We have found that the δ-lactone of gluconic acid specifically inhibits the process of the cleavage of flavonoid [6] and anthraquinone [7] glucosides by enzymes of fungal origin. It was of interest to study the influence of the inhibitor on the enzymes of glucoside-containing plants. We investigated the roots of Frangula alnus, which contain very powerful hydrolytic enzymes [8, 9]. The process of extraction of the hydroxymethylanthraquinones from the roots of Frangula by 70% ethanol was studied by percolation, with the addition to the extractant of from 0 to 5% (of the weight of the roots) of δ-gluconolactone. The contents of glucofrangulin, frangulin, and emodin were determined photocolorimetrically after their separation in a thin layer of silica gel. It was found that as little as 0.1% of the lactone appreciably inhibits the enzymatic decomposition of glucofrangulin and about 1% of the inhibitor is sufficient for the practically complete suppression of the action of the glucoside hydrolases.

To obtain the gluconolactone, an aqueous solution of calcium gluconate was passed through a column of the cation exchange resin KU-2-8 (H^+ form), and the gluconic acid was evaporated to a sirup from which the final product crystallized on standing.

The simplicity of the preparation of the lactone, its physiological inertness, and its fairly high inhibiting capacity permits the hope that it will find wide use as an inhibitor of the glucoside hydrolases of plants during the isolation of various glycosides from them, and also in the production of medicinal substances.

REFERENCES

1. L. Wolf and E. M. Karacsony, Planta med., 11, 432, 1963.
2. Nguyen-Hiep, P. G. Delaveau, and R. R. Paris, Ann. Pharm. Franc., 22, 573, 1964.
3. Nguyen-Hiep, P. G. Delaveau, and R. R. Paris, Ann. Pharm. Franc., 23, 297, 1965.
4. Sh. Okada, M. Yano, and J. Fukumoto, C. A., 62, 15004d, 1965.
6. Z. Kubiak, Dissert. pharmac., 16, 113, 1964.
6. P. I. Gvozdyak, KhPS [Chemistry of Natural Compounds], 3, 291, 1967.
7. P. I. Gvozdyak and Z. F. Yurkovskaya, Farm. zhurnal, 23, no. 6, 1968.
8. J. Pawlaczyk, Acta Polon. Pharmac., 18, 205, 1961.
9. Z. Kubiak and K. Rogoyska, Farmac. Polska, 23, 299, 1967.

16 December 1968

Khar'kov Chemical and Pharmaceutical Scientific-Research Institute

---

UDC 547.92+615.711.5

GLUCOEVONOGENIN AND GLUCOEVONOLOSIDE

S. G. Kislichenko, I. F. Makarevich, and D. G. Kolesnikov

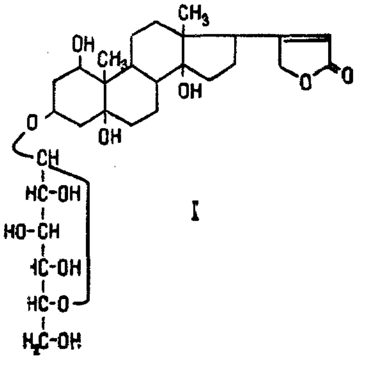
Khimiya Prirodnykh Soedinenii, Vol. 5, No. 3, pp. 193–194, 1969

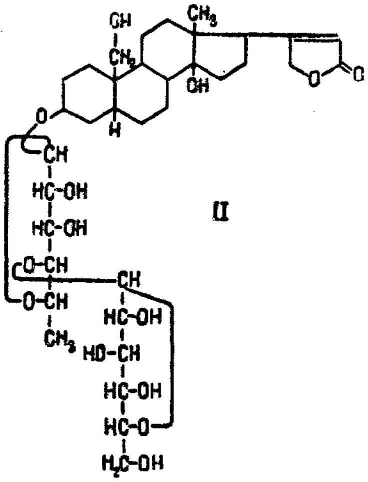
As reported previously, from the seeds of Evonymus europaea auct. (Central Russian variety of the European euonymus) we have isolated the cardenolides evonomonoside, evonoloside, and evonogenin. In this paper we give the results of an investigation of two new cardiac glycosides from native seeds of the plant mentioned.

