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METHANOLYSIS AS A MODEL REACTION FOR OLIGOSACCHARIDE SYNTHESIS OF SOME 6-SUBSTITUTED 2,3,4-TRI-*O*-BENZYL-D-GALACTOPYRANOSYL DERIVATIVES

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ABSTRACT

In the methanolysis of some 2,3,4-tri-*O*-benzyl-D-galactopyranosyl derivatives, the nature of the C-6 substituent appears to have minor influence on the steric outcome of the reaction compared with the effects of solvent and the nature of the leaving group at C-1. Different combinations of experimental parameters have given methyl glycosides mixtures, the compositions of which vary between 0–70% of α-anomer. Attempts to prepare oligosaccharides from 6-*O*-substituted 2,3,4-tri-*O*-benzyl-D-galactopyranosyl derivatives have shown methanolysis to be a less satisfactory model reaction than is the case for the D-glucose series, even when the alcohol and glycosyl derivative are in near equivalent quantities.

INTRODUCTION

In a systematic study of glycoside-forming reactions in progress in this laboratory[1], it has been shown that the steric outcome of reactions between equivalent amounts of glycosyl derivative having a nonparticipating group at C-2 and an alcohol may be controlled by judicious choice of leaving group, solvent, and substituents, especially at C-6. It is also known that substituents at other sites in the molecule affect the reaction[2–5]. With this knowledge, it has been possible to prepare α-(1→6) linked D-glucose oligomers[6–8] of very high purity, and in order to extend these results to the D-galactose series, we have studied the methanolysis of some 6-*O*-substituted 2,3,4-tri-*O*-benzyl-D-galactopyranosyl derivatives.

RESULTS AND DISCUSSION

D-Galactosyl derivatives show unexpected differences from their D-glucosyl analogs under the methanolysis conditions described herein. These differences can only be ascribed to the influence of the epimeric change at C-4 on the rest of the molecule. Reactions were always performed on an equivalent quantity of alcohol, usually in acetonitrile or ether. These solvents were found to be the limiting

cases for high yields of one or the other anomer in the D-glucose series[6]. In some cases, solvents with higher dielectric constants were employed: 1,2-Ethanediol carbonate ($\varepsilon = 95$) was added to reactions in acetonitrile, and hexamethylphosphoric triamide[9] (HMPT, $\varepsilon = 30$) was also used alone. With the *p*-tolylsulfonyloxy group as the leaving group at C-1, methanolysis gave mixtures of anomers which contained at least 30% of the minor product with all the solvents and C-6 substituents investigated. In contrast, some 1-*O*-tosyl-D-glucopyranose derivatives that give good yields of α anomers after reaction with alcohols are known, and in one case the yield of α anomer was 100% when ether was the solvent[6].

TABLE I

METHANOLYSIS OF SOME DERIVATIVES OF 2,3,4-TRI-*O*-BENZYL-D-GALACTOPYRANOSE[a]

C-6 Substituent	*Solvent*	*Yield* (%)	α *Anomer* (%)
C-1 Tosyloxy leaving group[b]			
Benzyl	Acetonitrile	93	64
Benzyl	Ether	89	48
N-Phenylcarbamoyl	Acetonitrile	75	57
N-Phenylcarbamoyl	Ether	73	44
N-Phenylcarbamoyl	Dichloromethane	98	43
N-Phenylcarbamoyl	Tetrahydrofuran	84	35
N-(*p*-Methoxyphenyl)carbamoyl	Acetonitrile	93	67
N-(*p*-Methoxyphenyl)carbamoyl	Ether	94	42
N-(*p*-Methoxyphenyl)carbamoyl	HMPT	100	50
N-(*p*-Methoxyphenyl)carbamoyl	1:1 Acetonitrile–1,2-ethanediol carbonate	97	63
p-Nitrobenzoyl	Acetonitrile	97	68
p-Nitrobenzoyl	Ether	87	29
p-Nitrobenzoyl	7:1 Acetonitrile–1,2-ethanediol carbonate	98	58
3-Acetylpropionyl	Acetonitrile	82	66
3-Acetylpropionyl	Ether	97	33
C-1 Brosyloxy leaving group[b]			
N-(*p*-Methoxyphenyl)carbamoyl	Acetonitrile	86	47
N-(*p*-Methoxyphenyl)carbamoyl	Ether	85	25
C-1 Trifluoroxy leaving group[c]			
Benzyl	Ether	100	26
p-Nitrobenzoyl	Ether	87	22
N-(*p*-Methoxyphenyl)carbamoyl	Ether	76	≈0
N-(*p*-Methoxyphenyl)carbamoyl	Ether[d]	90	≈0
N-(*p*-Methoxyphenyl)carbamoyl	Toluene	73	26
N-(*p*-Methoxyphenyl)carbamoyl	Dichloromethane	93	15
N-(*p*-Methoxyphenyl)carbamoyl	1,2-Dimethoxyethane	94	32
N-(*p*-Methoxyphenyl)carbamoyl	4:1 Ether–Dichloromethane[d]	97	5
N-(*p*-Methoxyphenyl)carbamoyl	4:1 Ether–1,2-dimethoxyethane[d]	92	16
3-Acetylpropionyl	Ether	92	12.5

