CONVOLININ - ALKALOID FROM CONVOLVULUS PSEUDOCANTHABRICA OF UZBEKISTAN FLORA

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Abstract. A number of alkaloids with a known structure and a novel base, convolinine, were separated from the total alkaloids found in the aerial portion of Convolvulis subhirsutus growing in Uzbekistan. It was discovered that it is an N-hydroxyethyl derivative of convolvin with the structure ()-3-(3,4-dimethoxybenzoyl)-N-hydroxyethyl-nortropane based on spectrum data and X-ray diffraction studies.

Key words: *Convolvulus pseudocanthabrica*, alkaloids, convolinine, (±)3α-(3,4-dimethoxybenzoyl)-N-hydroxyethyl-nortropane (N-hydroxyethyl-convolvin).

The Central Asian flora is abundant with members of the Convolvulus genus and Convolvulaaceae family of plants, sometimes known as bindweeds [1]. Some of them (C. subhirsutus, C. pseudocanthabrica, C. krauseanus, and C. erinaceus) are typical alkaloid transporters [2–3].

A.P. Orekhov and colleagues discovered convolvin and convolamine, two alkaloids from C. psrudocanthabrica, in 1932, and used this information to assess the alkaloid composition of plants in this family [4]. Later, C. subhirsutus, a widespread species in Kazakhstan, was examined by S. Yu. Yunusov and his colleagues [5]. The study produced 17 bases, 9 of which were brand-new, for which the structure was determined [6].

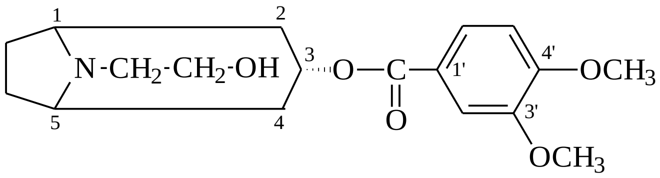
Ten alkaloids (convolvin, convolamine, convolidin, phyllalbine, convolvidin, convolicin, convolin, phyllalbine N-oxide, nortropine, and conpropine) were previously reported to have been isolated from the aerial portion of Convolvulus subhirsutus growing in Uzbekistan [7-8]. After removing convolvin and convolamine from the total alkaloids of the non-phenolic part of the roots of C. pseudocanthabrica, which were collected in the Fergana region, a new alkaloid with m.p. 215-216^{0}C, $[\alpha]_D$ $\pm 0^0$, which differed from the tropane bases described in the literature in terms of chemical and physical properties and chromatographic mobility, was named convolinin. (**1**).

The alkaloid has the composition $C_{18}H_{25}NO_5$. The IR spectrum of the base contains absorption bands of active hydrogen at 3290 cm^{-1}, ester carbonyl group (1749 cm^{-1}), 3,4-disubstituted benzene ring (1601, 1516, 878, 821 cm^{-1}), stretching and bending vibrations CH_3-, CH_2-, CH - groups (2922, 2854, 1463, 1419, 1284, 1274 cm^{-1}), ether bonds -HC-O- (1074, 1021 cm^{-1}).

Two aromatic methoxy group proton signals are seen as two three-proton singlets at 3.87 and 3.89 ppm in the alkaloid's PMR spectra. The existence of a multiplet typical of tropane bases with an intensity of eight proton units from four CH_2 groups of the tropane ring in locations C-2, C-4, C-6, and C-7 is observed in the 1.85-2.85 ppm range. A two-proton multiplet is discovered at 3.77 ppm, most likely from protons of the CH_2 group linked to the nitrogen atom. There is a multiplet at 3.59 ppm that is typical of alkaloids in the tropane series and has an intensity of two proton units from two methine protons at C-1 and C-5 of the tropane ring. A single-proton signal in the form of a triplet from Hα-3 is seen at 5.23 ppm, and this signal is indicative of tropane bases that have been esterified at the hydroxyl at C-3. Three protons of the aromatic ring with a distinctive structure for meta- and para-substitution are detected at 7.60–6.80 ppm. The isolated α-proton appears as a doublet with J≈2.0 Hz at 7.50 ppm, the other α-proton of the aromatic ring appears as a doublet of doublets (meta-constant J≈2.0 Hz and ortho-constant with J≈8.5 Hz) at 7.58 m .d. and finally the β-proton resonates at 6.84 ppm. in the form of a doublet (J≈8.5 Hz). At 2.75 ppm there is a signal with an intensity of two proton units in the form of a multiplet from the protons of the CH_2 group associated with the oxygen function.

