C(9)), and 4.48 (H–C(14)) and was thus assigned to C(8). In addition, correlations of the signal at δ(C) 88.6 with δ(H) 2.03 (H–C(5)), 3.29 (H–C(17)), and 4.96 (H–C(6)) suggested that the signal at δ(C) 88.6 could be assigned to C(7). *Doublet* signals at δ(C) 73.3 (C(1)), 76.2 (C(14)), and 83.7 (C(16)) coupled to signals of H–C(3) (δ(H) 2.02–2.06), H–C(12) (δ(H) 2.11–2.19), and CH$_2$(15) (δ(H) 2.43–2.51 and 3.25–3.37), respectively. The remaining oxygenated C-

Table 2. *^{1}H-NMR* (400 MHz) *and ^{13}C-NMR* (100 MHz, DEPT) *Data*[a]) *for* **2**, *and ^{13}C-NMR* (100 MHz, DEPT) *Data for* **15** *in $CDCl_3$*. δ in ppm, J in Hz.

Position	**2**				**15**
	δ(C)	δ(H)	HMBC (C → H)	NOESY	δ(C)
1	72.7 (*d*)	3.54–3.58 (*m*)	H–C(3)	CH_2(2), H_β–C(3), H–C(5)	72.2 (*d*)
2	29.1 (*t*)	1.48–1.51 (*m*)		H–C(1), CH_2(3)	29.1 (*t*)[b])
3	26.7 (*t*)	1.60–1.65 (*m*), 1.82–1.86 (*m*)	CH_2(18), CH_2(19)	H–C(1), CH_2(2), H–C(19)	26.8 (*t*)[b])
4	37.6 (*s*)		CH_2(2), H–C(5), H_α–C(6), CH_2(19)		37.6 (*s*)
5	40.7 (*d*)	1.81 (*d*, J = 7.3)	H_α–C(6), H–C(17), CH_2(18), CH_2(19)	H_β–C(2), H_β–C(6), H–C(10), CH_2(18)	41.8 (*d*)
6	32.3 (*t*)	1.39 (*d*, J = 14.7, H_α) 2.82–2.93 (*m*, H_β)	H–C(17)	CH_2(18) H–C(5)	33.4 (*t*)
7	85.6 (*s*)		H–C(5), CH_2(6), CH_2(15)		86.0 (*s*)
8	78.0 (*s*)		CH_2(6), H–C(10), H–C(14), H_β–C((15)		76.1 (*s*)
9	77.9 (*s*)		H–C(13), H_α–C(15)		47.6 (*d*)
10	48.5 (*d*)	1.89 (*dd*, J = 12.7, 5.3)	H–C(1), H_α–C(12), H–C(13)	H–C(5), H_β–C(12), H–C(14)	43.4 (*d*)
11	49.7 (*s*)		H–C(2), H–C(5), H_α–C(6), H–C(10), H_α–C(12), H–C(17)		49.3 (*s*)
12	26.4 (*t*)	1.56–1.63 (*m*, H_α) 2.08–2.18 (*m*, H_β)		H–C(16), H–C(17) H–C(10)	28.4 (*t*)[b])
13	39.5 (*d*)	2.20 (*dd*, J = 7.8, 5.3)	H_α–C(15)	H–C(14)	39.5 (*d*)
14	81.1 (*d*)	3.85 (*d*, J = 5.3)	H_α–C(12), H–C(16)	H–C(10), H–C(14)	75.4 (*d*)
15	37.6 (*t*)	3.04 (*dd*, J = 15.6, 8.8, H_α) 1.73 (*dd*, J = 15.6, 6.8, H_β)		H–C(17)	35.8 (*t*)
16	82.4 (*d*)	3.38 (*t*, J = 8.3)	CH_2(12), H–C(13), H–C(14), H_β–C(15), 16-MeO	H_α–C(12), H–C(17)	81.8 (*d*)
17	64.9 (*d*)	2.65 (*s*)	H–C(5), H–C(10), H–C(19)	H_α–C(12), H_α–C(15), H–C(16), *Me*CH_2–N	64.8 (*d*)
18	79.0 (*t*)	2.99 (*AB*, J = 8.3) 3.09 (*AB*, J = 8.3)	18-MeO	H_α–C(6), H–C(5) H_α–C(6), H–C(5)	78.6 (*t*)
19	56.3 (*t*)	2.39 (*AB*, J = 11.2)	H–C(5), H_β–C(6), H–C(17), CH_2(18)	*Me*CH_2–N	55.7 (*t*)
		2.69 (*AB*, J = 11.2)		H_α–C(3), H_α–C(6), *Me*CH_2–N	
N–*CH_2*Me	50.5 (*t*)	2.90 (*q*, J = 7.3)	H–C(17), *Me*CH_2–N	H–C(17)	50.4 (*t*)
N–CH_2*Me*	13.8 (*q*)	1.04 (*t*, J = 7.3)		CH_2(19)	13.6 (*q*)
16-MeO	56.5 (*q*)	3.28 (*s*)	H–C(16)		56.2 (*q*)
18-MeO	59.4 (*q*)	3.24 (*s*)	CH_2(18)		59.3 (*q*)

[a]) Chemical shifts were assigned on the basis of ^{1}H,^{1}H-COSY, HETCOR, and HMBC experiments. [b]) These assignments are revised.

