SUMMARY

1. Gitogenin, diosgenin, kogagenin, ruscogenin, and tokorogenin have been isolated for the first time from *Funkia ovata* Spr.

2. Structures of two new steroid glycosides — funkiosides E and G — have been determined.

LITERATURE CITED

1. P. K. Kintya, N. E. Mashchenko, N.I. Konovova, and G. V. Lazur'evskii, Khim. Prirodn. Soedin., 267 (1976).
2. N. E. Mashchenko, G. V. Lazur'evskii, and P. K. Kintya, Khim. Prirodn. Soedin., 123 (1977) [in this issue].
3. Y. Mori and T. Kawasaki, Chem. Pharm. Bull., 21, 224 (1973).
4. R. Kuhn and H. Trischman, Chem. Ber., 96, 284 (1963).
5. R. Tschesche and G. Wulff, Fortschr. Chem. Org. Naturstoffe, 30, 461 (1973).
6. W. Klyne, Biochem. J., 47, No. 4, xli (1950).

GLYCOSYLATION OF CARDENOLIDES.

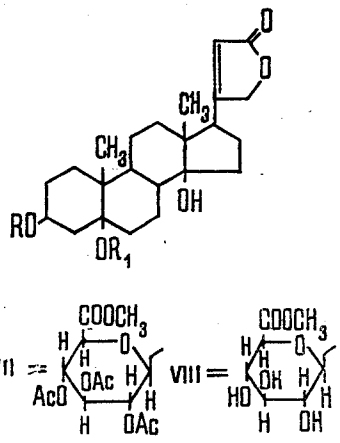
VI. PERIPLOGENIN MONO- AND DIGLUCOSIDURONIC ACIDS

N. Sh. Pal'yants and N. K. Abubakirov

UDC 547.918:547.926+615.711.5

Work on the synthesis of cardiac 3,5-bisglycosides [1] has been continued. The condensation of methyl 2,3,4-tri-O-acetyl-1-bromo-1-deoxy-α-D-glucuronate with periplogenin (I) under the conditions of the Koenigs–Knorr method [2] has led to the acetylated glycoside (III) and the less polar compound (V).

The NMR spectrum of the acetate (III) (see Fig. 1) contains the signals of the protons of three acetyl groups in the 2.00-2.05 ppm region, the three-proton singlet of a methoxycarbonyl group $COOCH_3$ at 3.69 ppm, and the doublet of the proton of a glucuronic acid residue at C-5' at 4.00 ppm (J = 10 Hz). This means that product (III) is the triacetate of the methyl ester of a periplogenin monoglucosiduronic acid. It is obvious that the sugar residue is attached to the OH group at carbon atom 3. The NMR spectrum of compound (III) clearly exhibits the doublet of the anomeric proton at 4.64 ppm with J = 7.5 Hz, which shows the β configuration of the glycoside bond [3]. Thus, substance (III) is periplogenin 3α-O-(methyl 2',3',4'-tri-O-acetyl-β-D-glucosiduronate).



I. R = R₁ = H
II. R = Ac; R₁ = H
III. R = VII; R₁ = H
IV. R = VIII; R₁ = H
V. R = R₁ = VII
VI. R = R₁ = VIII

Institute of the Chemistry of Plant Substances, Academy of Sciences of the Uzbek SSR, Tashkent. Translated from Khimiya Prirodnykh Soedinenii, No. 1, pp. 72-77, January-February, 1977. Original article submitted November 15, 1976.

