ALKALOIDS OF Aconitum monticola

STRUCTURE OF ACOMONINE

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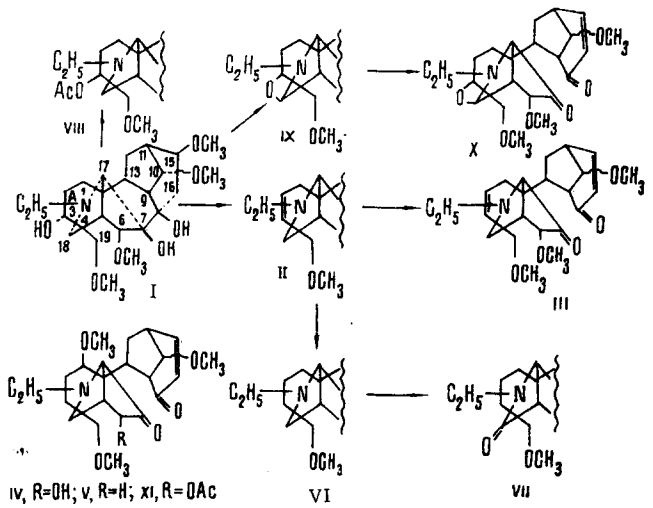
We have investigated the alkaloids from the roots of a plant which has not been studied previously, Aconitum monticola, collected in the Dzhungarian Ala-Tau (Kuyandysai) in the budding–flowering stage. Together with the known alkaloids songorine, songoramine, and norsongorine, from the combined alkaloids (4.1% of the weighed dry raw material) we have isolated with an amorphous base with the composition $C_{22}H_{35}NO_6$, a base $C_{22}H_{33}NO_6$ with mp 166-167° C, a base $C_{22}H_{33}NO_5$ with mp 161-164° C, and a new alkaloid $C_{25}H_{41}NO_7$ with mp 208-210° C (hydrochloride with mp 206-207° C) which we have called acomonine (I) [1].

According to its IR spectrum, acomonine contains hydroxy groups. On deuteration it was established that compound (I) contains three active hydrogen atoms. The NMR spectrum has the signals from a N-ethyl group and four methoxy groups [three-proton triplet at 1.13 ppm; three singlets (total 12H) at 3.27, 3.29, and 3.35 ppm]. Consequently, the expanded formula of acomonine is $C_{19}H_{21}(N-C_2H_5)(OH)_3(OCH_3)_4$. The results of a comparison of the empirical and expanded formulas of (I) show that its skeleton consists of six rings.

With p-toluenesulfonyl chloride acomonine gives anhydroacomonine, $C_{25}H_{39}NO_6$ (II), the oxidation of which with HIO_4 forms anhydrosecodesmethanolacomonine, $C_{24}H_{33}NO_5$ (III). The mass spectrum of (III) is similar to those of demethylenesecodesmethanoldelcorine (IV) and demethylenesecodesmethanoldeoxydelcorine (V) [2].

Anhydroacomonine was hydrogenated by the Adams method. The resulting deoxyacomonine $C_{25}H_{41}NO_6$ (VI) was oxidized with potassium permanganate. A substance $C_{25}H_{39}NO_7$ (VII) was obtained which contained a lactam carbonyl in a six-membered ring. In the mass spectrum of (VII) the intensity of the peak of the ion M−15 had risen sharply, as has also been observed on passing from the lycoctonine alkaloids to their 18-oxo derivatives [3]. On the basis of the facts given, it may be concluded that acomonine is based on the lycoctonine skeleton.



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The acetylation of acomonine with acetic anhydride in pyridine gave the monoacetate $C_{27}H_{43}NO_8$ (VIII). Its IR spectrum showed the absorption band of an ester carbonyl group, and in the mass spectrum the maximum peak was that of the ion M − 59. The NMR spectrum of (VIII) showed signals from one acetyl group and from the proton geminal to it. Thus, one of the hydroxy groups is secondary and the other two are, as shown below, tertiary, forming a α-glycol system.

Anhydroacomonine (II) is formed by the dehydration of the secondary hydroxyl, which explains the retention in product (II) of the glycol grouping at C_7-C_8 and also the presence in the NMR spectrum of compound (II) of one one-proton signals of two olefinic protons present in a six-membered ring at 5.32 ppm (doublet, J = 10 Hz) and 5.86 ppm (multiplet) [4].