[a]The ratio of methanol to glycosyl derivative was 1:1. [b]Reactions performed overnight at room temperature. [c]Reactions performed for 1 h at −78°. [d]Reaction performed for 3 h at −90°.

The results for the D-galactose series are reported in Table I. Ratios of α to β anomers formed were determined from the p.m.r. spectra of the crude products of reaction, based on the areas of the methoxyl signals, as previously described[10]. Yields were also determined from these spectra[11].

It is of interest that the solvent dependence in the two series is also different. In the D-glucose series, ether gave a greater ratio of α to β anomer than did acetonitrile[6], whereas in the D-galactose series this solvent effect was reversed, and a higher ratio of α to β anomer was observed with acetonitrile. Performing the reaction in mixtures of acetonitrile and 1,2-ethanediol carbonate, or in HMPT conferred no advantage. Indeed, an absence of any stereo-selectivity was noted in the latter case.

It appears that the nature or reactivity of the ion pairs derived from 2,3,4-tri-*O*-benzyl-1-*O*-tosyl-D-galactopyranose derivatives is significantly different from that of the corresponding D-glucose series. (Fig. 1). Apparently the rate of equilibration

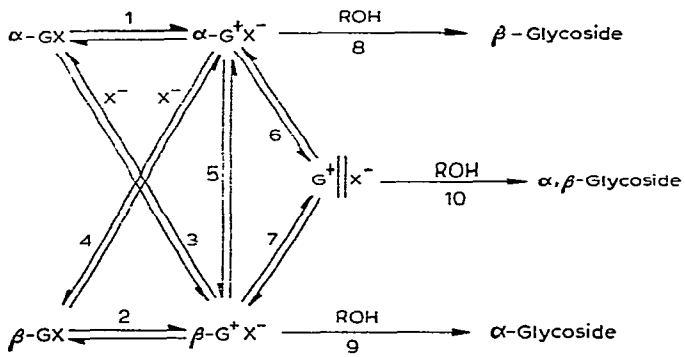

Fig. 1. Generalized mechanism of glycoside synthesis.

between α and β ion pairs is relatively low in the D-galactose series, and perhaps the position of equilibrium is less favorable. The presence of 15% of the β anomer of the tosyl derivative has been observed in ether solutions of 2,3,4-tri-*O*-benzyl-6-*O*-(*N*-phenylcarbamoyl)-1-*O*-tosyl-D-glucopyranose[6]. The use of acetonitrile perhaps enhances the rate of equilibration, and thus allows the more reactive β ion-pair to form more α isomer, but the competing rates in the system are too similar to give high stereoselectivities. Since HMPT is a highly ionizing solvent, complete separation of the ion pair may occur before alcoholysis, and thus no steric control by neighboring ion is possible.

Experiments with the *p*-bromophenylsulfonyl (brosyl) group, which was expected to lead to higher ratios of α to β anomer (owing to enhancement of ionization by stabilization of the incipient negative charge in the ion pair, and consequent increase in the rate of equilibration between the α and β ion-pairs), in fact gave lower ratios than the corresponding tosyl derivatives. The solvent effect, as observed with ether and acetonitrile, was the same however, greater amounts of the α anomer

being observed with the latter solvent. Apparently the brosyloxy group has not been used as a C-1 leaving group previously.

It was speculated that a 3-acetylpropionyl (levulinyl) group[12] at O-6 might yield interesting results if the carbonyl moiety of the acetyl group influences the ion pair at C-1 by through-space effects. The 3-acetylpropionyl group would also function well as a temporary blocking group on O-6 in any oligosaccharide synthesis, as it is simply removed by hydrazine hydrate at pH 7. However, the results followed the same trend as that of the other substituents.