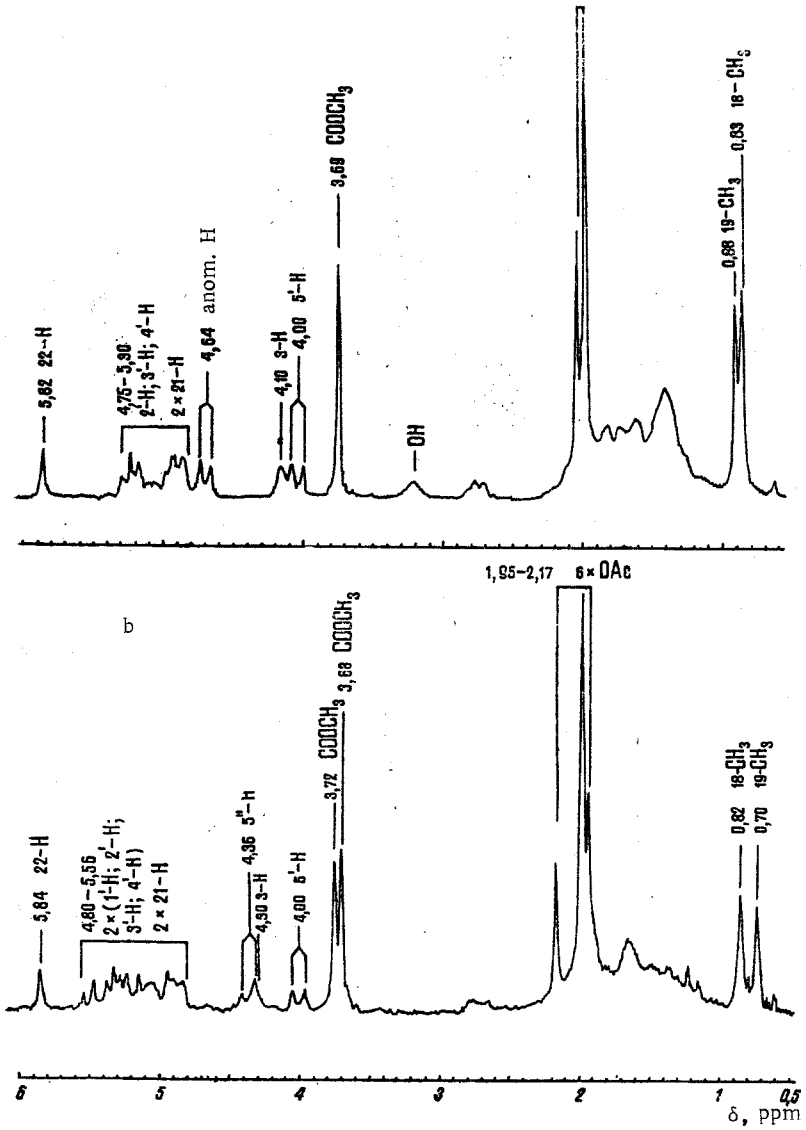


Fig. 1. NMR spectra of the triacetate of the methyl ester of periplogenin glucosiduronic acid (a) and of the hexaacetate of the dimethyl ester of periplogenin 3,5-diglucosiduronic acid (b).

TABLE 1. Chemical Shifts of the C-18 and C-19 Protons in Acetylated and Glycosylated Derivatives of Periplogenin (standard compound HMDS)

Periplogenin derivative	Solvent	Chemical shift of the protons (δ, ppm)	
		at C-18	at C-19
Periplogenin acetate (II) [4]*	$CDCl_3$	0,84	0,92
Triacetate of the glycoside (III)	—„—	0,83	0,88
Hexaacetate of the diglycoside (V)	—„—	0,82	0,70
Monoglycoside (IV)	C_5D_5N	0,90	0,96
Diglycoside (VI)	—„—	0,87	1,08

*In the original, the chemical shifts are given in relation to TMS as standard; we have recalculated them to HMDS.

In the NMR spectrum of product (V) (see Fig. 1), two three-proton singlets of methyl protons of ester groups ($COOCH_3$) at 3.68 and 3.72 ppm and two doublets relating to the two protons on the C_5 carbon atoms of the glucuronic acid residues at 4.00 and 4.36 ppm, each with J = 10 Hz, are clearly visible. These facts show that two sugar residues are attached to the aglycone and compound (V) is the hexaacetate of the dimethyl ester of a periplogenin diglucosiduronic acid. It is not a matter of doubt that one of the sugar residues is attached to the hydroxy group C-3 of periplogenin. The position of attachment of the second carbohydrate component was determined by a comparison of the NMR spectra of the acetates (III) and (V) and literature data.