Comminuted euonymus seeds were defatted with petroleum ether and extracted with 70% ethanol. The ethanolic extracts were evaporated to an aqueous residue. The aqueous solution was purified with petroleum ether and alumina. The cardenolides were extracted with chloroform and then with a mixture of chloroform and ethanol (2 : 1). The ethanolic-chloroformic extracts were evaporated and the mixture of cardenolides was separated by partition chromatography in the toluene—butan-1-ol (3 : 1–1 : 2)/water system. The stationary-phase carrier was alumina saturated with water. The combined fractions yielded evonoloside and two unknown cardenolides which we have called glucoevonogenin (I) and glucoevonoloside (II). Substance I has mp 212–220° C; $[\alpha]_D^{22}$ + 1.9 ± 2° (c 0.7; methanol; UV spectrum: $\lambda_{max}^{ethanol}$ 218 mμ (log ε 4.16). The biological activity, determined by P. I. Bezruk and Zh. A. Lyubetskaya is 0.14 mg/kg body weight in the cat.

Found, %: C 61.64; H 7.80; mol. wt. 563.5 (spectroscopic method). Calculated for $C_{29}H_{44}O_{11}$, %: C 61.25; H 7.79; mol. wt. 568.67.

The hydrolysis of I with the enzymes of the grape snail Helix pomatia gave an aglycone and a monosaccharide. The The aglycone had mp 273–276° C and from the results of paper chromatography, a mixed melting point, coloration with concentrated H_2SO_4, and the IR spectrum, it was identified as evogenin. The monosaccharide with mp 150–152° C, $[\alpha]_D^{22}$ + 53 ± 6° (c 0.2; water, after the establishment of equilibrium; phenylosazone, mp 202–208° C) was identified as D-glucose. An analysis of the molecular rotation of the glycoside I and its aglycone according to Klyne's rule [3] showed that the D-glucose is attached to the aglycone by a β-glycosidic bond. The sugar component is obviously present in the pyranose form since the glycoside is hydrolyzed with difficulty by dilute mineral acids. Thus, glucoevonogenin is 3-(β-D-glucosyl)-1β,5β,14β-trihydroxycard-20(22)-enolide.


I


II

Glucoevonoloside (II) was isolated in small amount in an amorphous but chromatographically individual state. The glycoside II has $[\alpha]_D^{20}$ −8.75 ± 5° (c 1.0; methanol). It is soluble in concentrated H_2SO_4 giving a coloration changing with time: 0 min, brown; 90 min, blue-green; 150 min, emerald. The hydrolysis of II with the enzymes of the grape snail formed a monoglycoside and a monosaccharide. After direct comparison with an authentic sample, the monoglycoside, with mp 158–160° C, $[\alpha]_D^{20}$ −13.3 ± 5° (c 0.37; methanol) was identified as evonoloside [2]. The monosaccharide was identified by paper chromatography as D-glucose. The facts presented, and also the results of a comparison of the polarity of glucoevonoloside with other cardiac glycosides and the observation of the dynamics of enzymatic hydrolysis permit the conclusion that it is the bioside cannogenol 3-α-L-rhamnosyl-β-D-glucoside, with the probable structure II.

REFERENCES

1. S. G. Kislichenko, I. F. Makarevich, and D. G. Kolesnikov, KhPS [Chemistry of Natural Compounds], 2, 440, 1966.

2. S. G. Kislichenko, I. F. Makarevich, and D. G. Kolesnikov, KhPS [Chemistry of Natural Compounds], 3, 241, 1967.

3. W. Klyne, Biochem. J., 47, no. 4, 1950.

2 January 1969

Khar'kov Chemical and Pharmaceutical Scientific-Research Institute

---

UDC 547.944/945

PREPARATION OF APOKHLORIN

A. Abdusamatov, Kh. A. Abduazimov, and S. Yu. Yunusov

Khimiya Prirodnykh Soedinenii, Vol. 5, No. 3, pp. 194–195, 1969

Apokhlorin (a preparation of methylapogalanthamine hydrochloride) (II) has been confirmed by the Pharmacological Committee of the Ministry of Public Health of the USSR as a new Soviet hypertensive preparation for wide medicinal use.