Table 3. *^{1}H-NMR* (270 MHz) *and ^{13}C-NMR* (67.5 MHz, DEPT) *Data*[a]) *for* **3** *in (D_5)Pyridine.* δ in ppm, J in Hz.

Position	**3** [10]	**3**		
	δ(C)	δ(C)	δ(H)	COLOC (C → H)
1	72.8	73.3 (*d*)	3.89–3.97 (*m*)	H–C(3)
2	28.1	30.1 (*t*)	1.68–1.76 (*m*, H_α)	
			1.86–1.92 (*m*, H_β)	
3	29.4	28.5 (*t*)	1.87–1.93 (*m*, H_α)	CH_2(19)
			2.02–2.06 (*m*, H_β)	
4	37.5	38.2 (*s*)		H–C(2), H–C(5), H–C(6), CH_2(19)
5	45.1	51.2 (*d*)	2.03 (*s*)	H–C(17), CH_2(19)
6	79.1	80.7 (*d*)	4.96 (*s*)	H–C(5), H–C(17)
7	–	88.6 (*s*)		H–C(5), H–C(6), H–C(17)
8	–	79.0 (*s*)		H–C(9), H–C(14), H–C(15)
9	45.1	46.4 (*d*)	3.58 (*t*, J = 5.3)	H–C(15)
10	39.2	45.1 (*d*)	2.07–2.11 (*m*)	H–C(13)
11	50.3	49.3 (*s*)		H–C(2), H–C(5), H–C(6), H–C(17)
12	28.7	30.8 (*t*)	2.57–2.60 (*m*, H_α)	
			2.11–2.19 (*m*, H_β)	
13	44.1	40.7 (*d*)	2.51–2.55 (*m*)	H–C(9)
14	76.4	76.2 (*d*)	4.48 (*t*, J = 4.4)	H–C(12)
15	36.6	35.3 (*t*)	2.43–2.51 (*m*), 3.25–3.37 (*m*)	
16	81.8	83.7 (*d*)	3.64 (*t*, J = 7.8)	H–C(15)
17	66.9	67.0 (*d*)	3.29 (*s*)	H–C(5), CH_2(19)
18	80.6	78.9 (*t*)	3.15 (*AB*, J = 5.3)	MeO–C(18′)
			3.36 (*AB*, J = 5.3)	
19	57.5	57.9 (*t*)	2.67 (*AB*, J = 11.4)	H–C(5), H–C(17)
			2.83 (*AB*, J = 11.4)	
N–*CH_2*Me	50.5	50.8 (*t*)	3.04–3.11 (*m*), 3.22–3.28 (*m*)	
N–CH_2*Me*	13.9	13.9 (*q*)	1.13 (*t*, J = 7.0)	
16-MeO	56.2	55.8 (*q*)	3.34 (*s*)	
18-MeO	59.6	59.0 (*q*)	3.16 (*s*)	

[a]) Chemical shifts were assigned on the basis of ^{1}H,^{1}H-COSY, HETCOR, and COLOC experiments.

atom was assigned through observation of correlations of the resonance at δ(C) 80.7 (C(6)) to H–C(5) (δ(H) 2.03) and H–C(17) (δ(H) 3.29). The remaining C-atoms were assigned through HETCOR and COLOC experiments. All C-atom assignment data are shown in *Table 3*.

Macroyesoenline (**1**) is an unusual alkaloid: to our knowledge it is only the third example after cochlearenine (**13**) [9] and dictysine (**14**) [11] of a C_{20}-diterpenoid alkaloid in which the unit usually present as an exocyclic CH_2 group has been converted to a vicinal diol. This suggests that in biological pathways, macroyesoenline could be generated from the ring-opening of epoxy precursors such as yesoxine (**16**) [14].

Experimental Part

General. All solvents were of anal. grade (*Wako Pure Chemical Industries*, Osaka, Japan). Column chromatography (CC): silica gel (SiO_2; 230–400 mesh; *Merck*). TLC: precoated SiO_2 plates;

visualization by spraying with *Dragendorff* reagent. M.p.: *Yanagimoto micromelting point* apparatus; uncorrected. Optical rotation: *Jasco Model DIP-340* polarimeter. IR Spectra: *Jasco Model FT/IR 7000* spectrometer; KBr pellets; in cm^{-1}. ^1H- and ^{13}C-NMR Spectra: *JEOL Model AL-400* and *GX-270* spectrometers; in $CDCl_3$ or (D_5)pyridine with tetramethylsilane as an internal standard; δ in ppm, J in Hz. EI-MS: *Hitachi Model M-2000* mass spectrometer; in m/z (rel. %).

Plant Material. The roots of *A. yesoense* var. *macroyesoense* (NAKAI) TAMURA (1.8 kg) were collected from Jozankei, Sapporo City, Hokkaido, Japan in August 1990 and August 1991.