According to the nature of the splitting of these signals the ethylene grouping is located, on the one hand, in the neighborhood of a quaternary carbon atom and, on the other hand, in the neighborhood of a methylene group. In the lycoctonine skeleton, such a grouping can only be in ring A. From a consideration of the mass spectrum of acomonine − M^+ 467 (22%), M − 15 (100%), M − 17 (44%), M − 18 (5%), M − 31 (61%), M − 33 (53%), M − 43 (13%), M − 45 (15%), M − 61 (10%), M − 71 (5%), M − 87 (5%) − the secondary hydroxy group can be located at C_1 in the β-orientation [5]. However, this is contradicted by the results of the oxidation of acomonine with potassium permanganate. As a rule, under these conditions lycoctonine alkaloids with a β-oriented hydroxy group at C_1 form 18-oxo derivatives, while acomonine is oxidized to anhydrohydroxyacomonine $C_{25}H_{39}NO_7$ (IX), which is an internal ether of an α-carbinolamine. Substance (IX) retains the α-glycol system, which is oxidized by periodic acid to anhydrohydroxysecodesmethanolacomonine $C_{24}H_{33}NO_6$ (X) and is reduced by sodium tetrahydroborate to acomonine.

The NMR spectrum of (IX) has a one-proton singlet due to the proton of a −O−CH−N< grouping at 4.48 ppm. The hydroxy group participating in the formation of this grouping has the α-orientation. The C_1 position for it is excluded on the basis of the mass spectrum of acomonine, since in such cases the peak of the ion due to the ejection of a substituent from this position is the maximum peak and considerably exceeds all the other peaks [3]. We are left with C_3 as the possible position for the hydroxy group. This is confirmed by the NMR spectrum of acetylacomonine (VIII) in which the signal of the geminal proton at 4.7 ppm appears in the form of a quadruplet with J_1 = 7 Hz adn J_2 = 10 Hz. Finally, if the hydroxy group under consideration were located at C_2, the appearance fo a multiplet in the NMR spectrum could be expected. The observed coupling constants with the α-orientation of the C_3-hydroxy group agree well with the conformation of ring A in the distorted chair form.

The NMR spectra of acomonine and some of its derivatives contain one-proton triplets at ~3.6 ppm with J = 5 Hz corresponding in each case to a β-proton at C_{10} geminal to a methoxy group. The appearance of this signal shows the absence of substituents at C_9 and C_{11} [6].

The IR spectrum of anhydrosecodesmethanolacomonine (III) has the absorption bands of ketone groups in six-membered and five-membered rings at 1655 and 1735 cm^{-1}, respectively. The band of the ketone in a six-membered ring is displaced, which is due to its conjugation with a double bond. The UV spectrum of (III) is characteristic for α,β-unsaturated ketones (λ_{max} 232 nm, log ε 3.87). This is also shown by the NMR spectrum of (III) which contains the signals from four olefinic protons one of which is located in the weak field at 6.9 ppm (quartet).

Possible Positions for the α-Glycol Grouping − 7,8 and 8,9. It was mentioned above that position 9 is not substituted; furthermore, the mass spectrum of (III) is characteristic for the spectra of the seco products formed from a diol system at C_7 and C_8 and is similar to those of the corresponding products from delcorine and deoxydelcorine [2, 7]. In the oxidation of anhydroacomonine (II), in addition to the cleavage of the diol system the elimination of a molecule of methanol takes place. Such a phenomenon indicates the presence of a methoxy group in the β-position to a carbonyl group. And since this carbonyl group, according to the IR spectrum, is located in a six-membered ring, the second methoxy group must be present at C_{15}. The extreme ease of elimination of the methoxy group is apparently connected with its β (axial) orientation [6].

The results of a study of the mass spectra of the seco products of a series of lycoctonine alkaloids proved to be extremely useful in establishing the position of the following methoxy group. A distinguishing feature of these spectra is the presence of a strong peak in the region of low mass numbers. Thus, in the spectrum of demethylenesecodesmethanoldelcorine (IV) the peak of an ion with m/e 100 is observed, while in the spectrum of the deutero analog it is shifted by one unit (m/e 101), and in the spectrum of the acetate of demethylenesecodesmethanoldelcorine (XI) the peak of an ion with m/e 142 corresponds to this fragment.

