STRUCTURE OF EZOCHASMANINE, EZOCHASMACONITINE, ANISOEZOCHASMACONITINE AND SYNTHESIS OF VILMORRIANINE A

Hiromitsu Takayama, Miyuki Ito, Miyuki Koga, and Shin-ichiro Sakai*

Faculty of Pharmaceutical Sciences, Chiba University,
Yayoi, Chiba, 260 Japan

Toshihiko Okamoto

Faculty of Pharmaceutical Sciences, University of Tokyo
Hongo, Bunkyo-ku, Tokyo, 113 Japan

Abstract--- The structures of new diterpene alkaloids, ezochasmanine, ezochasmaconitine, and anisoezochasmaconitine, have been established as 3-hydroxychasmanine, 14-acetyl-8-benzoylchasmanine, and 14-acetyl-8-anisoylchasmanine respectively, in correlation with a known alkaloid chasmanine.

Vilmorrianine A (8-acetyl-14-anisoylezochasmanine) was synthesized from ezochasmanine via benzoylation, trichloroethoxycarbonylation, acetylation and elimination reaction of trichloroethoxycarbonyl group.

Three new minor diterpene alkaloids ezochasmanine 1, ezochasmaconitine 2, and anisoezochasmaconitine 3, together with major alkaloids pseudokobusine 4, chasmanine 5 and jesaconitine 6 were isolated from Aconitum yesoense Nakai.[1)]

We describe the structural elucidation of these minor bases 1, 2, 3, and a synthesis of vilmorrianine A which was isolated from Aconitum vilmorrianum Kom. and the structure of which was postulated by Yang Tsung-ren et al. in China.[2)]

Ezochasmanine 1 showed following properties; mp 115-118°(ether), $C_{25}H_{41}N\,O_7 \cdot \frac{1}{3}\,H_2O$, m/e 467 M^+, $[\alpha]_D^{21}$= + 40.3 ($CHCl_3$). The 100 MHz ^{1}H-NMR spectrum of 1 in $CDCl_3$ exhibited the signals due to one triplet proton on secondary alcoholic carbon-14[3)] (δ 4.10 t. J=5 Hz), four methoxyl groups (3H, s. 3.33, 3.21, 6H, s. 3.31) and a N-ethyl group (3H, t. 1.08, J=7 Hz, N-$CH_2\underline{CH_3}$). Comparison of ^{13}C-NMR spectral data of 1 with that of 5[4)] (Table) clearly revealed the site of additional hydroxy group in the former compound to chasmanine 5. The presence of a hydroxyl group at C-3 in 1 was detected by observing the downfield shift of the C-3 (37ppm) resonance in comparison with 5. The observed upfield shift of the signal due to C-19 of 1 by 6.6 ppm comparing the corresponding carbon shift of chasmanine 5[4)] can be ascribed to the deletion of 1,3-diaxial hydrogen-hydrogen interaction[5)]

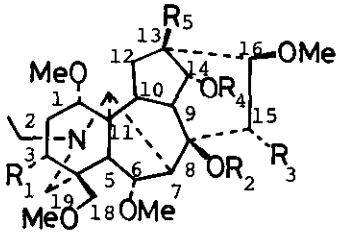
Table ^{13}C-NMR Spectra of Chasmanine 5[4] and Ezochasmanine 1 (δ_C, ppm; $CDCl_3$)

Carbon atom	5	1	Carbon atom	5	1
C-1	86.1	83.2	C-14	75.5	75.5
C-2	26.0	33.9	C-15	39.2	39.1
C-3	35.2	72.2	C-16	82.2[a]	82.0[b]
C-4	39.5	43.5	C-17	62.4	62.2
C-5	48.8	48.5	C-18	80.8	77.4
C-6	82.5[a]	82.2[b]	C-19	54.0	47.4
C-7	52.8	52.4	N-CH_2	49.3	49.1
C-8	72.6	72.5	CH_3	13.6	13.7
C-9	50.3	48.8	OCH_3 1′	56.3	56.4
C-10	38.4	38.1	6′	57.2	57.3
C-11	50.4	50.2	16′	55.9	56.0
C-12	28.6	28.1	18′	59.2	59.2
C-13	45.7	45.3			

a - b) Assignments bearing the same superscript may be interchanged.

between C-3-H and C-19-H and thus the presence of C-3-α-hydroxyl group in 1 was demonstrated. Acetylation of 1 was carried out in Ac_2O with pyridine at 3° for over night to yield the mono acetate 7, ($C_{27}H_{43}N\ O_8$ m/e 509 M^+, mp 58-62°, hygroscopic) and the diacetate 8, ($C_{29}H_{45}N\ O_9$ m/e 551 M^+, amorphous). More vigorous acetylation using Ac_2O, p-TsOH at 80-90°, however, afforded the triacetate 9, ($C_{31}H_{47}N\ O_{10}$ m/e 593 M^+, mp 221-224°). Additional evidence regarding the sterochemistry of α-hydroxyl group at C-3 can be obtained from ^{1}H-NMR of mono acetate 7.