It can be assumed that convolinin has a substitution at the nitrogen atom, and the substituent is the -C_2H_4OH radical, based on the aforementioned spectral characteristics, the data of the IR spectrum (the presence of active hydrogen absorption bands), the PMR spectrum, and also taking into account the molecular weight of the alkaloid (M^+335), the unsubstituted nitrogen-containing tropane nucleus. The -CH_2-CH_2-OH group at the nitrogen atom may be inferred from the PMR spectrum's data.

Concluding from the evidence at hand, convolinin appears to be an N-hydroxyethyl derivative of the alkaloid convolvin.



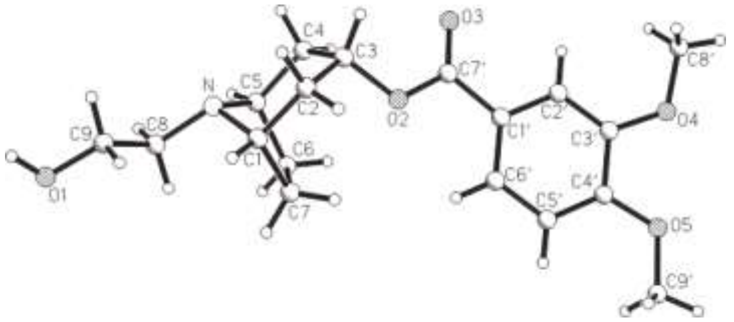


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Figure 1. Spatial structure of the molecule 1

A single crystal of convolinin was subjected to X-ray diffraction analysis (XRD) in order to determine the compound's structure. Convolinine, an alkaloid, crystallizes in the centrosymmetric space group Pbca, and as a result, the crystal includes both of the molecule's enantiomers. Figure 1 depicts molecule 1's spatial structure as determined by X-ray diffraction data. It was feasible to determine the relative arrangement of the C3 center thanks to the X-ray diffraction data, which supported the structure that had been suggested based on the spectral data. The veratroyloxy group is a substituent that is displaced from the tropane nucleus' plane of symmetry and has an 11-axial orientation with regard to it; its torsion angle, C4C3O2C1, is equal to 84.3^0. In general, the orientation and location of the substituent at the C3 position is similar to that observed in o-benzoyltropine hydrochloride. It has flat pieces that are slightly rotated in relation to one another, including an aromatic ring with *п*- and *м* -methoxyl groups and an ester group; the torsion angle C3’C2’C1’O3 is 12.9^0. The position C3 substituent is often oriented and arranged similarly to that of o-benzoyltropine hydrochloride [9].

A bath conformation is adopted by the seven-membered ring. The five-membered heterocycle resembles an N-envelope in shape. Within ±0.008 Å units, the six-membered ring with oxygen functions is planar. The C8-C9 distance is 1.421(8) (due to the significant thermal vibration of the C9 atom) while the other distances and bond angles in the molecule are normal [10].

The O1-H from the original molecule and the N atom, which were changed by the symmetry element 21 (x, 0.25.0), generated an O-H... N type of hydrogen bond that is present in the crystal. The following are the H-bond specifications: The H...N is 0.82, the O-H...N angle is 142, and the N...O distance is 2.867. Because of these H-bonds, a crystallographic axis *a* forms an endless "ribbon" in the crystal.