It can be seen from Table 1 that the chemical shifts of the C-18 protons of the first three compounds have approximately the same value. The protons of the methyl group at C-19 behave differently. While the C-19 protons of periplogenin acetate (II) and of the monosubstituted acyl glycoside (III) give signals at 0.92 and 0.88 ppm, respectively, the signals of the C-19 protons of the acetylated bisglycoside (V) appear at 0.70 ppm. It is obvious that the sugar residue that has caused this diamagnetic shift must be located close to the C-19 methyl group, i.e., of the two probable positions of attachment of the second sugar — the OH group at C-5 and C-14 — preference must be given to the hydroxy group at C-5. The correctness of this choice is also shown by the fact that in the NMR spectra of product (V) the signal of the proton at C-3, which is located close to carbon atom 5 of the steroid nucleus, is shifted downfield by 0.2 ppm as compared with the analogous signal of the monoglycoside (III). Consequently, substance (V) is the hexaacetate of the dimethyl ester of periplogenin 3,5-diglucosiduronic acid.

It has been shown previously [1] for the case of strophanthidin 5-rhamnoside and 3,5-bisglycoside as examples, that the replacement of the hydroxy group at C-5 by a sugar residue has no substantial influence on the position in the NMR spectrum of the three-proton singlet of the C-18 angular methyl group. For periplogenin diglucosiduronic acid, we come to the conclusion that the shift of the protons at C-19 has analytical value and can be used for the recognition of other glycosides of analogous structure.

The action on compound (III) of a solution of sodium methoxide in absolute methanol gave the methyl ester of the β-glucosiduronic acid (IV), and on compound (V) it gave the dimethyl ester of periplogenin 3,5-diglucosiduronic acid (VI). The analytical results confirmed that in the first place a monoglycoside and in the second a diglycoside was obtained. As can be seen from Table 1, in the case of the diglycoside (IV), in contrast to the hexaacetate (V), the free sugar residue at C-5 does not screen but, conversely, descreens the protons at C-19, shifting the signal by 0.12 ppm.

There is no doubt that the carbohydrate component at C-3 in the bisglycoside (VI) is attached in the same way as in compound (VI) by a β-glycosidic bond. From a calculation by the method of molecular differences [5] ($[M]_D$ VI —336.5°; $[M]_D$ IV —110.8°; $\Delta[M]_D$ —225.7°) it follows that the second sugar residue in compound (VI) is attached to the C-5 OH group by a β-glycosidic bond [6].

Thus, compound (VI) is periplogenin 3β,5β-di-O-(methyl β-D-glucosiduronate).

EXPERIMENTAL

For thin-layer chromatography (TLC), we used KSK silica gel with 5% of gypsum. Methyl 2,3,4-tri-O-acetyl-1-bromo-1-deoxy-α-D-glucuronate was obtained under the conditions described by Bollenback et al. [7].

Periplogenin 3β-O-(Methyl 2',3',4'-tri-O-acetyl-β-D-glucosiduronate) (III) and Periplogenin 3β,5β-di-O-(Methyl 2',3',4'-tri-O-acetyl-β-D-glucosiduronate) (V) from (I). Periplogenin (I) (1.17 g = 3 mmoles) was dissolved in 60 ml of a mixture of dichloroethane and toluene (5:1 by volume). The solution was heated to the boil with stirring. After the first 10 ml of solvents had been distilled off, 1 g of CaO and 0.58 g of Ag_2CO_3 were added to the reaction mixture. Then a solution of 2.50 g (6.25 mmoles) of the acetobromo derivative of methyl glucuronate in a mixture of dichloroethane and toluene was added dropwise. The acetobromo derivative was added over an hour at such a rate that the volume of the reaction mixture remained constant. Simultaneously, silver carbonate was added in two portions of 0.58 each. The reaction mixture was then boiled for another 20 min. The course of the reaction was monitored by LGC in the chloroform—ethanol system (15:1). The reaction mixture was filtered, the precipitate on the filter was washed several times with chloroform, and

the solution was evaporated to dryness in vacuum. The residue was chromatographed on silica gel, being eluted with benzene containing gradientwise increasing amounts of chloroform. This gave 684 mg of (V), 905 mg of (III), and 180 mg of the initial (I) in the crystalline form with yield of 22.3, 42.6, and 15.4%, respectively. Compound (V), $C_{49}H_{66}O_{23}$, had mp 250°C (from ethanol); $[\alpha]_D^{29°}$ −29.0 ± 3° (c 1.38; chloroform); $\lambda_{max}^{C_2H_5OH}$: 218 nm (log ε 4.25); ν_{max}^{KBr}, cm^{-1} 3530 (OH), 1760 (C=O), 1635 (C=C), 1225 (C–O–C). The NMR spectrum is given in Fig. 1.