The signals at 4.88ppm (d.d., J_1= 6, J_2= 10Hz) revealed an axial conformation of β-hydrogen group at C-3. The above observations lend credence to structure 1 for ezochasmanine. Chasmanine 5 can be visually derived from ezochasmanine 1 by elimination of the 3-α-hydroxyl group. The following is the transformation of ezochasmanine 1 to chasmanine 5 for the structural proof of 1. Mono benzoylation of 1 using 1.7 equivalents of benzoyl chloride in pyridine at ~18° for 2hr resulted in formation of unexpected 14-benzylezochasmanine 10 ($C_{32}H_{45}N\ O_8$ m/e 571 M^+, amorphous, δ 5.11, 1H, t, J= 5Hz, C-14-H) in 68% yield. For protecting the 3-hydroxyl group, using a lage excess of the β-trichloroethoxycarbonylchloride[6] in pyridine, 10 could be converted in 83% yield to the derivative 11 ($C_{35}H_{46}N\ O_{10}Cl_3$ m/e 745 M^+, 747 M^++ 2, amorphous). The remained tertiary hydroxyl group was acetylated with Ac_2O and p-TsOH at 85-92° for 2hr. The resulting triacyl derivative 12 was treated with Zn powder in HOAc at room temperature (5hr) to remove the protecting group at 3-position. The final product is 8-acetyl-14-benzoylezochasmanine 13 [prism, mp 113-114° (AcOEt-hexane), toxic, $C_{34}H_{47}N\ O_9$ m/e (%) 613 M^+(3), 582 M^+ - OCH_3(62), 553 M^+- HOAc (56), 105(100), δ 5.02 (1H, t. J= 5Hz, C-14-β-H), 1.36 (3H,



1 R_1= OH, R_2=R_3=R_4=R_5= H Ezochasmanine

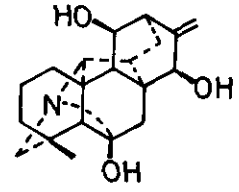
2 R_1= H, R_2=-COC$_6$H$_5$, R_3=R_5= H, R_4=-COCH$_3$
Ezochasmaconitine

3 R_1= H, R_2=-COC$_6$H$_4$OMe(p), R_3=R_5= H, R_4=-COCH$_3$
Anisoezochasmaconitine

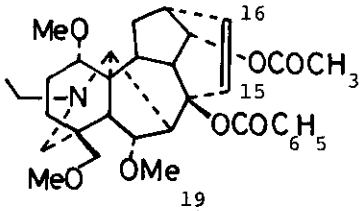
5 R_1=R_2=R_3=R_4=R_5= H Chasmanine

6 R_1=R_3=R_5= OH, R_2=-COCH$_3$, R_4=-COC$_6$H$_4$OMe(p)
Jesaconitine

18 R_1=R_2=R_3=R_5= H, R_4=-COCH$_3$



4 Pseudokobusine



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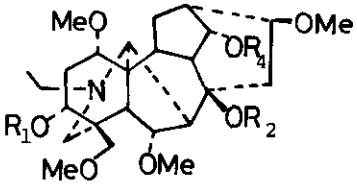


