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PHYTOECDYSTEROIDS OF THE PLANT Dianthus hoeltzeri

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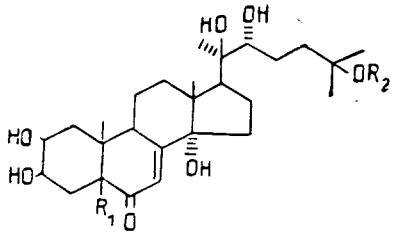
UDC 547.926

We have previously reported the presence of phytoecdysteroids in representative of the genera Silene [1, 2] and Melandrium [3], belonging to the family Caryophyllaceae. Continuing a study of plants of this family for the presence of ecdysteroids, we have investigated the the peigeal part of Dianthus hoeltzeri Winkl. The plant raw material was collected in 1989 on the northern slopes of the Pskem range in the environs of the village of Burchmulla.

The air-dry comminuted raw material (1 kg) was exhaustively extracted with methanol. Then the solvent was distilled off and the dry residue was dissolved in 60% aqueous ethanol. The insoluble part, which did not contain ecdysteroids, was separated off. The ethanol was evaporated off and the residual aqueous solution was shaken with chloroform and then with ethyl acetate. The ethyl acetate extract was chromatographed on a column of silica gel. On elution with chloroform—methanol (9:1), three ecdysteroids were isolated



I R_1=H; R_2=Ac
II. R_1=OH; R_2=H
III. R_1=R_2=H

Substance (I), $C_{29}H_{46}O_8$, mp 196-198°C (from acetone), $[\alpha]_D^{20}$ +59.7±2° (c 0.87; methanol) was obtained with a yield of 0.035% (here and below, the yield is calculated on the weight of the air-dry material). ν_{max}^{KBr} (cm^{-1}); 3400 (OH), 1655 (Δ^7-6-keto grouping), 1725, 1275 (ester group). This compound was identified from its spectral indices and by a direct TLC comparison with an authentic sample as viticosterone E [4, 5].

Phytoecdysteroid (II), $C_{27}H_{44}O_8$, mp 251-252°C (from acetone), $[\alpha]_D^{20}$ +92.4±2° (c 0.56; methanol), yield 0.052%. ν_{max}^{KBr} cm^{-1}); 3400 (OH); 1685 (Δ^7-6-keto grouping). Substance (II) was identified from its spectral characteristics and by a direct TLC comparison with an authentic sample as polypodine B [5, 6].

Compound (III), $C_{27}H_{44}O_7$, mp 241—242°C (from acetone), $[\alpha]_D^{20}$ +60.1±2° (c 0.36; methanol), yield 0.3%, was identified from its physicochemical constants and spectral characteristics and by a direct comparison with an authentic sample as ecdysterone [5].

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ALKALOIDS OF Buxus colchica

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Plants of the genus Buxus (family Buxaceae) are known as containing steroid alkaloids [1-3]. Buxus colchica Pojark (Colchian box) (endemic to the Caucasus) has long been used in folk medicine [3].

Our investigations of this species have shown that the plant is rich in pharmacologically active steroid alkaloids. The total level of bases in the flowering phase in the leaves and green branches found by extraction with ethanol amounted to 3%; and, in the fruit-bearing phase, in the leaves to 0.9%, in green shoots to 0.75%, in lignified branches to 1.1% in the capsules of the fruit to 0.56%, and in the seeds to 1.5%. For the purpose of isolating the total alkaloids and individual bases, we used young branches 15-20 cm long collected in the flowering phase.

The comminuted air-dry raw material was extracted with 80% ethanol. After appropriate purification, the alkaloids were separated into ether-extracted (2.2%) and chloroform-extracted (0.8%) fractions.

The ethereal fraction yielded four bases the identification of which was made from a comparison of their physicochemical constants and spectral characteristics (IR, UV, PMR, ^{13}C-NMR, and mass spectroscopy) with information in the literature.

Base (I), with the composition $C_{25}G_{42}N_2O$, mp 229-231°C, $[\alpha]_D^{20}$ +90° (c 0.2; chloroform) was isolated when the ethereal fraction was precipitated with a mixture of ethanol and ethyl ether. Compound (I) was identified as pseudocyclobuxine D [4].

After the separation of the pseudocyclobuxine D, the mother liquor was evaporated and the residue was dissolved in ethyl ether and subjected to citrate-phosphate polybuffer separation. Fractions with pH 7, 6, 5, and 4 were obtained. Alkaloids (II) and (III) were isolated from the pH 7 fraction by the methods of reprecipitation and stepwise crystallization.

Base (II), $C_{25}H_{42}N_2O$, mp 236-238°C, $[\alpha]_D^{20}$ +94° (c 0.2; chloroform) proved to be cyclobuxine D [4].

Base (III) with mp 167-170°C $[\alpha]_D^{20}$ +104° (c 0.2; chloroform). Mass spectrum: M^+ 401. The identification of this alkaloid is proceeding.

Base (IV), $C_{27}H_{48}N_2$, was obtained from the pH 6 fraction by reprecipitation with a mixture of hexane and ethyl ether. After recrystallization, mp 187-189°C $[\alpha]_D^{20}$ −66° (c 0.2; chloroform). Base (IV) was identified as L-cycloprotobuxine C, isolated previously from Buxus sempervirens L. [1, 5].

The investigation of the alkaloids of Buxus colchica is continuing.

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