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ALKALOIDS OF *Aconitum kirinense* THE STRUCTURE OF AKIRAMINE

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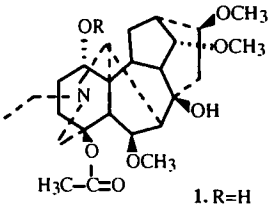
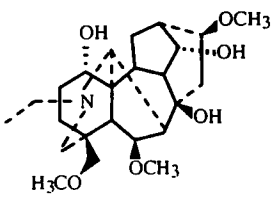
The known alkaloid tuguaconitine and a new base, which has been called akiramine, have been isolated from the epigeal part of Aconitum kirinense *Nakai. The structure of akiramine as 4β-acetoxy-1α,8β-dihydroxy-6β,14α,16β-trimethoxy-N-ethylaconitine has been established by a study of its spectral characteristics.*

Aconitum kirinense Nakai is a perennial herbaceous plant growing in Russia, Japan, and China. In Russia, the plant is found in the Far East in oak woods, on dry slopes, and in rock crevices [1]. We have previously [2—6] described the isolation from the epigeal part of this plant gathered in Primorskii krai (Maritime Territory) in the fruit-bearing period of the known diterpene alkaloids excelsine and lepenine and the new bases 8-acetylexcelsine, akiran, akiranine, akirine, and lepenine N-oxide. Continuing the study of the alkaloid composition of *Aconitum kirinense*, we have isolated two bases. One of them, with the composition $C_{23}H_{39}NO_7$, mp 197—199°C, was identified by a comparison of spectral characteristics and physicochemical constants as tuguaconitine [7]. The second proved to be new and has been called akiramine (**1**).

Akiramine (**1**) has the composition $C_{23}H_{39}NO_7$, mp 162—164°C (acetone). The IR spectrum of the alkaloid has absorption bands of hydroxy and ester groups and ether bonds. According to its PMR spectrum, the base contains a N-ethyl group, three methoxy groups, and an acetoxy group. Its mass spectrum showed that akiramine belonged to the group of alkaloids with a lycoctonine skeleton and was close to that of akiran (**2**), which contains an acetoxy group at C-4. In its functional composition akaramine differs from akiran by the presence of a hydroxy group in place of one of the methoxy groups of akiran.



1. R=H
2. R=CH_3



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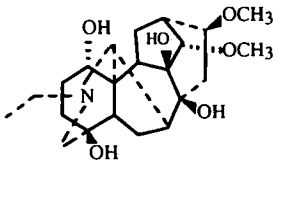

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In the PMR spectrum of akiramine the signals of a number of methine protons showing the positions of the oxygen substituents were observed. The presence of an α-methoxy group at C-14 was confirmed by the signal of the geminal proton (3.55 ppm, 1H, t, J = 5 Hz). The presence of an α-hydroxy group at C-1 followed from the observation of the signal of the hemihydroxylic proton (3.72 ppm, 1H, t, J = 2.5 Hz). The second methoxy group in akiramine is located at C-6 and has the β-configuration since the PMR spectrum contains the signal of a proton geminal to it (4.02 ppm, 1H, d, J = 7 Hz). The above-described assignments of the substituents and positioning of the other functional groups were elucidated in an analysis of the ^{13}C NMR spectrum of akiramine. The multiplicities of the signals were determined by using the DEPT technique.

The ^{13}C NMR spectrum of akiramine revealed the signals of the 25 carbon atoms the multiplicities of which are given in Table 1.

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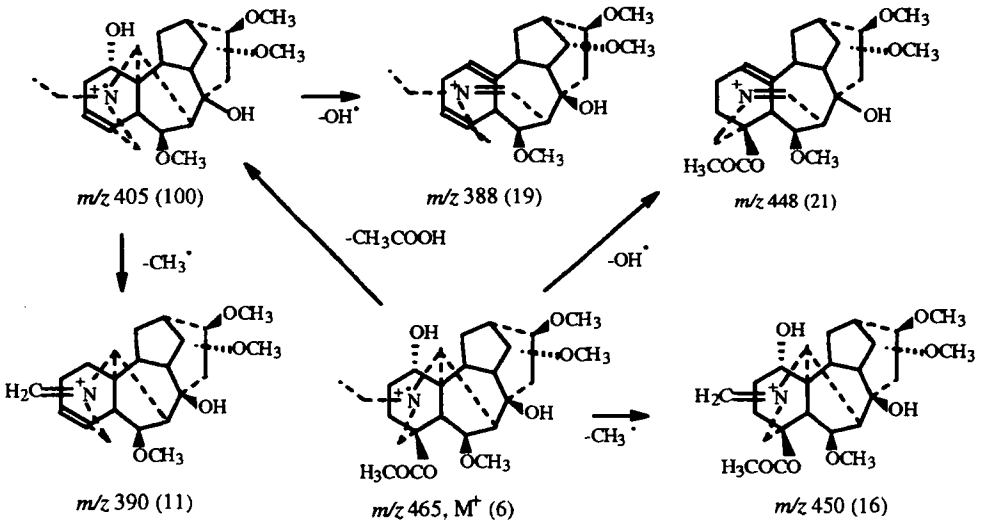
TABLE 1. Chemical Shifts of the Carbon Atoms in the ^{13}C NMR Spectra of Akiramine (1), Subcusine (3), and Lappaconidine (4)