7 R_1=-COCH$_3$, R_2=R_4= H

8 R_1=R_4=-COCH$_3$, R_2= H

9 R_1=R_2=R_4=-COCH$_3$

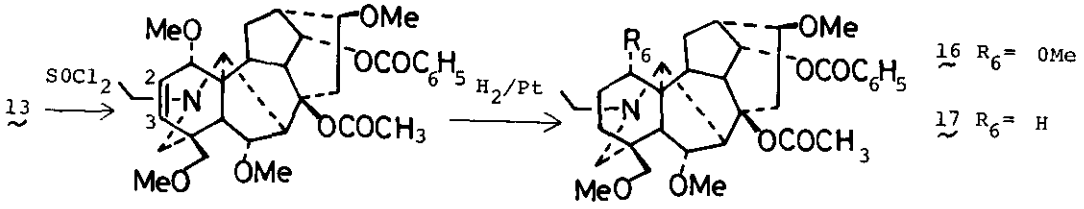
10 R_1=R_2= H, R_4=-COC$_6$H$_5$

11 R_1=-COOCH$_2$CCl$_3$, R_2= H, R_4=-COC$_6$H$_5$

12 R_1=-COOCH$_2$CCl$_3$, R_2=-COCH$_3$, R_4=-COC$_6$H$_5$

13 R_1= H, R_2=-COCH$_3$, R_4=-COC$_6$H$_5$

14 R_1= H, R_2=-COCH$_3$, R_4=-COC$_6$H$_4$OMe(p)
Vilmorrianine A



15

16 R_6= OMe

17 R_6= H

16 $\xrightarrow{OH^-}$ Chasmanine 5

s. $-OCOCH_3$)]. 8-Acetyl-14-anisoylezochasmanine 14 [prism, mp 169° (AcOEt-hexane), toxic, CD (MeOH) $\Delta\varepsilon$ (nm); +1.9(256), +3.9(216), $[\alpha]_D^{25}$ = + 24.1 ($CHCl_3$)] was obtained by the same method. Alkaloid 14 and vilmorrianine A were identified by comparison of mixed melting point, IR and CD spectra. Refluxing of 8-acetyl-14-benzoylezochasmanine 13 (= 13,15-dideoxyaconitine) in excess thionyl chloride[7] gave the dehydrated product 15 [= $\Delta^{2,3}$-3,13,15-trideoxyaconitine, amorphous, $C_{34}H_{45}N\ O_8$ m/e 595 M^+, δ 6.04 (1H, d.d. J_1= 10, J_2= 4Hz, C-2-H), 5.79 (1H, d. J= 10Hz, C-3-H)] in 71% yield after the purification by Al_2O_3 column chromatography. After catalytic reduction using PtO_2 as catalyst, followed by separation using preparative tlc (Et_2O -saturated 25% aq-NH_4OH), 13 gave rise to two reduced products 16 [in 59% yield, mp 148.5-151°, CD(MeOH)$\Delta\varepsilon$(nm) +0.9(272), +6.4(228)] and 17 [in 25% yield, mp 174-178°, $C_{33}H_{45}N\ O_8$ m/e 567.3198 M^+]. ^{1}H-NMR spectrum of the latter compound 17 showed signals of three OCH_3 (3,35, 3.28, 3.17 each 3H, s.), one acetyl group (1.41 3H, s.) and C-14-H (5.06 1H, t. J= 4.5Hz). From above observations, the structure 17 (1-demethoxy-3,13,15-trideoxyaconitine[7]) was ascribed to the latter reduced compound. The first reduced compound 16 was found identical with known 8-acetyl-14-benzoylchasmanine[8] by comparison of mixture melting point, IR, CD and ^{1}H-NMR spectra. Finally, 16 was hydrolyzed in alkaline solution to form chasmanine 5.

Ezochasmaconitine 2 showed following properties; mp 163-165° prism (acetone), $C_{34}H_{47}N\ O_8$ m/e 597 M^+, CD(MeOH), $\Delta\varepsilon$ (nm) +1.2(235), +1.1(210). The 100 MHz ^{1}H-NMR spectrum of 2 in $CDCl_3$ exhibited several signals due to five aromatic protons (7.45-8.00), four methoxyl groups 2.94(3H,s.), 3.24(6H,s.), 3.32(3H,s.) and one acetyl group 1.76(3H,s.) and one proton (4.82 t. J= 4.5Hz) arising on acetylated secondary alcoholic carbon at C-14. On the basis of these data, we assumed a structure of 8-benzoyl-14-acetylchasmanine 2 for ezochasmaconitine. Then derivation of 5 to 2 was carried out as follows. Acetylation of 5 by use of F_3CCOOH and HOAc provided 14-acetylchasmanine 18 [amorphous, $C_{27}H_{43}N\ O_7$ m/e 493 M^+, δ 4.79 (1H, t.J= 5Hz, C-14-H), 2.02 (3H,s. C-14-$OCOCH_3$)] in 63% yield. Treatment of 18 with benzoic acid anhydride (10 eq.) in the presence of p-TsOH·H_2O (1eq.) in boiling toluene for 12hr gave the 8-benzoyl-14-acetylchasmanine 2 [mp 166°, m/e 597 M^+, $[\alpha]_D^{19}$= +26.1 ($CHCl_3$)] in 20% yield and 8-benzoyl-14-acetyl-$\Delta^{15,16}$-16-demethoxychasmanine 19 [mp 147.5-149°, $C_{33}H_{43}N\ O_7$ m/e 565 M^+, C-16-olefinic proton (6.06, 1H, d.d. J_1=7, J_2=10Hz), C-15-olefinic proton (6.67, 1H, d. J=10Hz)] in 39% yield