Compound (III), $C_{36}H_{50}O_{14}$, had mp 178-181°C (from a mixture of chloroform and ether); $[\alpha]_D^{24}$ +0.8 ± 3° (c 1.18; chloroform); $\lambda_{max}^{C_2H_5OH}$: 217 nm (log ε 4.23): ν_{max}^{KBr}, cm^{-1}: 3560 (OH), 1750 (C=C); 1630 (C=C); 1220 (C–O–C). The NMR spectrum is given in Fig. 1.

Periplogenin 3β-O-(Methyl β-D-glucosiduronate) (IV) from (III). The triacetate (III) (350 mg) was dissolved in 15 ml of absolute methanol and 3 ml of 0.1 N CH_3ONa solution was added. After 15 min, JU-2-8 cation-exchange resin was added to the reaction mixture until it was neutral. The solution was filtered off from the resin, and this was washed several times with methanol. The methanolic solution was evaporated to dryness. The residue was chromatographed on silica gel with elution by chloroform containing gradientwise increasing amounts of methanol. This gave 220 mg of (IV). The yield in compound (III) was 76.6%. The monoglycoside (IV), $C_{30}H_{44}O_{11}$, had mp 228-230°C (from ethanol); $[\alpha]_D^{24}$ −19.1 ± 3° (c 1.20; methanol); $\lambda_{max}^{C_2H_5OH}$: 217 nm (log ε 4.18); ν_{max}^{KBr}, cm^{-1}: 3450-3500 (OH), 1740, 1625 (butenolide ring). NMR spectrum (C_5D_5N), ppm: 0.89 (3H at C-18, s); 0.96 (3H at C-19 s); 3.54 (3H at $COOCH_3$, s); 4.90, 5.22 (2H at C-21, q, centers of doublets, J = 18 Hz); 6.00 (H at C-22, s).

Periplogenin 3β,5β-di-O-(methyl β-D-glucosiduronate) (VI) from (V). The hexaacetate (V) (350 mg) was dissolved in 22 ml of absolute methanol, and 9 ml of a 0.1 N solution of CH_3ONa in methanol was added. The reaction mixture was worked up as described above for the preparation of (IV). The residue was chromatographed on silica gel with elution by mixtures of chloroform and methanol in ratios of 100:5 and 100:8 (by volume). Elution with the chloroform–methanol (100:8) yielded 105 mg of chromatographically homogeneous (VI). The yield on the hexaacetate (V) was 40%. The bisglycoside (VI), $C_{37}H_{54}O_{17}$, was an amorphous substance with $[\alpha]_D^{24}$ −43.7 ± 3° (c 0.55; methanol); $\lambda_{max}^{C_2H_5OH}$: 216 nm (log ε 4.18); ν_{max}^{KBr}, cm^{-1}: 3400-3500 (OH), 1745, 1625 (butenolide ring). NMR spectrum (C_5D_5N), ppm: 0.86 (3H at C-18, s), 1.08 (3H at C-19, s), 3.60 (3H at $COOCH_3$, s), 3.62 (3H at $COOCH_3$, s), 6.00 (H at C-22, s).

SUMMARY

The Koenigs–Knorr condensation of the acetobromo derivative of methyl glucuronate with periplogenin has given the methyl ester of periplogenin monoglucosiduronic acid and the dimethyl ester of periplogenin 3,5-diglucosiduronic acid.

LITERATURE CITED.

1. N. Sh. Pal'yants, M. V. Gorovits, and N. K. Abubakirov, Khim. Prirodn. Soedin., 765 (1976).
2. W. Koenigs and E. Knorr, Ber., 34, 957 (1901).
3. M. Mamsui and M. Okada, Chem. Pharm. Bull., 19, 395 (1971).
4. K. Tori and K. Aono, Ann. Rept. Shionogi Res. Lab., 15, 130 (1965).
5. W. Klyne, Biochem. J., 42, No. 2, xli (1950).
6. T. E. Timell, W. Entermann, F. Spencer, and E. J. Soltes, Can. J. Chem., 43, 2296 (1965).
7. G. N. Bollenback, J. W. Long, D. G. Benjamin, and J. A. Lindquist, J. Am. Chem. Soc., 77, 3310 (1955).