In view of the fact that in demethylenesecodesmethanoldelcorine there is only one hydroxyl at C_6, it is clear that the substituent at C_6 takes part in the formation of this fragment. In the mass spectra of the corresponding seco products, which we obtained from lycoctonine and browniine, containing a methoxy group at C_6, there are the peaks of an ion with m/e 114. In the mass spectrum of (III) there is a strong peak of an ion with m/e 114, i.e., there is a methoxy group at C_6 in acomonine. The oxidation of acomonine by potassium permanganate forms anhydrohydroxyacomonine, and deoxyacomonine forms an 18-oxo derivative, and therefore the β-orientation follows for the C_6-methoxy group [8].

Taking into account the presence of a lycoctine skeleton in (I), the locations in it of all the substituents mentioned, and the absence of $\rangle N-CH_3$, $-C-CH_3$, $-CH_2OH$ and $\rangle NH$ groups, the remaining methoxy group must be located at C_{19}.

Thus, acomonine has structure (I). We may note that acomonine is the first lycoctonine alkaloid having no substituent at C_1.

EXPERIMENTAL METHOD

The homogeneity of the substances was checked by chromatography in a thin layer of type KSK silica gel in the benzene–methanol (4 : 1) system. The NMR spectra were taken in deuterochloroform on a JNM-4H-100/100 MHz instrument with HMDS as internal standard (values given in the δ scale); the mass spectra were taken on an MKh-1303 instrument fitted with a system for the direct introduction of the sample into the ion source; and the IR spectra were taken on UR-20 instrument (tablets with KBr).

The Isolation of the Alkaloids. The comminuted air-dry roots of Aconitum monticola (90 kg) were wetted with 5% sodium carbonate solution, and the alkaloids were exhaustively extracted with chloroform at room temperature. The extracts were treated once with 10% sulfuric acid and then repeatedly with 5% sulfuric acid. On standing in the refrigerator, the 10% sulfuric acid solution deposited 100 g of a crystalline sulfate. This was treated with 5% sodium carbonate solution and the alkaloids were extracted with ether and chloroform (A). The ether-soluble fraction, on treatment with acetone, yielded 59 g of songorine with mp 190-195° C, and the evaporated mother solution yielded 28 g of songorine hydrochloride with mp 248-250° C. After the hydrochloride had been separated off, the solution was evaporated, the residue was dissolved in water, and after the addition of sodium carbonate to an alkaline reaction the bases were extracted successively with hexane, ether, and chloroform. The hexane fraction was separated on a column of alumina, and elution with hexane–ether yielded 0.08 g of songoramine with mp 215-217° C. The alkaloids were extracted from the chloroform extract A with 5% sulfuric acid in portions (8 × 2 ml). The acid fractions were made alkaline, and the bases were extracted with ether. From them in methanol was obtained 0.38 g of a hydrochloride with mp 212-214° C, which was converted into an amorphous base with the composition $C_{22}H_{35}NO_6$, mol. wt. 409. The aqueous solution after the treatment of the crystalline sulfate with 5% sodium carbonate solution and extraction of the alkaloids with ether and chloroform (A) was evaporated to dryness and the residue was repeatedly extracted with chloroform at the boil. The solvent was evaporated off and from the residue was isolated 0.13 g of norsongorine with mp 304-305° C (methanol). After the separation of the crystalline sulfate, the 10% acid solution was combined with the 5% acid solution, the mixture was made alkaline with sodium carbonate with cooling, and the alkaloids were extracted with chloroform. This gave 3.6 kg of combined chloroform-soluble alkaloids. The total amount of alkaloids was 4.1% of the weight of the air-dry roots. The combined chloroform-soluble alkaloids were dissolved in 5% sulfuric acid, the solution was made alkaline with sodium carbonate, and the alkaloids were extracted first with ether and then with chloroform.

When 2.9 kg of the combined ether-soluble alkaloids was treated with acetone, 92.7 g of acomonine separated out with mp 208-210° C (acetone–methanol). The solvent was evaporated off from the mother solution and in ethanol 443 g of a mixture of hydrochlorides with mp 150-155° C was obtained. On a thin-layer chromatogram, this mixture showed two spots with R_f 0.3 and 0.8. Part of this mixture (28.8 g) was recrystallized from methanol and then from ethanol. This gave 9.31 g of songorine hydrochloride with mp 248-250° C. The combined mother solutions after recrystallization were evaporated to two thirds, and 2 g of a hydrochloride with mp 125-128° C (main spot with R_f 0.8) separated out. The hydrochloride was dissolved in water, the solution was made alkaline with sodium carbonate, and the bases were separated according to their solubilities into petroleum ether, diethyl ether, and chloroform fractions. By means of acetone, 0.5 g of a base with the composition $C_{22}H_{23}NO_6$, mp 166-167° C (from acetone), mol. wt 407, was obtained from the petroleum ether and diethyl ether fractions. The solvent was evaporated off and the resi-

due was treated with ether. By means of a mixture of ether and acetone, 0.2 g of a base with the composition $C_{22}H_{33}NO_5$, mp 161-164° C (ether–acetone), mol. wt. 391, was obtained.

