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CAROTENOIDS OF MULBERRY LEAVES AND OF SILKWORM EXCRETA

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At the present time, great attention is being devoted to the search for new cheap sources of carotene [1]. The raw material for this may be certain industrial wastes and, in particular, silkworm excreta (SWE) the complex study of the composition of which has already been carried on over a number of years [2].

In the present communication we present the result of a comparative study of the carotenoid complex of the SWE and of the product consumed by the silkworm — mulberry leaves.

Mulberry leaves and the excreta of silkworm caterpillars of the IVth and Vth instars collected in the Samarkand silkworm station (UzSSR) in 1985 were investigated.

The total carotenoids (Cs) were extracted with acetone—methanol (1:1) and were freed from chlorophylls and lipids by saponification. The preliminary separation of the Cs into carotenes and xanthophylls was carried out by chromatography on columns filled with sucrose [3]. Individual representatives were obtained by TLC on silica gel, using the heptane—methyl ethyl ketone (5:3) solvent system for the separation of the xanthophylls and hexane—acetone (96:4) for the carotenes. During the work, the pigments were protected from degradation by the addition of stabilizers [4] to the eluting solvent systems and by the performance of the operations in the absence of bright light.

The pigments were detected on the plates visually from their colorations, or, in the case of colorless and weakly colored Cs, staining with iodine vapor was used.

The compounds were identified on the basis of chromatography in the presence of markers, by staining with rhodanine, antimony trichloride, and strong acids [5], by spectrophotometry in the visible and ultraviolet regions [6], and by the use of the HCl epoxide test [7] for the presence of a hypsochromic shift in the spectra of the xanthophylls.

The amounts of the pigments were determined colorimetrically on the basis of molar extinction coefficients [6].

As a result of the investigations performed, the complete identity of the carotenoids of the leaves and of the wastes of the silkworm industry was established. It can be seen from the results presented below that the amount of carotenoids in the SWE was higher (~1.4 fold) than in the mulberry leaves. These results correlate with information [8] on the considerable accumulation of chlorophylls in SWE as compared with the leaves.

The relative amounts (% on the total weight) of carotenoids in mulberry leaves and SWE were as follows:

Carotenoids	Mulberry leaves	SWE
Phytoene	2,3	0,8
Phytofluene	1,8	0,5
β-Carotene	41,4	30,3
α-Carotene	3,5	1,2
Hydroxy-α-carotene	2,2	3,4
Lutein	22,4	29,2
Zeaxanthin	2,7	2,5
Violaxanthin	9,0	11,0
Luteoxanthin	3,4	5,6

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Neoxanthin	8.1	9.6
Unidentified xanthophylls	3.2	5.9
Total amount, g/kg dry weight	8.8	12.2

The increase (1.3-fold) of the amount of oxygen-containing forms of the carotenoids in the SWE must be noted, which obviously indicates the occurrence of carotene-oxidation processes in the digestive tract of the silkworm caterpillar. The predominant forms of the carotenoids are β-carotene, lutein, violaxanthin, and neoxanthin, which is extremely characteristic for the leaves of many plants [6].

Thus, the investigations performed have shown that SWE contains a considerable amount of various carotenoids which, together with chlorophyll [2] can, obviously, likewise be utilized in the national economy.

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IRIDOID GLYCOSIDES OF *Verbascum sinuatum*

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Continuing investigations of iridoids of species of mullein [1-3], we have studied the iridoid glycosides of *Verbascum sinuatum* L.

The epigeal part of this plant, collected in the flowering phase in the basin of Lake Sevan, Armenian SSR, was exhaustively extracted with methanol. Successive chromatography, on columns of polyamide sorbent and of silica gel, of an aqueous solution of the methanolic extract that had been washed with organic solvents and freed from flavonoids with alumina gave the new iridoid glycoside (I). From the remainder of the iridoid fraction, aucubin, catalpol, and 6-α-L rhamnopyranosylaucubin (II) [4] were isolated.

Substance (I) — $C_{30}H_{38}O_{16}$, mp 266-266.5°C (methanol—water) $\alpha\ ^{20}_{546}$ −178.5° (s 0.84; pyridine—methanol), ν^{KBr}_{max} 3200-3600 (OH), 1700 (C=O), 1640 1655, 1630 ($C_3 = C_4$), 1516 (Ar) cm^{-1}; $\lambda^{C_2H_5OH}_{max}$ 206, 222, 312 nm. ^1H NMR spectrum (DMSO-d_6, δ, ppm): 7.56 d (2H, J = 9 and 1 Hz; arom); 7.58 d (1H, J = 16 Hz, H-β); 6.82 d (2H, J = 9 and 1 Hz, arom.); 6.46 dd (1H, J = 6 and 1.5 Hz, H-3); 6.43 d (1H, J = 16 Hz, H-α); 5.29 d (1H, J = 5.5 Hz, H-1); 5.01 dd (1H, J = 6 and 3 Hz, H-4); 4.85 d (1H, J = 2, H''); 4.67 d (1H, J = 7.5 Hz, H'); 1.2 d (3H, J = 6.2 Hz, CH_3 of

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