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ALKALOIDS OF GENTIANA. STRUCTURE OF GENTIANADINE AND GENTIANAMINE

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The alkaloids of plants of the genus Gentiana (family Gentianaceae) have received little chemical study. In some species gentianine [1–5], gentianadine [6, 7], and gentialutine [8] have been found.

We have studied the alkaloids of the epigeal part of G. turkestanorum Gand. and G. olivieri Griseb. (table).

From the total alkaloids of G. turkestanorum, we isolated an amorphous base $C_{11}H_{31}O_3N$ with mp 375°–380° C (decomp.). The base has not been described in the literature and we have called it gentiananine. Its IR spectrum had absorption bands at 1730 cm^{-1} (lactone carbonyl) and 1600 cm^{-1} (aromatic ring). UV spectrum of gentiananine: λ_{max} 220, 265 mμ (log ε 3.88, 3.50), which is similar to that for gentianine.

From the benzene-soluble fraction we isolated a crystalline base with mp 77°–78° C and composition $C_8H_7O_2N$, which we have called gentianadine. The IR spectrum of gentianadine has a band at 1730 cm^{-1} (δ-lactone). The UV spectrum is very similar to that of dihydrogentianine. Gentianadine does not contain a double bond. It differs from gentiananine by a C_2H_2 group. We have assumed that gentianadine has the structure of gentiananine without a vinyl group. All the properties mentioned of this base and its derivatives agree with literature data for the known synthetic substance 4-(2'-hydroxyethyl)nicotinic lactone [4]. However, a small difference was observed in the melting points of the base itself and of its picrate. Consequently, to confirm our assumption we carried out some reactions characteristic for this compound.

Plant	Site of collection	Date of collection	Vegetation period	Total alkaloids, %	Alkaloids present			
					gentianine	gentiananine	gentianadine	gentianamine
G. turkestanorum	Karagai-Tugai, KirgSSR	26.VII 1965	budding	0.66	+	+	—	—
	Nura Gorge and Karagai-Tugai, KirgSSR	25.VIII 1964	flowering	0.64	+	+	+	—
	Alai, Zaalaiskii Range, Basin of the R. Nura KirgSSR	5.VIII 1965	flowering	0.30	+	+	—	+
	Kar-Kara, KazSSR	17.VI 1965	flowering	0.71	-	+	—	—
G. olivieri	Kaplanbek, southern Kazakhstan	6.V 1964	flowering	0.35	+	+	—	—
	Basin of the R. Karatag, TadzhSSR	20.V 1964	flowering	0.67	+	+	—	—
	Basin of the R. Karatag, TadzhSSR	16.V 1965	flowering	0.46	+	+	—	—

From the products of the oxidation of gentianadine with potassium permanganate in alkali we isolated an acid with mp 260°–262° C (decomp.). A mixture of this acid with 3,4-pyridinedicarboxylic acid obtained by oxidizing 4-methylnicotinic acid gave no depression of the melting point. The IR spectra of the two acids were identical. The decarboxylation of gentianadine [4] gave an oily substance possessing an unsaturated nature which gave a picrate with mp 155°–157° C. A mixture with 4-vinylpyridine picrate obtained from gentianic acid gave no depression of the melting point. Thus, the production of 3,4-pyridine dicarboxylic acid and of 4-vinylpyridine confirms the identity of gentianadine (I) as 4-(2'-hydroxyethyl) nicotinic lactone [4].

The mother liquor after the separation of the bases gave three spots on a paper chromatogram, two of which corresponded to gentianadine and gentianine.

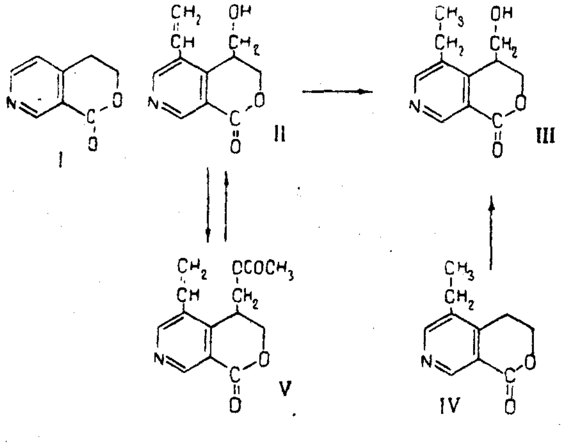
From G. turkestanorum we have isolated a new alkaloid $C_{11}H_{11}O_3N$ with mp 149°–150° C (acetone), which we have called gentianamine. Ethylation of gentianamine with methyl iodide gave a methiodide, which shows the tertiary nature of the nitrogen atom. The IR spectrum of gentianamine has bands at 3200 cm^{-1} (hydroxy), 1720 cm^{-1} (lactone carbonyl), 1660 cm^{-1} (double bond), and 1585 cm^{-1} (pyridine). The presence of a hydroxy group was also confirmed by the production of an O-acetyl derivative (V). UV spectrum: λ_{max} 218 and 250 mμ (log ε 3.96, 4.46), which is very similar to the UV spectrum of gentianine.