Acomonine (I). After recrystallization from a mixture of acetone and methanol and drying in vacuum, substance (I) melted at 208-210° C. Found %; C 64.20; 64.30; H 9.20; 8.95; N 2.89; 2.82. $C_{25}H_{41}NO_7$. Calculated %: C 64.24; H 8.78; N 2.99.

Anhydroacomonine (II). To a solution of 0.2 g of acomonine in 20 ml of pyridine cooled to 0° C was added 0.24 g of p-toluenesulfonyl chloride. The mixture was left at 0° C for 18 h. Then the solvent was evaporated off and the residue was dissolved in 2% sulfuric acid, the solution was washed with benzene and was made alkaline with sodium carbonate with cooling and the reaction product was extracted with petroleum ether. On evaporation it crystallized. In this way, 0.08 g of substance (II) was obtained with mp 133-136° C (hexane–acetone), ν_{max}^{KBr} 3040 cm^{-1} (=CH); mol. wt. 449.

Anhydrosecodesmethanolacomonine (III). To 0.2 g of anhydroacomonine was added 0.23 g of periodic acid in 20 ml of water, and the mixture was left at 20° C for 96 h. Then the solution was washed with ether and was made alkaline with sodium carbonate and the reaction product was extracted with hexane. The crystalline residue after evaporation of the hexane was treated with acetone, and 0.054 g of substance (III) with mp 162-164° C (acetone), mol. wt. 415, was obtained. NMR spectrum: one-proton signal at 4.9 ppm (doublet J = 10 Hz), 5.7 ppm (multiplet), 6.05 ppm (doublet), J = 10 Hz, and 6.9 ppm (multiplet).

Deoxyacomonine (VI). Anhydroacomonine (0.07 g) was hydrogenated over platinum in ethanol for 8 h. After the usual working up, 0.06 g of (VI) was obtained with mp 129-132° C, mol. wt. 451.

Oxodeoxyacomonine (VII). Deoxyacomonine (0.06 g) was oxidized with potassium permanganate in aqueous acetone [8]. The aqueous residue after the acetone had been driven off was acidified with a 5% solution of sulfuric acid, and the reaction product was extracted with chloroform. The chloroform extract was treated with saturated sodium carbonate solution, dried with sodium sulfate, and evaporated. The residue was separated preparatively in a fixed layer of silica gel in the benzene–methanol (7:1) system. The product was eluted with chloroform and the solvent was evaporated off. This gave 0.01 g of (VII) with mp 189-192° C; ν_{max}^{KBr} 1640 cm^{-1}; mol. wt. 465.

Acomonine Monoacetate (VIII). A mixture of 0.1 g of acomonine, 2 ml of acetic anhydride, and 1 ml of pyridine was left at room temperature for four days. The solvent was evaporated off, the residue was dissolved in 2% sulfuric acid, the solution was then made alkaline with sodium carbonate with cooling, and the reaction product was exhaustively extracted with ether. This gave 0.08 g of (VIII) with mp 76-78° C (acetone); ν_{max}^{KBr} 1730 cm^{-1}; mol. wt. 509. NMR spectrum: three-proton singlet at 1.99 ppm and one-proton quadruplet at 4.7 ppm (J_1 = 7 Hz, J_2 = 10 Hz).

Anhydrohydroxyacomonine (IX). A. Acomonine (0.1 g) was oxidized with potassium permanganate in aqueous acetone [8]. The aqueous solution after the acetone had been driven off was acidified with sulfuric acid, and the reaction product was extracted with chloroform. The chloroform extract was washed with saturated sodium carbonate solution, dried with sodium sulfate, and evaporated. This gave 0.02 g of (IX) with mp 208-210° C (from acetone); mol. wt. 465.

