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Note

Stereospecific syntheses of β-anomeric ester glucosyluronic acid derivatives by the orthoester glycosylation method*

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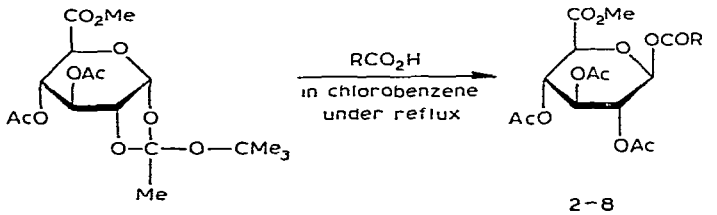
D-Glucuronic acid conjugation plays an important role in the metabolism of animals. D-Glucosyluronic esters, 1-*O*-acyl derivatives of D-glucuronic acid, have been isolated from excreta of animals administered such drugs containing carboxylic acid groups, as benzoic acid[1], salicylic acid[2], *p*-aminosalicylic acid[3], indomethacin[4], and isopanoic acid[5]. Great difficulty is experienced in isolation and identification of such esters from the excreta of animals, because they are unstable and decompose readily during manipulation. Therefore, derivatization of the isolated glucosyluronic esters to a stable form, such as *O*-acetyl methyl esters, is widely used for their identification.



1 2–8

This paper describes an alternative route for preparation of the *O*-acetyl methyl ester of glucosyluronic esters by use of the orthoester glycosylation method[6–8]. The conventional pathway to the glucosyluronic esters is through condensation of methyl 1-bromo-1-deoxy-2,3,4-tri-*O*-acetyl-α-D-glucopyranuronate with the silver or potassium salt of a carboxylic acid[1,3], or with a carboxylic acid in the presence of a catalyst such as silver oxide[9]. The glucosyluronic esters are also prepared by the acid chloride[10] and the carbodiimide methods[11,12], but both routes sometimes give a mixture of anomers. In the previous paper, we reported 1-*O*-acylation of D-glucopyranose using glucopyranose 1,2-*tert*-butyl orthoacetate[6]. The reaction of 3,4,6-tri-*O*-acetyl-α-D-glucopyranose 1,2-(*tert*-butyl orthoacetate) with a free carboxylic acid in chloro-

*Part IV of Studies on Glycosylation.

TABLE I

ANALYTICAL DATA FOR METHYL (2,3,4-TRI-*O*-ACETYL-1-*O*-ACYL-β-D-GLUCOPYRANOSYL)URONATES

	Acyl group	*m.p.* (°C)	$[\alpha]_D^{20}$(°)[a]	*Formula*	*Calc.*			*Found*			*T.l.c.*[b] R_F
					C	*H*	*N*	*C*	*H*	*N*	
2	propanoyl[c]	153–154	+ 9.4	$C_{19}H_{22}O_{11}$	49.23	5.68		49.40	5.76		0.45
3	benzoyl[d]	144–145	−16.3	$C_{20}H_{22}O_{11}$	54.79	5.06		55.04	5.09		0.56
4	salicyloyl	162–162.5	−37 0	$C_{20}H_{22}O_{12}$	52.87	4.88		53.00	4.92		0.49
5	cinnamoyl[e]	175–176	− 2.0	$C_{22}H_{24}O_{11}$	56.88	5.21		56.82	5.24		0.44
6	*p*-nitrophenylacetyl	166	−18.8	$C_{21}H_{23}O_{13}N$	50.71	4.66	2.82	50.75	4.57	3.05	0.34
7	hippuroyl	160	+ 2.3	$C_{22}H_{25}O_{12}N$	53.33	5.09	2.83	53.49	5.19	3.10	0.08
8	nicotinoyl	122–124	−12.9	$C_{19}H_{21}O_{11}N$	51.94	4.82	3.19	51.87	4.82	3.29	0.19

[a]In $CHCl_3$, *c* 1.0. [b]Solvent: (10:1) benzene–ether. [c]Pravdic and Keglević[11] prepared the 1-*O*-propanoyl derivative enriched in the β anomer (m.p. 149–151°, $[\alpha]_D$ +9°). [d]Compare lit.[1] m.p. 145°, $[\alpha]_D^{28}$ −16.6°; lit.[11] m.p. 142–143°, $[\alpha]_D$ −16°. [e]Compare lit.[11] m.p. 170–171°, $[\alpha]_D$ −15°.

benzene under reflux afforded the 1-*O*-acyl-β-D-glucopyranose tetraacetate in good yield. This method was successfully applied for synthesis of the 1-*O*-acylglucopyranose derivatives of all of the carboxylic acids examined. Therefore, we are interested in application of the orthoester method to prepare the glucosyluronic ester derivatives.