Carbon atom	Compound			Carbon atom	Compound		
	1	3	4		1	3	4
1	71.9	72.4	72.5	14	81.8	76.0	90.4
2	29.5	27.3	29.8	15	39.9	40.6	45.1
3	30.4	30.0	33.5	16	83.0	81.5	83.6
4	79.9	37.0	70.7	17	64.0	65.2	63.1
5	49.0	44.2	48.2	18	-	77.7	-
6	84.5	82.4	27.4	19	59.1	57.6	60.4
7	51.4	50.8	47.0	N-CH_2 |	48.0	48.5	46.5
8	75.0	75.1	76.3	CH_3	12.7	12.9	13.1
9	44.1	46.1	77.6	6'	57.6	57.1	-
10	44.0	45.4	36.3	14'	57.7	-	58.1
11	49.6	48.3	50.4	16'	56.2	56.3	56.3
				18'	-	59.1	-
12	30.5	29.3	23.1	O=C |	169.6	-	-
13	37.6	39.8	48.4	CH_3	21.9	-	-



Scheme 1. Main directions of the mass-spectrometric fragmentation of akiramine (1).

The assignment of the signals of the carbon atoms was made by a comparison with the spectra of subcusine (3) [8] and lappaconidine (4) [9]. The akiramine molecule contains four quaternary carbon atoms. One of them was revealed at 169.6 ppm and corresponds to the carbonyl carbon of an acetoxy group, while the second was shown at 79.9 ppm and can be assigned to the C-4 carbon atom linked with the acetoxy group. The third singlet signal was found at 75.0 ppm and corresponds to the C-8 carbon atom, and, finally, the fourth was detected at 49.6 ppm and corresponds to C-11. The third methoxy group in akiramine is located at C-16 and has the β-orientation, as was confirmed by a doublet signal at 83.0 ppm. Consequently, akiramine has the structure (1).

Possible directions of the mass-spectrometric fragmentation of akiramine are given in Scheme 1 and agree well with

the proposed structure. The ion with the maximum intensity in the mass spectrum of (1) results from the splitting out of an acetic acid molecule at the expense of the acetoxy group at C-4. Similar fragmentation is observed in the spectra of other alkaloids containing an acyl residue at C-4 [10]. The further breakdown of the molecule is connected with the splitting out of methyl and hydroxyl radicals with the participation of the N-ethyl group and the hydroxy group at C-1. In addition to the above-described main direction of the mass-spectrometric fragmentation of akiramine, its mass spectrum showed the peaks of ions connected with the ejection of hydroxyl and methyl radicals directly from the molecular ion.

EXPERIMENTAL

Melting points are uncorrected. Mass spectra were taken on a MKh 1310 spectrometer with a system for the direct injection of the sample into the ion source, PMR and ^{13}C NMR spectra on a UNITY 400 Plus instrument (Varian) in deuterochloroform with HMDS as internal standard, and IR spectra on a Perkin-Elmer Model 2000 Fourier IR spectrometer in tablets with KBr.

For chromatography we used type LS silica gel (Czech republic). The individuality of the substances was checked by TLC using silica gel and alumina as sorbents in the systems: 1) chloroform—methanol (100:1) and 10:1); and 2) (benzene—methanol (4:1).

Separation of the Total Alkaloids. The mother solutions from the hexane fraction of the total mixture of *Aconitum kirinense* alkaloids [5] were rechromatographed on a silica gel column, and elution with benzene—methanol (50:1) permitted the isolation of 0.12 g of akiramine and 0.07 g of tuguaconitine.

Tuguaconitine. mp 197—199°C (acetone—ether). IR spectrum (ν, cm^{-1}): 3515, 3440, 2960, 2950, 1470, 1390, 1230, 1110, 880, 750, 710.

PMR spectrum (δ, ppm): 1.03 (3H, t, J = 7.0 Hz, N-CH_2CH_3), 3.42, 3.41, 3.36 (each 3H, s, 3×OCH_3), 3.54 (1H, t, J = 5 Hz, H-14β), 3.80 (1H, s), 4.30 (1H, s).

Mass spectrum, m/z (%): 437 (80), 422 (100), 406 (36), 394 (18), 311 (5), 268 (25).

Akiramine (1). IR spectrum (ν, cm^{-1}): 3586, 3480, 2936, 2351, 1730, 1683, 1652, 1634, 1558, 1539, 1471, 1456, 1409, 1245, 1230, 1145, 1100, 1082, 1020, 982, 856, 697, 583, 498.

PMR spectrum (δ, ppm): 1.06 (3H, t, J = 7 Hz, N-CH_2CH_3), 1.97 (3H, s, $OCOCH_3$), 3.28, 3.29, 3,35 (each 3H, s, 3×OCH_3), 3.55 (1H, t, J = 5 Hz, H-14β), 3.66 (1H, t, J = 2.5 Hz), 3.72 (1H, t, J = 2.5 Hz, H-1β), 4.02 (1H, d, J = 7 Hz, H-6α), 4.29 (1H, s).

Details of the ^{13}C NMR spectrum of akiramine are given in Table 1, and of the mass spectrum in Scheme 1.

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