The trifluoromethylsulfonyloxy (triflyloxy) group was first reported as a C-1 leaving group for D-glucopyranosyl derivatives by Kronzer and Schuerch[11]. In the D-galactose series, remarkably different results have been obtained with this group. Under the most favorable conditions with preformed triflyl derivatives, yields approaching 100% were obtained for methyl 2,3,4-tri-*O*-benzyl-6-*O*-[*N*-(*p*-methoxyphenyl)carbamoyl]-β-D-galactopyranoside (see Table I). This contrasts with a yield of 97% for an α-linked product in an analogous experiment in the D-glucose series[11] with *N*-phenyl-carbamate as the C-6 substituent. [The change of substituent on C-6 was necessitated by the poor solubility of 2,3,4,-tri-*O*-benzyl-6-*O*-(*N*-phenylcarbamoyl)-α-D-galactopyranosyl bromide in ether at the temperature of the reaction]. Recently, Maradufu and Perlin[13] have prepared and isolated D-glucose derivatives with brosyloxy and triflyloxy substituents on C-4, and these authors have prepared by substitution of these leaving groups various substituted methyl galactopyranosides for testing as substrates of D-galactose oxidase.

Other leaving groups, such as nitrate and perchlorate, were also investigated, but preliminary experiments were not encouraging. Rates were very low, a major drawback, and the reactions showed no useful stereochemical advantage.

An attempt to prepare a β-linked disaccharide from 2,3,4-tri-*O*-benzyl-6-*O*-[*N*-(*p*-methoxyphenyl)carbamoyl]-α-D-galactopyranosyl bromide and methyl 2,3,4-tri-*O*-benzyl-α-D-galactopyranoside *via* a triflyl intermediate gave products having the approximate ratio of β to α anomer of 3:1, as determined from the ^{13}C n.m.r. spectrum of the products, after purification by preparative t.l.c. The signals were partially assigned by comparison with the products of a parallel preparation with a tosyl intermediate, where the product was expected, on the basis of the results of methanolysis, to be richer in the α-linked disaccharide. The signal at 104.4 p.p.m. (relative to tetramethylsilane) was assigned to the β-C-1 atom of the disaccharidic link, and that at 100.6 p.p.m. to the corresponding α-C-1 resonance. The signal due to the α-C-1 of the methyl glycoside appeared at 99.15 p.p.m. In a corresponding mixture of (1→6)-linked disaccharides in the D-glucose series[8], the glycosyl α-C-1 signal was upfield from the α-C-1 methoxyl signal. Presumably the change at C-4 in the aglycon accounts for this difference.

The poor correlation between the results of methanolysis and those of the reaction with a primary alcohol group is disappointing in view of the good agreement found in the D-glucose series[6–8]. This sugar is, of course, unique in possessing all equatorial

hydroxyl groups, and in cases where hindrance by axial substituents is possible, some extra assistance may be required to achieve good stereoselectivity.

EXPERIMENTAL

General. — N.m.r. spectra were determined with Varian A-60-A or XL-100 spectrometers on solutions in chloroform-*d* and with tetramethylsilane as internal standard. Optical rotations were recorded with a Perkin–Elmer 141 automatic polarimeter. Melting points were determined with a Meltemp instrument and are uncorrected. Microanalyses were performed by Galbraith Laboratories, Inc.

Materials. — Spectrograde dichloromethane and acetonitrile were dried with calcium hydride, ether with sodium wire. 1,2-Ethanediol carbonate (Jefferson Chemical Co., Inc., Houston, Texas 77052) was distilled[14] at pressures above 72 torr and stored in a dark bottle. Silver *p*-toluenesulfonate (Eastman Organic Chemical, Rochester, New York 14650) and silver trifluoromethanesulfonate (Willow Brook Labs., Inc., Waukesha, Wisconsin 53186) were dried *in vacuo* in the dark before use. Sodium *p*-bromobenzenesulfonate was converted into the silver salt by treatment with silver nitrate in aqueous solution; the precipitate was washed and stored *in vacuo*, protected from light.