Thus, the above data allow us to suggest the structure of (±)-3α-(3,4-dimethoxybenzoyl)-N-hydroxyethyl-nortropane for convolinin.

After the *C. pseudocanthabrica plant's* alcoholic extract was used to extract the alkaloids, it was neutralized to pH 7, diluted 1:1 with water, and then alternately extracted with gasoline, chloroform, ethyl acetate, and n-butanol. On a silica gel column, the obtained fractions were chromatographed. As a consequence, from the gasoline portion, crystals with a melting point of 135–136° were separated and recognized as β -sitosterol. A crystal with a melting point of 81–82° was produced by treating the residue (fraction after precipitation, or filtrate) with acetone. This crystal's melting point is not lower than that of a genuine sample of acetamide.

Acetone was also used to treat the chloroform fraction, and when it was condensed, it produced crystals with a melting point of 201–202°. The greatest absorption lines typical of 6,7-dioxycoumarins are seen in the UV spectrum at 297 and 345 nm. Active hydrogen (3115 cm^{-1}-ON), -CN (3045 cm^{-1}), aromatic ring (1605, 1562, 1506), and carbonyl (1702 cm^{-1}) have absorption lines in the IR spectra, respectively. The C-3 and C-4 protons show as two doublets (J=9.6 Hz) in the PMR spectra at 6.15 and 7.79 m.u., respectively. 6.71 m.u. and 7.05 m.u. at the C-5 proton in a single structure. Proton C-8 was visible as a singlet. 3.85 m.u. In the stronger field, a three-proton singlet of the aromatic methoxyl group was seen. Both the N-4 component signal and the N-8 signal exhibit a widening, which is brought on by their long-distance spin-spin interaction. The 192 mass peaks in the chromatogram-mass spectrum served as proof of the molecular weight of coumarin.

The obtained substance was identified as scopoletin, which is isolated for the first time from plants of the genus *Convolvulus*, based on spectrum data and comparison with known coumarin.

A compound having a melting point of 140–141° was identified after the fraction containing ethyl acetate was subjected to chromatography on a silica gel column.

Using the chromato-mass spectrum, its molecular mass, which is equivalent to 208 atomic mass, was identified. Maximum absorption lines in the UV spectrum that are typical of aromatic systems were found at 218, 245, and 330 nm. In the IR spectra, 1,3,4-trivalent ring absorption lines were seen at 1682, 1588, 870, and 814 cm^{-1}. The methyl signal of the ethyl group has a three-proton triplet at (J=7.2 Hz) at 1.22 m.u. in the strong field of the PMR spectrum. 2.N, J=7.2 Hz), 4.13 m.u. in quartet form of O-SN2 protons, and 6.16 m.u. They include 6.71 m.u. at (1N, d, J=8.4, H-51), 6.85 m.u. at (1H, ddd, J=1.6, H-21), and 6.97 m.u. at (1H, d, J=1.6, H-61). These signals are for β - protons in the aromatic cycle of the pyrocatechin system.

This substance is the ethyl ester of caffeic acid, which we have never before detected from a plant, according to spectral characterizations.

Caffeic acid and its esters are recognized to be intermediates in the production of coumarin and flavonoids.

Using the chromatography-mass spectroscopy technique, peaks for aminoalcohol tropine and coumarin ethoxyumbelliferone with molecular masses of 190 and 141, respectively, were found in the alcohol extract of the plant.

It was discovered that plants of the genus Convolvulus contain the separated compounds for the first time.

We have detected these compounds for the first time.

In comparison to the first carbohydrate samples, paper chromatography was used to establish that the residual aqueous fraction included mannose, glucose, and galactose.

Experimental Part

IR spectra were taken on a Perkin-Elmer Fourier IR spectrometer, model 2000 (Vaseline), NMR spectra were recorded on a UNITY-400^+ instrument with an operating frequency of 400 MHz (solvent - CD_3OH, internal standard - HMDS, δ-scale), mass spectrum - on the chromatomass - spectrometer "HP 6C/MS 6890/5973".