On catalytic hydrogenation, gentianamine forms dihydrogentianamine with mp 169°–170° C, the IR spectrum of which has lost the band at 1660 cm^{-1}. The IR spectrum of dihydrogentianamine has one maximum at 270 mμ (log ε 3.60). Thus, the formula of gentianamine can be represented in the following way:

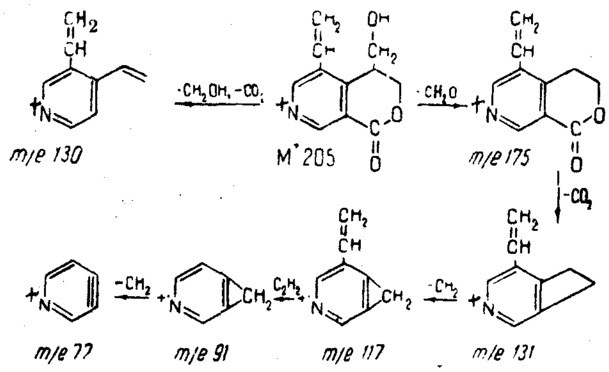
$$C_{10}H_{10}(OH)\ (C{=}O)\,(-O-)\,(>N-)\,(=).$$

When gentianamine was oxidized by a modified Percheron method [9, 10] no C–CH_3 group was found but in the case of dihydrogentianamine acetic acid was detected. This shows the presence of a vinyl group in the pyridine part of the molecule of gentianamine (II). When this alkaloid was oxidized with potassium permanganate in alkali, 3,4,5-pyridinetricarboxylic acid was obtained, this also being formed by the oxidation of gentianine. The action of formaldehyde on dihydrogentianine (IV) [4] gave us a substance identical with dihydrogentianamine (III). The IR spectra of these substances were also identical.

Consequently, gentianamine is a derivative of vinylpyridine and has the structure (II). The mass spectrum of gentianamine confirms the proposed structure (II).



The results of a comparison of the mass spectra of gentianamine and gentianine confirm the following decomposition mechanism:



From the neutral part of the extractive substances of G. turkestanorum we isolated a yellow substance with mp 269°–270° C (from amyl alcohol) having the composition $C_{14}H_{10}O_6$ which, from its properties and IR spectrum, is apparently identical with bellidifolium (pigment B) isolated from G. bellidifolia [11].

Experimental

The UV spectra were taken on a SF-4 spectrophotometer in ethanol and the IR spectra on a UR-10 double-beam spectrometer. The mass spectra were obtained on a MKh-1303 instrument fitted with a system for introducing the sample directly into the ion source at a temperature of 107° C with an ionization energy of 40 eV.

Isolation of the alkaloids. By the usual chloroform extraction, 9 kg of the air-dry epigeal part of G. turkestanorum yielded 43.4 g of ether alkaloids and 16.6 g of chloroform alkaloids, amounting to 0.64% of the weight of the dry raw material. Treatment of the total ether alkaloids with acetone gave 15.5 g of gentiananine. The gentiananine was purified by reprecipitation with acid from dilute alkaline solution. The amorphous gentianine precipitated was washed with water to neutrality, mp 375°–380° C (decomp.).

The substance is insoluble in inorganic solvents and dilute mineral acids. It is readily soluble in caustic alkalies.

Found, %: C 63.7; H 5.86; N 7.2. Calculated for $C_{11}H_{13}O_3N$, %: C 63.7; H 6.3; N 6.7.

The material of the mother liquor after the removal of the gentiananine was separated according to solubilities in various organic solvents. The ether-soluble fraction yielded 8.91 g of gentianine with mp 81°–82° C.

After the removal of the solvent, the mother liquor was treated with benzene. The benzene solution of the alkaloid was evaporated to small bulk. The residue crystallized. Treatment with a mixture of ethanol and ether yielded another 3.7 g of gentianine. On cooling, the mother liquor crystallized. It yielded 2 g of gentiananine with mp 77°–78° C (absolute ether), R_f 0.65 (butanol– acetic acid– water). Gentianadine is readily soluble in methanol, acetone, chloroform, and caustic alkalies, and sparingly soluble in ether and petroleum ether.

Found, %: C 64.76; H 4.9; N 9.45. Calculated for $C_8H_7O_2N$, %: C 64.4; H 4.7; N 9.3; mol. wt. 149 (mass spectrometry).