Extraction and Isolation. The dried and powdered root was extracted with 90% MeOH at r.t. and evaporated to give a residue (629 g). This was dissolved in 5% HCl (*v*/*v*, 70 ml) and extracted with hexane (300 ml × 6). The acidic fraction was basified at pH 10 in the ice bath with aq. NH_3. Extraction with $CHCl_3$ (400 ml × 13) and evaporation of the extract gave a crude mixture of alkaloids (39.7 g). The hexane fraction was extracted with 5% HCl (*v*/*v*, 150 ml × 6) and the acidic fraction was basified at pH 10 in the ice bath with aq. NH_3. Extraction with $CHCl_3$ (200 ml × 10) and evaporation of the extract gave a crude mixture of alkaloids (7.1 g). The combined crude alkaloidal mixture was chromatographed on a SiO_2 (*Merck*) column and eluted with $CHCl_3$ (sat. with aq. NH_3)/MeOH mixture of increasing polarity. Fractions were collected and pooled according to their TLC pattern to give five main fractions ($A-E$). Each fraction was chromatographed, and 13 compounds were isolated: from *Fr. A:* **4** (531 mg), **5** (5440 mg), and **12** (65 mg); *Fr. B:* **6** (14 mg), **7** (1356 mg), and **11** (4316 mg); *Fr. C:* **1** (83 mg), **2** (20 mg), **8** (2056 mg), and **11** (1076 mg); *Fr. D:* **13** (8 mg), **3** (11 mg), **8** (175 mg), and **9** (1052 mg); *Fr. E:* **9** (1056 mg) and **10** (64 mg).

Macroyesoenline (=*(1α,7β,13*R*,15β)-13,15,16,17-Tetrahydroxy-4,21-dimethyl-7,20-cycloatidan-1-yl Acetate*; **1**). Colorless, amorphous solid. $[\alpha]_D^{20} = -33.8$ ($c = 0.13$, EtOH). IR (KBr): 3350 (OH), 1720 (C=O), 1240. ^1H- and ^{13}C-NMR: *Table 1*. EI-MS (70 eV): 421 (48, M^+), 404 (29, $[M-OH]^+$), 390 (11, $[M-CH_2OH]^+$), 362 (100, $[M-OCOMe]^+$). HR-EI-MS (70 eV): 421.2485 (M^+, $C_{23}H_{35}NO_6^+$; calc. 421.2463).

9-Hydroxyvirescenine (=*(1α,7β,14α,16β)-20-Ethyl-16-methoxy-4-(methoxymethyl)aconitane-1,7,8,9,14-pentol*; **2**). Colorless needles. M.p. 236–238°. $[\alpha]_D^{20} = +10.0$ ($c = 0.12$, EtOH). IR (KBr): 3320 (OH). ^1H- and ^{13}C-NMR: *Table 2*. EI-MS (70 eV): 439 (81, M^+), 424 (100, $[M-Me]^+$), 422 (74, $[M-OH]^+$). HR-EI-MS (70 eV): 439.2552 (M^+, $C_{23}H_{37}NO_7^+$; calc. 439.2567).

Delphinifoline (=*(1α,6β,7β,14α,16β)-20-Ethyl-16-methoxy-4-(methoxymethyl)aconitane-1,6,7,8,14-pentol*; **3**). Colorless needles. M.p. 238–241°. IR (KBr): 3400 (OH). ^1H- and ^{13}C-NMR: *Table 3*. EI-MS (70 eV): 439 (94, M^+), 424 (94, $[M-Me]^+$), 422 (100, $[M-OH]^+$). HR-EI-MS (70 eV): 439.2593 (M^+, $C_{23}H_{37}NO_7^+$; calc. 439.2567).

Cochlearenine (=*(1α,7β,15β)-21-Ethyl-4-methyl-7,20-cycloatidane-1,15,16,17-tetrol*; **13**). Colorless, amorphous solid. IR (KBr): 3340 (OH). ^1H-NMR (270 MHz): 0.71 (*s*, Me(18)); 1.05 (*t*, $J = 7.3$, 3 H, N–CH_2*Me*); 1.27 (*d*, $J = 9.7$, H–C(5)); 1.49–1.53 (*m*, H–C(7)); 1.82–1.90, 2.17–2.25 (2*m*, each 1 H of CH_2(2)); 1.97–2.00 (*m*, CH_2(12)); 2.09 (*d*, $J = 5.6$, H–C(13)); 2.23, 2.50 (2*d*, $J = 10.5$, each 1 H of CH_2(19)); 2.42–2.47 (*m*, N–*CH*$_2$Me); 3.49, 4.21 (2*d*, $J = 11.4$, each 1 H of CH_2(17)); 3.72 (*s*, H–C(20)); 3.84 (*dd*, $J = 10.9$, 6.6, H–C(1)); 4.03 (*s*, H–C(15)). ^{13}C-NMR (67.5 MHz): *Table 1*. EI-MS (70 eV): 377 (67, M^+), 360 (52, $[M-OH]^+$), 346 (39, $[M-CH_2OH]^+$), 186 (100). HR-EI-MS (70 eV): 377.2555 (M^+), $C_{22}H_{35}NO_4^+$; calc. 377.2563).

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