Isolation and separation of the amount of alkaloids: [7-8].

Isolation of convolinin. After removing the primary alkaloids, convolvin and convolamine, the mother liquors of the non-phenolic portion of the total alkaloids of the roots were dried, combined in 1:25 ratio with an equivalent amount of silica gel, and put to a silica gel column. Eluted with a 1:1 hexane-benzene combination, followed by benzene and a 3:1 benzene-acetone mixture. Crystals having a chloroform-methanol (2:0.1) system m.p. of 215-216^{0}C (20 mg), and Rf of 0.45.

X-ray diffraction experiment used a CuK-radiation-based ω/2θ-scanning approach on an Xcalibur diffractometer (Oxford Diffraction, T=298 K, Ruby CCD detector, graphite monochromator). The crystals are translucent and around 0.45x0.06x0.08 mm in size, having an acicular form. A adequate set of experimental reflections could not be obtained due to the little amount of the separated material making it unable to grow single crystals of the quality and size needed for X-ray diffraction analysis.

Crystallographic data and main refinement parameters for compound 1: $C_{18}H_{25}NO_5$, M_r =335.39, orthorhombic system, space group Pbca, a=8.367(2), b= 10.783(2), c=38.427(8) Å, V=3466.9(12) $Å^3$, Z=8, $d_{выч.}$=1.266 $г/см^3$, μ_{exp} = 0.093 $мм^{-1}$, number of measured reflections $2\theta \leq 110.67^o$ 2126, number of reflections with $I \geq 2\sigma(I)$ 1388, number of refined parameters 220, $R_1(I \geq 2\sigma(I))$=0.0791, wR_2=0.1821 (over all reflections), GOOF=1.103. The absorption correction was introduced by the Multi-scan method.

The SHELXS-97 software program was used to solve the structure directly. Calculations for structure refinement were done with the SHELXL-97 software. In the full-matrix anisotropic approximation, the least squares approach (by F^2) was used to refine all non-hydrogen atoms. The "rider" model has been improved to incorporate hydrogen atoms in places determined geometrically. The hydroxyl group's H atom was discovered using a different method of electron density synthesis, and it was then refined isotropically, "binding" it to the appropriate oxygen atom.

Materials for X-ray diffraction investigation were deposited at the Cambridge Crystallographic Data Center in the form of a CIF file. (CCDC: W=758625).

References

1. Флора Узбекистана, Изд-во АН УзССР, Ташкент, 1961, **5**, 128 с.
2. А. М. Гаппаров, Н. А. Раззаков, С. Ф. Арипова, Узбекский биологический журнал, 2007, **3**, 83.
3. Алкалоиды, под ред. С. Ю. Юнусова, Ташкент, Изд-во «Фан», 1981, 310 с.
4. А. П. Орехов, Р. А. Коновалова, Журн. общ. химии, 1937, **4**, 646.
5. С. Ю. Юнусов, Т. Т. Шакиров, Н. В. Плеханова, Докл. АН УзССР, 1958, **10**, 17.
6. С. Ф. Арипова, Автореф. дисс… докт. хим. наук, Ташкент, 1991.
7. А. М. Гаппаров, Н. А. Раззаков, С. Ф. Арипова, Химия природ. соедин., 2007, 242 [Chem. Nat. Comp., 2007, **43**, 3, 291 (Engl. Transl.)].
8. А. М. Гаппаров, С. Ф. Арипова, Н. А. Раззаков, В. У. Хужаев, Химия природ. соедин., 2008, 601 [Chem. Nat. Comp., 2008, **44**, 6, 743 (Engl. Transl.)].
9. T. A. Hamor, J. Chem. Soc., Perkin Trans., 1976, **2**, 1359.
10. F. N. Allen, O. Kennard, D. G. Watson, J. Chem. Soc., Perkin Trans II., 1987, **12**, 1.