Gentianadine hydrochloride, mp 195°–196° C (from ethanol).

Gentianadine picrate, mp 156°–158° C (from ethanol).

Oxidation of gentianadine (I). With stirring, an aqueous solution of 0.85 g of potassium permanganate was added to a solution of 0.25 g of gentianadine in 12 ml of 1% caustic potash. The reaction mixture was then heated on a water bath for 4 hr. The manganese dioxide was separated off and washed with hot water. The filtrate was evaporated to dryness. The residue was acidified with an alcoholic solution of hydrochloric acid. The amorphous precipitate of acid that deposited was filtered off with suction, and washed with dilute hydrochloric acid, mp 260°–262° C (decomp.). Yield 0.072 g.

3,4-Pyridinedicarboxylic acid. With stirring, a solution of 0.286 g of potassium permanganate in 15 ml of water was added to a solution of 0.12 g of 4-methylnicotinic acid in 6 ml of 1% caustic potash. The mixture was heated in the water bath for 6 hr. The excess of potassium permanganate was decolorized by the addition of sodium hydrogen sulfite. The subsequent treatment was carried out as for the oxidation of (I). This gave 0.064 g of an acid with mp 263°–265° C (decomp., water).

4-Vinylpyridine. A mixture of 0.47 g of gentianadine, 1.5 ml of water, 0.3 g of caustic potash, and 1.5 g of soda-lime was heated in a bunsen burner flame. 4-Vinylpyridine distilled over; it gave a crystalline picrate with mp 153°–155° C (ethanol). The picrate of 4-vinylpyridine from gentianic acid melts at 155°–157° C (ethanol) [4].

From 9.3 kg of G. turkestanorum collected on 5 August 1965 in the basin of the R. Nura (KirgSSR) 20.5 g of total ether alkaloids and 7.5 g of total chloroform alkaloids was isolated by the usual chloroform extraction. The combined alkaloids amounted to 0.3% of the weight of these dry raw materials.

From the total ether alkaloids 10.8 of gentianine crystallized. The mother liquor from the ether alkaloids was evaporated to dryness and the residue was treated with acetone. This gave 2.2 g of gentiananine.

A solution of 5 g of the material from the mother liquor from the ether alkaloids in 2% sulfuric acid was filtered, washed with ether, and treated with an aqueous solution of sodium perchlorate. The resinous precipitate of perchlorate that deposited was treated with water at 40°–45° C. The aqueous solution was filtered and made alkaline with ammonia, and the base was extracted with ether and chloroform. Elimination of the solvent by distillation yielded 2 g of an ether fraction and 0.8 g of a chloroform fraction of the bases. When the ether fraction (2 g) was treated with a mixture of ether and ethanol, gentianamine precipitated with mp 149°–150° C (from acetone). Yield 0.93 g. Gentianamine is readily soluble in methanol, ethanol, and chloroform. This base does not pass into organic solvents from solutions of caustic alkalies. R_f 0.52 in a thin-layer chromatogram in the methanol– ethyl acetate (9 : 1) system with iodine as the revealing agent.

Found, %: C 65.1; H 5.63; N 7.1. Calculated for $C_{11}H_{11}O_3N$, %: C 64.4; H 5.4; N 6.8; mol. wt. 205 (mass spectroscopy).

Gentianamine oxalate, mp 158°–159° C (from ethanol).

Gentianamine nitrate, mp 127°–128° C (from ethanol– ether).

Gentianamine picrate, mp 146°–147° C (from ethanol).

The sodium salt of gentianamine was obtaining by mixing alcoholic solutions of caustic soda and the base, mp 144°–146° C (with foaming)(ethanol– ether).

Gentianamine methiodide, mp 157°–158° C (from alcohol).

O-Acetylgentianamine. A mixture of 0.087 g of the base, 0.5 ml of pyridine, and 1.5 ml of acetic anhydride was left for 98 hr, the excess of acetic anhydride was distilled off in vacuum, the residue was dissolved in 2% hydrochloric acid, and the solution was made alkaline with ammonia and extracted with ether. After the ether had been distilled off, the crystalline O-acetyl derivative of gentianamine was obtained. Yield 0.052 g, mp 96°–97° C (ether).

Saponification of O-acetylgentianamine. A solution of 30 mg of O-acetylgentianamine in 3 ml of 15% methanolic caustic potash was heated in the water bath for 2 hr, after which it was evaporated to dryness. The residue was dissolved in 10% sulfuric acid and made alkaline with ammonia. The base was extracted with ether. After the ether had been distilled off, gentianamine with mp 149°–150° C was obtained.