after the purification by preparative tlc. Identity of ezochasmanine and the synthetic alkaloid 2 was established by comparison of mixture melting point, IR, ^{1}H-NMR and CD spectra.

Anisoezochasmaconitine 3 showed following properties; mp 136-138.5° (ether), $C_{35}H_{49}N\,O_9$ m/e 627 M^+, CD(MeOH)$\Delta\varepsilon$ (nm) +0.95(256), +3.5(213), UVλ_{max}^{EtOH} 259nm. The 100 MHz ^{1}H-NMR spectrum of 3 in $CDCl_3$ showed the signals arising from p-anisoyl group [δ 7.93 (2H,d. J= 9Hz), 6.86 (2H,d. J= 9Hz) aromatic protons, 3.84 (3H, s. $OC\underline{H}_3$)] and the signals of C-14-H (4.80, 1H, t. J=4.5Hz), C-14-$OCOC\underline{H}_3$ (1.78, 3H, s.)]. 8-Anisoyl-14-acetylchasmanine [mp 140°(ether), m/e 627 M^+, $[\alpha]_D^{18}$ = +14.1 ($CHCl_3$), UVλ_{max}^{EtOH} 259nm (logε = 4.29)] was synthesized from chasmanine 5 (in 33% over all yield) under the same manner described above and identified with the natural anisoezochasmaconitine 3 by comparison of mixture melting point and other physical data of them. Ezochasmaconitine 2 and anisoezochasmaconitine 3 are the first examples of the diterpene alkaloids possessing the aroyl groups at C-8 and acetyl group at C-14. All of the formerly known congeners have the aroyl groups at C-14 and acetyl group at C-8. Mild toxicity was observed for the two new bases, 2 and 3.

Structure of chasmanine 5 was confirmed by X-ray crystallographytic study[9] of the 14-benzoylchasmanine hydrochloride salt and total synthesis[10], and therefore, structures of the titled alkaloids were also established by correlation with 5.

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REFERENCES

1. S.Sakai et al., Presented in part at the 100th Annual Meeting of the Pharmaceutical Society of Japan, Tokyo, April 1980. Abstracts, p 224.
2. Yang Tsung-ren, Hao Xiao-jiang, Chow Jun; Acta Botanica Yunnanica 1 41 (1979).
3. S.W.Pelletier, Z.Djarmati, S.Lajsic, W.H.Decamp; J.Am.Chem.Soc. 98 2617(1976)
4. S.W.Pelletier, Z.Djarmati; J.Am.Chem.Soc., 98 2626 (1976).
5. H.Beierbeck, J.K.Saunders; Can.J.Chem., 54 2985 (1976).
6. T.B.Windholz, D.B.R.Johnston; Tetrahedron Letters; 2555 (1967).
7. R.E.Gilman, L.Marion; Can.J.Chem. 40 1713 (1962).
   R.E.Gilman, L.Marion; Tetrahedron Letters; 923 (1962).
   Y.Tsuda, L.Marion; Can.J.Chem. 41 3055 (1963).
8. O.Achmatowicz.Jr., Y.Tsuda, L.Marion, T.Okamoto, N.Natsume, H.Chang, K.Kajima; Can.J.Chem. 43 825 (1965).
9. S.W.Pelletier, W.H.DeCamp, Z.Djarmati; J.Chem.Soc., Chem.Comm., 253 (1976).
10. T.Y.R.Tsai, C.S.J.Tsai, W.W.Sy, M.N.Shanbhag, W.C.Liu, S.F.Lee, K.Wiesner; Heterocycles, 7 217 (1977).
    K.Wiesner, T.Y.R.Tsai, K.P.Nambiar; Can.J.Chem., 56 1451 (1978).

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