RESULTS AND DISCUSSION

Methyl 3,4-di-*O*-acetyl-α-D-glucopyranuronate 1,2-(*tert*-butyl orthoacetate) (**1**) was prepared by the general procedure for the D-glucopyranose 1,2-(*tert*-butyl orthoacetate) derivative[8]. Thus, methyl 2,3,4-tri-*O*-acetyl-1-bromo-1-deoxy-α-D-glucopyranuronate[13] was treated with *tert*-butyl alcohol in nitromethane containing 2,6-lutidine to give **1**. The orthoester **1** showed a signal for the orthoacetyl group at 1.83 p.p.m. as a singlet in the n.m.r. spectrum. The orthoester **1** was allowed to react with an excess of various carboxylic acids in chlorobenzene under reflux for 15–30 min. Evaporation of the mixture and subsequent crystallization of the residue from ethanol gave methyl (2,3,4-tri-*O*-acetyl-1-*O*-acyl-β-D-glucopyranosid)uronate in good yields. The carboxylic acids used were propanoic, benzoic, salicylic, cinnamic, *p*-nitrophenylacetic, hippuric, and nicotinic acids, and the 1-*O*-acyl derivatives of methyl D-glucuronate (compounds **2–8**) prepared are listed in Table I. The products (**2–8**) were pure by t.l.c. and elemental analyses.

By the reaction employed, exclusive production of β-anomers was anticipated from the results reported on 1-*O*-acylation of glucopyranose by the orthoester method[6]. Proof that 1-*O*-acylglucuronic acid derivatives (**2–8**) were β anomers was provided by their optical rotations and their n.m.r. and i.r. spectra. As shown in Table I, compounds **2–8** showed negative or low, positive specific rotations. The specific rotation of the 1-*O*-propanoyl derivative (**2**) was in good agreement with that of methyl β-D-glucopyranuronate tetraacetate ($[\alpha]_D^{23}$ +7.4° (*c* 2, chloroform)[13]; compare the α anomer, $[\alpha]_D^{20}$ +119.2° (*c* 1.02, chloroform)[14]).

TABLE II

N.M R. SPECTRA OF METHYL (2,3,4-TRI-*O*-ACETYL-1-*O*-ACYL-β-D-GLUCOPYRANOSYL)URONATES[a]

	Acyl group	*H-1 (δ, and $J_{1,2}$ in Hz)*	*H-5 (δ, Hz)*
2	propanoyl	5.84, d, *J* 6 8	4.23, m
3	benzoyl	6.02, d, *J* 7 5	4 27, m
4	salicyloyl	6.00, d, *J* 8 0	4 31, m
5	cinnamoyl	5 96, d, *J* 7 2	4 30, m
6	*p*-nitrophenylacetyl	5 83, d, *J* 6 4	4 24, m
7	hippuroyl	5.89, d, *J* 6 8	4 27, m
8	nicotinoyl	6 07, d, *J* 7 0	4 35, m

[a]Other signals of the sugar moiety were commonly observed as follows: 2 0–2.1 (9H, 3 × $COCH_3$), 3.7–3 8 (3H, s, CO_2Me), 5 0–5.6 (3H, m, H-2,3,4)

In the ^{1}H-n.m.r. spectra (Table II), the anomeric-proton signal was observed as a doublet in the range of 5.83–6.07 p.p.m. and its spacing was 6.4–8.0 Hz, confirming that the 1-*O*-acyl products (**2–8**) were exclusively the β anomers. The products also showed the H-5 signal of the sugar moiety between 4.24–4.35 p.p.m. as a multiplet, affording additional evidence for the β configuration. Thus, the H-5 signals of β anomers of the acetylated methyl glucopyranuronates appeared near 4.2 p.p.m. as multiplets, whereas those of the α anomers appeared[15] as doublets at 4.3–4.5 p.p.m. having coupling constants of 10 Hz. The i.r. spectra of the products showed the characteristic absorption band of β anomers near 905 cm^{-1}, but not that of the α anomer (930 cm^{-1})[16].

