A Novel 19,21-Secohetisan Diterpenoid Alkaloid from *Aconitum tanguticum*

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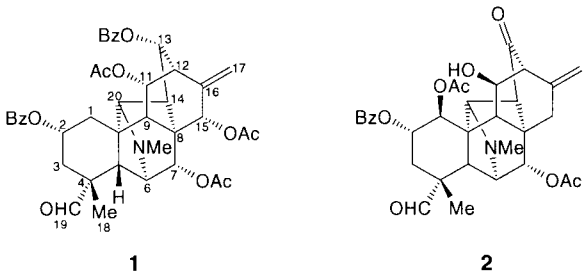
A novel 19,21-secohetisan diterpenoid alkaloid, tangutisine A (**1**), was isolated from *Aconitum tanguticum* (MAXIM.) STAPF. Its structure was determined by HR-EI-MS and 1D- and 2D-NMR spectroscopy and by comparison with the data of known 19,21-secohetisan derivatives.

Introduction. – *Aconitum tanguticum* (MAXIM.) STAPF. is a plant belonging to the family of the *Ranunculaceae*. It is widely used in traditional Tibet folk medicine as a herb to treat fever and pneumonia [1]. Previous chemical investigations of *A. tanguticum* mainly focused on the isolation and structure elucidation of diterpenoid alkaloids with the atisine and aconitine skeleton [2–5]. Further investigation of the aerial parts of *A. tanguticum* led now to the isolation of the novel 19,21-secohetisan diterpenoid alkaloid, tangutisine A (**1**) (see *Fig.*). It is noteworthy that few naturally occurring alkaloids bearing the 19,21-secohetisan skeleton were isolated until now [6–9]. Tangutisine A (**1**) is a new member of the family of barbaline, barbisine (**2**), vakoganavine, and 15-deacetylvakoganavine. We report the isolation and characterization of **1**.



1 **2**

Figure. *Structure of tangutisine A* (**1**) *and barbisine* (**2**)

Result and Discussion. – Tangutisine A (**1**) was isolated as colorless needles. Its molecular formula $C_{41}H_{43}NO_{11}$ was deduced by HR-EI-MS (M^+ at m/z 725.2836; calc. 725.2844) and its structure was determined as (2α,4α,7α,11α,13α,15α)-7,11,15-tris-

(acetyloxy)-2,13-bis(benzoyloxy)-21-methyl-19,21-secohetisan-18-al by 1D- and 2D-NMR spectroscopy and comparison with data of barbaline (**2**) [9]. It is noteworthy that tangutisine A (**1**) is the first reported 19,21-secohetisan diterpenoid alkaloid from a natural source in which the carbonyl group at C(13) is reduced to an (esterified) OH group.

The ^{1}H- and ^{13}C-NMR (*Table*) of **1** exhibited resonance for an exocyclic CH_2 group (δ(H) 5.23 and 5.36; δ(C) 117.8 and 142.9), a tertiary Me group (δ(H) 1.00; δ(C) 26.3), a MeN group (δ(H) 2.43; δ(C) 33.4), and a tertiary aldehyde function (δ(H) 9.37; δ(C) 197.8), which are consistent with similar characteristic signals of previously reported 19,21-secohetisan diterpenoid alkaloids [6–9]. The NMR data also indicated the presence of three acetate and two benzoate residues. The DEPT spectrum showed the presence of another two CH_2 groups at δ(C) 31.9 and 34.2. A CH proton at δ(H) 5.51 (H–C(2)) exhibited vicinal coupling patterns with two adjacent CH_2 protons in the ^{1}H,^{1}H-COSY plot. The lack of additional vicinal couplings towards the two CH_2 groups indicated that the C_3 fragment was connected to two quaternary C-atoms. In the HMBC spectrum, the cross-peaks between the CH proton and the quaternary C-atoms at δ 43.9 and 52.9 confirmed our deduction. The tertiary Me protons exhibited long-range coupling with the quaternary C-atom at δ 43.9, the tertiary aldehyde function, and the tertiary C-atom at δ 59.1. Thus, the aldehyde and the tertiary Me group were placed at C(4) (δ 43.9), and the other quaternary C-atom was attributed to C(10) (δ 53.9). Two C*H* signals, correlated with δ(C) 40.5 and 60.1, showed long-range couplings with δ(C) 52.9 (C(10)) in the HMBC spectrum. HMQC and HMBC experiments established that the C-atoms at δ 65.8 and 60.1 were attached to the MeN moiety. In the ^{1}H,^{1}H-COSY plot, the C*H* signals correlated to δ(C) 65.8, 40.5, 73.5, 43.6, 73.1, and 49.1 showed successive couplings. These NMR data along with HMQC and HMBC data suggested the presence of a ring system in **1** similar to that of barbaline (**2**) [9], which consists of C(5), C(6), MeN, C(20), C(14), C(13), C(12), C(11), C(9), and C(10). H–C(5) displayed a very weak vincinal coupling to H–C(6). This information indicated that H–C(6) is β-oriented. The molecular models showed that H–C(5) is axial, and the dihedral angle with H–C(6) is approaching 90°. H–C(6) only showed vincinal coupling with H–C(7), whose signals were correlated to δ(C)

Table. *^{1}H- and ^{13}C-NMR* ($CDCl_3$) *Data of Tangutisine A* (**1**). δ in ppm, *J* in Hz.