B. To 0.5 g of acomonine in 13 ml of 1 N $HClO_4$ solution was added 1 g of $Ce(SO_4)_2 \cdot 4H_2O$ in 10 ml of water and the mixture was made alkaline with ammonia to pH 4-5. It was stirred for 39 min, and then the precipitate was filtered off and the solution was extracted with chloroform. The chloroform fraction was treated with a saturated solution of sodium carbonate and dried with sodium sulfate. The solvent was evaporated off to give 0.3 g of (IX) with mp 207-209° C (acetone); perchlorate of (IX) with mp 203-204° C (ethanol).

Reduction of (IX). Sodium tetrahydroborate was added to an aqueous ethanolic solution of the perchlorate of (IX) and after the vigorous evolution of hydrogen the reaction mixture was heated in the boiling water bath for 2 h. Then the solvent was evaporated off and the residue was dissolved in water, and the solution was acidified and extracted with chloroform. The aqueous solution was made alkaline with sodium carbonate with cooling, and the product was extracted with ether. The solvent was evaporated off the residue was treated with acetone. The product so obtained was identical with acomonine.

Anhydrohydroxysecodesmethanolacomonine (X). A mixture of 0.09 g of (IX) and 0.11 g of periodic acid in 10 ml of water was left at room temperature for 4 days. Then the solution was washed with ether and made alkaline with sodium carbonate and the product was extracted with ether. After the solvent had been driven off, the alkaline ether-soluble fraction was treated with hexane. This gave 4 mg of (X) with mp 138-140° C (petroleum ether–acetone); ν_{max}^{KBr} 1670, 1750 cm^{-1}; mol. wt. 463.

Desmethanolsecolycoctonine. A mixture of 1 g of lycoctonine and 1 g of periodic acid in 50 ml of water was left at 20° C for 10 days. Then the aqueous solution was washed with ether and made alkaline with sodium carbonate and the reaction product was extracted with ether. The ethereal extract was evaporated and the residue was crystallized from acetone. This gave 0.1 g of desmethanolsecolycoctonine with mp 186-188° C; ν_{max}^{KBr} 1670, 1750 cm^{-1}; mol. wt. 433.

Desmethanolsecobrowniine. A mixture of 0.96 g of browniine and 1 g of periodic acid in 60 ml of distilled water was left for 12 days. Then the aqueous solution was washed with chloroform and was made alkaline with sodium carbonate and the product was extracted with chloroform. The chloroform solution was evaporated, and the residue was chromatographed on a column of silica gel. This gave 0.03 g of desmethanolsecobrowniine with mp 106-109° C (with foaming), ν_{max}^{KBr} 1680, 1740 cm^{-1}; mol. wt. 433.

SUMMARY

The roots of Aconitum monticola have yielded songorine, songaramine, norsongorine, an amorphous base with the composition $C_{22}H_{35}NO_6$, a base $C_{22}H_{33}NO_6$ with mp 166-167° C, a base $C_{22}H_{33}NO_5$ with mp 161-164° C, and the new alkaloid acomonine. The structure of acomonine has been established on the basis of chemical transformations and spectral properties: It consists of a lycoctonine nucleus with an α-hydroxy group at C_3, an α-methoxy group at C_{10}, β-methoxy groups at C_6 and C_{15}, an α-glycol system at C_7 and C_8, and a methoxy group at C_{19}.

LITERATURE CITED

1. V. Nezhevenko, M. S. Yunusov, and S. Yu. Yunusov, Khim. Prirodn. Soedin., 409 (1974).
2. A. S. Narzullaev, M. S. Yunusov, and S. Yu. Yunusov, Khim. Prirodn. Soedin., 497 (1973).
3. M. S. Yunusov, Ya. V. Rashkes, V. A. Tel'nov, and S. Yu. Yunusov, Khim. Prirodn. Soedin., 515 (1969).
4. N. Bhacca and D. A. Williams, Applications of NMR Spectroscopy in Organic Chemistry, Holden-Day, San Francisco (1964).
5. M. S. Yunusov and S. Yu. Yunusov, Khim. Prirodn. Soedin., 85 (1972).
6. M. S. Yunusov and S. Yu. Yunusov, Khim. Prirodn. Soedin., 335 (1970).
7. V. A. Tel'nov, M. S. Yunusov, and S. Yu. Yunusov, Khim. Prirodn. Soedin., 583 (1970).
8. O. Achmatowicz, Jr., Y. Tsuda, and L. Marion, Can. J. Chem., 43, No. 8, 2336 (1965).