N.m.r. spectra showed the acetyl group to be retained at O-2 of the sugar moiety, because signals from three acetyl groups were observed. This fact was supported by the i.r. spectra, in which no absorption band of a hydroxyl group was observed.

The reaction with such hydroxycarboxylic acids as salicylic acid produced the 1-*O*-acyl derivative (**4**) and no 1-*O*-aryl derivative was obtained. This result indicates that the condensation of the ortho ester with the carboxyl group proceeded more rapidly than with the phenolic group.

The ortho esters are susceptible to acid, and glycosylation by use of ortho esters proceeds in the presence of an acid catalyst[8]. Consequently, when the reaction is employed without catalyst, the carboxylic acid itself may play a role as catalyst at the first stage of reaction. After protonation of the *O-tert*-butyl group, the reaction seems to proceed by a mechanism similar to that reported for 1-*O*-alkylation of glucopyranose derivatives by the orthoester method[17].

In conclusion, the orthoester method constitutes a stereospecific way for preparing 1-*O*-acyl-β-D-glucosyluronic acids. As formation of β-D-glucosyluronic acid derivatives is a common metabolic pathway in drug metabolism, the orthoester method described here is a suitable procedure for the preparation of the *O*-acetylated methyl glycosides of glucosyluronic esters.

EXPERIMENTAL

General methods. — Melting points are uncorrected. Specific rotations were measured in chloroform at room temperature with a Yanagimoto OR-50 polarimeter. The ^{1}H-n.m.r. spectra were determined in chloroform-*d* solution with a Hitachi R-22 spectrometer (90 MHz) and tetramethylsilane was used as the internal standard. The i.r. spectra were recorded on a Jasco IRA-1 spectrometer. Thin-layer chromatography was performed on precoated plates of silica gel F_{254} (E. Merck, Darmstadt, Germany) with 10:1 benzene–ether or chloroform throughout this experiment.

*Methyl 3,4-di-*O*-acetyl-α-*D*-glucopyranuronate 1,2-*(tert*-butyl orthoacetate)* (**1**). — Methyl 2,3,4-tri-*O*-acetyl-1-bromo-1-deoxy-α-D-glucopyranuronate[13] (1.0 g) and *tert*-butyl alcohol (0.5 mL) were dissolved in nitromethane (1.0 mL) containing 2,6-lutidine (0.5 mL), and the mixture was kept for 3 days at room temperature. The

mixture was poured into cold water (20 mL) and extracted with ether. The ethereal layer was washed with water (10 mL), M silver nitrate (10 mL), and finally with water (10 mL × 4), and dried over sodium sulfate. The extract was evaporated under diminished pressure and the residue was dried over sodium hydroxide in a vacuum desiccator. The oily product (450 mg) failed to crystallize. N.m.r. ($CDCl_3$): δ, 1.32 (9H, s, *tert*-butyl), 1.83 (3H, s, orthoacetyl), 2.04 and 2.09 (each 3H, s, $COCH_3$), 3.77 (3H, s, CO_2CH_3). Other signals between 4–6 p.p.m. (H-1–H-5) were not assigned.

*Preparation of methyl (2,3,4-tri-*O*-acetyl-1-*O*-acyl-β-*D*-glucopyranosyl)uronates (compounds* **2–8**). — Methyl 3,4-di-*O*-acetyl-α-D-glucopyranuronate 1,2-(*tert*-butyl orthoacetate) (**1**, 450 mg, 1.2 mmol) and a carboxylic acid (2 mmol) in chlorobenzene (10 mL) were boiled for 15–30 min under reflux. The mixture was then cooled in an ice-bath. When an excess of the carboxylic acid crystallized, it was removed by filtration. The chlorobenzene was then evaporated off under diminished pressure, and the residue was crystallized from ethanol to give the 1-*O*-acyl products (compounds **2–8**) as colorless needles, yields, about 60–70%.

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