Dihydrogentianamine. A solution of 0.1078 g of the alkaloid in 20 ml of methanol was shaken with a platinum catalyst (from 0.0536 g of PtO_2) in an atmosphere of hydrogen. After 3 hr, 1 molar proportion of hydrogen had been adsorbed. The residue after the evaporation of the solvent was crystallized from acetone. This gave 0.0848 g of dihydrogentianamine with mp 170°–171° C.

3,4,5-Pyridinetricarboxylic acid. With constant stirring, an aqueous solution of 0.936 g of potassium permanganate was added to a solution of 0.15 g of the base in 15 ml of 1% caustic soda. The mixture was heated on a water bath for 8 hr. Excess of potassium permanganate was decolorized by the addition of methanol. Manganese dioxide was separated off, the filtrate was evaporated to dryness, and the residue was treated with an alcoholic solution of hydrochloric acid. This gave 30 mg of an acid with mp 262°–264° C (decomp., water).

The action of formaldehyde on dihydrogentianine [2, 4]. A mixture of 1.78 g of dihydrogentianine and 6 ml of formalin was heated at 100°–110° C on a sand bath for 24 hr. After the excess of formaldehyde had been eliminated, the reaction mixture was dissolved in 5% sulfuric acid, the acid solution was made alkaline with ammonia, and the reaction product was extracted with ether. After the ether had been distilled off, a white crystalline precipitate of dihydrogentianamine with mp 172°–173° C (acetone) deposited. Yield 0.85 g.

Four hundred grams of the epigeal part of G. olivieri collected on 20 May 1964 in the gorge of the R. Karatag, TadzhSSR, yielded 2.13 g total ether alkaloids and 0.65 g of total chloroform alkaloids (0.67% of the weight of the plant). Treatment of the total ether alkaloids with a mixture of ether and ethanol gave gentianine (1 g) and, from the mother liquor, on treatment with acetone, gentiananine (0.3 g). When the mother liquor was chromatographed on paper in the butanol– acetic acid– water system it gave three spots with R_f 0.63, 0.70, and 0.81, the first two corresponding to gentianadine and gentianine.

Five kilograms of the epigeal part of G. olivieri collected on 6 May 1964 in Kaplanbek (South Kazakhstan region) yielded 11.08 g of total ether alkaloids and 6.5 g of total chloroform alkaloids (0.3% of total alkaloids on the weight of the dry raw material.)

When the total ether alkaloids were treated with acetone, 1.8 g of gentiananine was isolated. The mother liquor was chromatographed on alumina. Fractions 1–3 (ethereal eluate) gave 0.8 g of gentianine, and the total chloroform alkaloids yielded 1.2 g of gentiananine.

Bellidifolium. 375 g of the plant was extracted with chloroform. The extract was concentrated to 150 ml to give a yellow precipitate of bellidifolium (pigment B) with mp 269°–272° C (from amyl alcohol). Yield 3 g (0.8% of the weight of the dry raw material). IR spectrum: 3430 cm^{-1}, 1660, 1630, 1615, 1590, 1500, 1498, 1095, 1050 cm^{-1}.

Found, %: C 61.3; H 3.79. Calculated for $C_{14}H_{10}O_6$, %: C 61.3; H 3.6.

Summary

1. Gentianine and the three new alkaloids gentiananine, gentianadine, and gentianamine have been isolated from Gentiana turkestanorum and G. olivieri; gentianadine is identical with synthetic 4-(2'-hydroxyethyl) nicotinic lactone.

2. Gentianamine has the structure 5-vinyl-4-(1'-hydroxymethyl-2'-hydroxyethyl) nicotinic lactone.

3. Dihydrogentianine has been converted into dihydrogentianamine.

REFERENCES

1. H. G. Boit, Ergebnisse der Alkaloid-chemie bis, 141, Ber. 1960, 1961.
2. N. F. Proskurnina, ZhOKh, 14, 1148, 1944.
3. N. F. Proskurnina, V. V. Shpanov, and R. A. Konovalova, DAN SSSR, LXVI, 437, 1949.
4. T. R. Govindachari, K. Nagarajan, and S. Rajappa, Chem. and Ind., 1017, 1956; J. Chem. Soc., 551, 1957.
5. I. T. Eskin, Tr. Instituta kraevoi meditsiny, Frunze, no. II, 70, 1959.
6. T. Steinegger, Weibel Pharm. Acta Helv., 26, 259, 1951.
7. Fu and Sun, C. A. 8310, 1959.
8. RZhKh, 22, Zh, 365, 1965.
9. F. Percheron and R. Goutarel, Bull., 10, 1198, 1957.
10. P. Kh. Yuldashev and S. Yu. Yunusov, DAN UzSSR, no. 6, 38, 1962.
11. K. R. Markham, Tetrah., 20, 991, 1964.

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