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DIGOXIGENIN 3-α-L-RHAMNOSIDE

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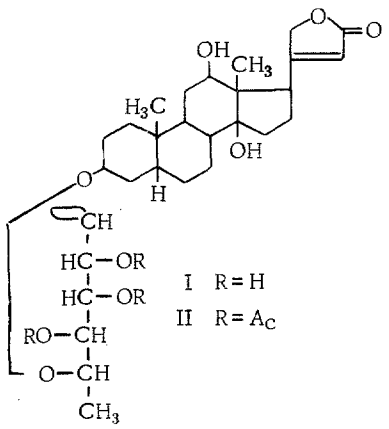
We have obtained digoxigenin 3-(α-L-rhamonside) semisynthetically from digoxigenin and L-rhamnose by the Königs-Knorr method [1]. On approaching the present investigation, we proposed to synthesize a bioside in which rhamnose would be added to the aglycone as two positions—at C_3 and C_{12}. The preparation of this glycoside is of interest for a study of the dependence of the biological action on the position of the sugar component in the aglycone. However, during the work it was found that acetylrhamnosyl bromide condenses only with the hydroxyl at C_3. The OH group at C_{12} is in the equatorial position and nevertheless in this case proves to be unreactive. We assume that this is due to steric hindrance that such a voluminous substituent as acetylrhamnosyl bromide experiences on approaching the hydroxyl at C_{12}. The methyl group at C_{13} and the butenolide ring at C_{17} apparently act as the main hindrances. In other reactions (cf., for example, the acetylation of the cardenolides [2]) where the substituent is less voluminous, the equatorial hydroxyl at C_{12} of digoxigenin proves to be considerably more reactive than the axial hydroxyl at C_3.

As S. I. Lutokhin has established, the monoglycoside (I) synthesized, $C_{29}H_{44}O_9$, processes a high biological activity (0.161 ± 0.013 mg/kg body weight of the pigeon). Its IR spectrum has one absorption maximum at 219 mμ (log ε 4.14). Results of a comparison of the molecular rotations of the glycoside and the aglycone, in accordance with Klyne's rule [3], show that the L-rhamnose is attached by an α-glycosidic bond. First of all, to prove the position of addition of the sugar residue to the aglycone, the triacetate of the glycoside (II) was obtained. The latter was isolated by the chromatography on alumina of the mixture of cardenolides formed after the condensation of acetylrhamnosyl bromide with digoxigenin. Then, an analysis of the rate of acetylation of the triacetate (II) carried out by the method that we have proposed [2] showed that the OH group undergoing acetylation is equatorial. Since in the initial agylcone of the two secondary OH groups one is axial (at C_3) and the other equatorial (C_{12}), the determination of the position of addition of the sugar component can be made unambiguously: the L-rhamnose adds to digoxigenin at the C_3 axial hydroxyl. The glycoside synthesized, therefore, corresponds to formula (I). In properties, it is identical with substance γ obtained from Antiaris toxicaria Lesch. by Reichstein et al. [4, 5], and provisionally characterized as digoxigenin 3-(α-L-rhamnoside).



I R = H

II R = Ac

Experimental

The substances were analyzed after drying for 2 hr over phosphorus pentoxide at 80° C in vacuum (0.01 mm Hg). The following systems of solvents were used for the chromatography of the cardenolides on paper: chloroform—tetrahydrofuran (1 : 1)/formamide; m-xylene—methyl ethyl ketone (1 : 1)/formamide; benzene/formamide.

A reaction flask fitted with a stirrer was charged with 1 g of digoxigenin having mp 220-222° C, 30 ml of anhydrous dioxane, and 10 g of silver carbonate. Then 5 g of acetylrhamnosyl bromide dissolved in 30 ml of toluene was added over 1.5 hr at the boiling point of the reaction mixture. After this it was boiled for another 30 min. At the end of the reaction, the solution was filtered, the residue was washed with chloroform, and the filtrate was evaporated in vacuum. The residue was chromatographed on 40 g of alumina (activity grade III). Chloroform elution yielded 740 mg of the triacetate (II) in the form of an amorphous but chromatographically individual substance. By eluting the column with a mixture of ethanol and chloroform (5 : 95), 310 mg of crystalline digoxigenin with mp 219-221° C was obtained.

Digoxigenin rhamnoside triacetate (II). The substance has $[\alpha]_D^{25}$ −15.3 ± 3° (c 0.82; chloroform), dissolves in concentrated sulfuric acid forming a coloration changing with time: 0 min—yellow, 2 min—yellow-brown, 50 min—pink. Under the action of acetic anhydride in pyridine, it is acetylated almost completely in 1 hr (22–25° C; ratio of acetic anhydride to pyridine 1 : 2).

The triacetate (II) was dissolved in 100 ml of methanol saturated with gaseous ammonia and the solution was left at room temperature for 18 hr. The completeness of the saponification of the acetyl groups was checked by chromatography. The methanol and ammonia were driven off in vacuum. The residue was dissolved in 150 ml of a mixture of ethanol and chloroform (1 : 3), the solution was washed free from salt with water (3 × 5 ml), dried with anhydrous sodium sulfate, and evaporated. The glycoside was crystallized from acetone, giving 510 mg of the pure substance.

Digoxigenin 3-(α-L-rhamnoside) (I). The glycoside melts at 268–274° C, $[\alpha]_D^{26}$ −20.1 ± 2° (c 1.76; methanol), and dissolves in concentrated sulfuric acid giving a coloration changing with time: 0 min—yellow, 15 min—yellow-orange, 30 min—pink. Found, %: C 64.71; H 8.09; M 538. Calculated for $C_{29}H_{44}O_9$, %: C 64.90; H 8.26; M 536.67.

Conclusions

Digoxigenin 3-(α-L-rhamnopyranoside) has been synthesized from digoxigenin and L-rhamnose with a yield of 37%; it has been shown that in the condensation of acetylrhamnosyl bromide with the agylcone practically no reaction takes place at the C_{12} hydrdroxyl because of steric hindrance.

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