Position	δ(H)	δ(C)	Position	δ(H)	δ(C)
H_α–C(1)	2.98 (*dd*, *J* = 3.6, 15.1)	31.9	H–C(20)	3.67 (*s*)	65.8
H_β–C(1)	1.86 (*dd*, *J* = 3.6, 15.1)		MeN	2.43 (*s*)	33.4
H–C(2)	5.51 (*m*)	68.3	2-BzO:		
H_α–C(3)	2.29 (br. *d*, *J* = 15.5)	34.2	C(1)	–	128.9
H_β–C(3)	1.49 (*dd*, *J* = 3.2, 15.5)		H–C(2), H–C(6)	7.62 (*dd*, *J* = 1.1, 7.3)	129.4
C(4)	–	43.9	H–C(3), H–C(5)	7.33 (overlap)	128.3
H–C(5)	2.04 (*s*)	59.1	H–C(4)	7.49 (*m*)	132.9
H–C(6)	3.05 (*d*, *J* = 3.3)	60.1	C=O	–	165.3
H–C(7)	5.28 (*d*, *J* = 3.6)	70.5	13-BzO:		
C(8)	–	51.5	C(1)	–	129.7
H–C(9)	3.01 (br. *s*)	49.1	H–C(2), H–C(6)	7.75 (*dd*, *J* = 1.2, 7.5)	129.4
C(10)	–	52.9	H–C(3), H–C(5)	7.08 (overlap)	128.3
H–C(11)	5.57 (*dd*, *J* = 9.0, 2.6)	73.1	H–C(4)	7.33 (overlap)	133.2
H–C(12)	2.81 (*d*, *J* = 2.6)	43.6	C=O	–	166.0
H–C(13)	5.14 (*m*)	73.5	7-AcO:		
H–C(14)	2.91 (*dd*, *J* = 1.9, 9.7)	40.5	Me*C*O	–	169.4
H–C(15)	5.68 (*s*)	71.1	*Me*CO	2.17 (*s*)	21.0
C(16)	–	142.9	11-AcO:		
H_a–C(17)	5.36 (*s*)	117.8	Me*C*O	–	170.4
H_b–C(17)	5.23 (*s*)		*Me*CO	2.09 (*s*)	21.4
Me(18)	1.00 (*s*)	26.3	15-AcO:		
CH(19)=O	9.37 (*s*)	197.8	Me*C*O	–	169.9
			*Me*CO	2.07 (*s*)	21.6

71.3 (C(7)). Long-range $^1H,^{13}C$ coupling indicated that C(7) must be bound to the quaternary C(8) (δ 54.6). ^1H- and ^{13}C-NMR were in accordance with previously reported 19,21-secohetisan diterpenoid alkaloids. The CH_2=C(17) protons showed long-range coupling with C(15) (δ 71.1) and C(12) (δ 43.6) in the HMBC plot. H–C(7), H–C(11), and H–C(15) exhibited long-range coupling with the corresponding acetate C=O group, and H–C(2) and H–C(13) with the corresponding benzoate C=O group. Thus, all the substitution positions were established.

The relative configuration of **1** was determined by the 2D-NOESY plot. Based upon comparison of NMR data of **1** with those reported for 19,21-secohetisan-type diterpenoid alkaloids and upon biogenetic rationale, the β orientation is attributed to Me–C(4). The NOE for Me–C(4)/H–C(6) established the β configuration of H–C(6) and further confirmed our previous deduction. In turn, cross-peaks for H–C(6)/H–C(7) and H–C(7)/H–C(15) indicated that H–C(7) and H–C(15) are also β-oriented. In addition, NOEs for H–C(5)/H–C(9) and H–C(9)/H–C(11) established the β-orientation of H–C(11).

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Experimental Part

General. Column chromatography (CC): silica gel (200–300 mesh) from *Qingdao Marine Chemical Factory*, Qingdao, P. R. China. TLC: silica gel GF_{254} from *Qingdao Marine Chemical Factory*. M.p.: *XT-4* apparatus; uncorrected. Optical rotations: *Horiba-SEPA-300* polarimeter (*Horiba*, Tokyo, Japan). UV Spectra: *UV-210A* spectrometer; λ_{max} in nm. IR Spectra: *Bio-Win FT135* spectrophotometer; KBr pellets; $\tilde{\nu}$ in cm^{-1}. ^1H-, ^{13}C-, and 2D-NMR Spectra: *Bruker DRX-AV-500* spectrometer; δ in ppm, J in Hz. MS: *Autospec 3000* spectrometer, at 70 eV for EI; in m/z (rel. int.).

Plant Material. The aerial part of *Aconitum tanguticum* (MAXIM) STAPF. was collected at Diqing, Yunnan Province, P. R. China in June, 2001. The voucher specimen was deposited at the School of Pharmacy, Yunnan University (No. L-AT-WYB-2).

Extraction and Isolation. The air-dried and powdered aerial parts (6 kg) were extracted 3 × with EtOH. The extract was concentrated and filtered, and the filtrate was partitioned with AcOEt. The AcOEt extract (43 g) was subjected to CC (silica gel, gradient $Me_2CO/CHCl_3$): *Fractions 1–11*. *Fr. 3* (0.8 g) was further subjected to CC (silica gel, petroleum ether/Me_2CO/Et_3N 6:1:0.1; then silica gel, Et_2O/Et_3N 100:1): **1** (18 mg).

Tangutisine A (=*(2α,4α,7α,11α,13α,15α)-7,11,15-Tris(acetyloxy)-2,13-bis(benzoyloxy)-21-methyl-19,21-secohetisan-18-al*; **1**). Colorless needles. M.p. 259–261°. $[\alpha]_D^{16} = +36.84$ ($CHCl_3$, $c = 0.095$). UV: 241, 274, 304. IR (KBr): 1743, 1742, 1602, 1278, 1244, 1036, 711. EI-MS: 724(1), 697(25), 576(73), 516(13), 268(9), 146(28), 105(100), 77(45). HR-EI-MS: 725.2845 ($C_{41}H_{43}NO_{11}^+$; calc. 725.2836).

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