The structures of the substances isolated were confirmed by UV, IR, and PMR spectroscopies and the results of a study of the products of acid and enzymatic hydrolysis, and also by comparison with authentic substances. This is the first time that substances (I), (V), and (VI) have been isolated from plants of the genus Lathyrus, while (II) is new for this species.

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CHEMICAL COMPOSITION OF THE ROOTS OF Gentiana asclepiadea

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The herbage and roots of milkweed gentian Gentiana asclepiadea L., family Gentianaceae, are widely used in folk medicine for improving digestion [1]. Eleven flavone C-glycosides derived from apigenin and luteolin and three xanthone C-glycosides have been isolated from the epigeal part of this plant previously [2-5]. The pyridine alkaloid gentianine and the secoiridoid gentiopicrin and its 6'-O-glucoside have been found in its roots [6-8], and 13 phenolcarboxyolic acids have also been detected [9].

We have studied the roots gathered in Vitosha in October, 1987. A herbarium specimen (No. 147197) is being kept in the herbarium of the Botanical Institute in Sofia. The comminuted freshly gathered roots (10 kg) were exhaustively extracted with alcohol. The concentrated extract was diluted with water and was reextracted with ethyl acetate, and the ethyl acetate was driven off in vacuum. The dry powder so obtained (40 g) was suspended in water and treated with diethyl ether.

The aqueous residue was separated on columns of polyamide (with the eluents water—ethanol (9:1) and (7:3)) and by preparative paper chromatography in the $CHCl_3$—CH_3COOH—H_2O (20:15:5) system, and substances (I-III) were obtained.

The ethereal solution was extracted with a 10% aqueous solution of caustic potash. The alkaline solution was neutralized with hydrochloric acid and extracted with ethyl acetate. Both fractions (neutral and phenolic) were separated by preparative chromatography on columns of Kieselgel Merck silica gel, with elution by benzene—ethyl acetate ((3:1) and (3:2)). As a result, substances (IV-VI) were obtained.

Substances I and II were identified on the basis of data from chromatographic analysis, acid hydrolysis UV, and PMR spectroscopy [10], and at the same time compared with set pure samples.

Substance (I) (281 mg) was isovitexin (apigenin 6-C-β-glucoside). mp 245-247°C; R_f 0.51 (15% acetic acid, Filtrak 1 paper); R_f of its isomer 0.28 (acid hydrolysis with 10% HCl, 90 min); λ_{max}^{MeOH} 271, 335 nm. PMR spectrum (ppm, DMSO, 100 MHz): 13.58 (s, 1H, 5-OH); 10.40 (2H, 7, 4'-OH); 7.88 (d, J = 8 Hz, 2H, H-2', 6'); 6.90 (d, J = 8 Hz, 2H, H-3', 5'); 6.74 (s, 1H, H-3); 6.48 (s, 1H, H-8); 4.62 (d, J = 10 Hz, 1H, H-1"); 3.00—4.20 (m, sugar protons).

Substance (II) (50 mg) was isoorientin (luteolin 6-C-β-glucoside). mp 230-232°C; R_f 0.35 (15% acetic acid, paper); R_f of its isomer 0.14 (acid hydrolysis with 10% HCl, 90 min); λ_{max}^{MeOH} 272, 340 nm. PMR spectrum (ppm, DMSO, 100 MHz): 13.52 (s, 1H, 5-OH); 7.36 (m, 2H,

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H-2', 6'); 6.84 (d, J = 8 Hz, 1H, H-5'); 6.58 (s, 1H, H-3); 6.50 (s, 1H, H-8); 4.52 (d, J = 10 Hz, H-1"); 3.00–4.10 (m, sugar protons).

Substance (III) – magniferin (1,3,6,7-tetrahydroxyxanthone 2-C-glucoside) was isolated in minor amounts. λ_{max}^{MeOH} 242, 259, 317, 367 nm. It was identified with the aid of UV spectroscopy and a chromatographic comparison with an authentic specimen of mangiferin isolated from Mangifera indica L. and Cratoxylum pruniflorum Kurz. [11].

Substance (IV) (2.2 mg) was isogentisin (1,3-dihydroxy-7-methoxyxanthone). mp 239-241°C; λ_{max}^{MeOH} 232, 256, 305, 365; +NaOMe 230, 266, 340, 380; +$AlCl_3$ 270, 322, 410; +$AlCl_3$/HCl 270, 322, 410. Mass spectrum (m/z): 258 (M^+, 100%, $C_{14}H_{10}O_5$); 243 $(M - CH_3)^+$; 227 $(M - OCH_3)^+$. The melting point found and the UV spectrum of substance (IV) agreed fully with those given in the literature [12].

Substance (V) (49 mg) was β-sitosterol, mp 137-138.5°C. The chromatographic behavior (in four systems), melting point, and IR spectra of substance (V) and of β-sitosterol were identical.

Substance (VI) (2.8 mg) was ursolic acid, mp 268-270°C. It was identified with the aid of a chromatographic comparison with an authentic sample (in four systems).

Substances (I-III) have been isolated from the leaves of milkweed genitan previously. This is the first time that substances (IV)–(VI) have been isolated from this plant.

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