1,6-Di-*O*-acyl-2,3,4-tri-*O*-benzyl-D-galactopyranoses were prepared by the method of Fréchet and Schuerch[10] from 2,3,4-tri-*O*-benzyl-D-galactopyranose[15]. The latter compound was obtained by benzylation of 2,3,4-tri-*O*-acetyl-1,6-anhydro-β-D-glucose[16], followed by acetolysis and deacetylation to give a syrup which, in our hands, failed to crystallize from ether–petroleum ether but had an optical rotation in agreement with the literature[15]. The properties of the 1,6-di-*O*-acyl derivatives are reported in Table II.

*Methyl 2,3,4-tri-*O*-benzyl-α-D-galactopyranoside.* — Detritylation of methyl 2,3,4-tri-*O*-benzyl-6-*O*-trityl-α-D-galactopyranoside[15] with hydrogen bromide in glacial acetic acid by the conventional procedure gave, after chromatography on silicic acid, the required product as a syrup that crystallized after two months, yield 32%, $[\alpha]_D^{25}$ +59.4° (*c* 3.37, chloroform).

Anal. Calc. for $C_{28}H_{32}O_6$: C, 72.39; H, 6.74. Found: C, 72.42; H, 6.96.

*Preparation of 6-*O*-substituted 2,3,4-tri-*O*-benzyl-D-galactopyranosyl derivatives and reaction with methanol.* — 6-*O*-Substituted 2,3,4-tri-*O*-benzyl-D-galactopyranosyl bromides were prepared by the method of Kronzer and Schuerch[11] from the corresponding 1,6-di-*O*-acyl derivatives, or in one case from 2,3,4,6-tetra-*O*-benzyl-1-*O*-(*N*-phenylcarbamoyl)-α-D-galactopyranose[11]. The products were obtained as syrups by removal of solvent, thoroughly dried under high vacuum, and used immediately to prepare the tosyl, brosyl, or triflyl derivatives on a high vacuum line, as described by Eby and Schuerch[6], in a glass apparatus consisting of two chambers separated by a fritted glass filter. The D-galactosyl derivatives were formed in one arm in acetonitrile, for the tosyl and brosyl derivatives, and in ether for the triflyl derivatives. In the first case, the acetonitrile could be distilled off and replaced by a second solvent. One

TABLE II

PROPERTIES OF 1,6-DI-*O*-ACYL-2,3,4-TRI-*O*-BENZYL-D-GALACTOPYRANOSES

Substituent	*Solvent of crystallization*[a]	*Crystalline anomeric form*	*Yield* (%)	*M.p.* (°)	$[\alpha]_D^{25}$ (°)[b]	*Elemental analysis*[c]		
						C	*H*	*N*
N-(*p*-Methoxyphenyl)carbamoyl	1	*β*	32–38	204–5	+1	68.97 68.77	5.92 5.92	3.75 3.90
N-Phenylcarbamoyl	2	*β*	35	158–9	−5	71.49 71.69	5.85 5.79	4.07 4.18
p-Nitrobenzoyl	2,3	*β*	33	151–2	−6.8	65.77 65.81	4.85 4.76	3.75 3.61
3-Acetylpropionyl		mixture (α to *β* = 1:2)	95	syrup		68.71 68.78	6.54 6.77	

[a](1) Dichloromethane–ethanol, (2) ethanol, (3) ethyl acetate–pentane. [b]In chloroform. [c]First line, calculated results, second line, found results.

equivalent of methanol was added, and the mixture was filtered into the second chamber. In the case of the triflyl derivatives, this step was dispensed with for convenience, as the reactions were performed at −78° or below. The reactions were allowed to proceed overnight, or in the case of the triflyl derivatives for 1 h. (The time allowed for preformation of the tosyl, brosyl, and triflyl derivatives was 15 min. Precipitation of silver bromide then appeared to be complete). The reaction mixtures were processed by conventional techniques, and the crude products examined by p.m.r. The yields and ratios of α to β anomers were calculated (see Table I).

Preparation of disaccharide mixtures. — The procedure just outlined was followed, with the modification that methyl 2,3,4-tri-*O*-benzyl-α-D-galactopyranoside was dried in the second chamber, and the D-galactosyl derivatives were added to this compound when they had formed in the usual way. The products were purified by preparative t.l.c. on glass plates coated with silica gel (E. Merck G_F-254), developed with ether as solvent. The ratios of products were determined from ^{13}C n.m.r. spectra by a technique similar to that used with the proton spectra. These results may have less quantitative accuracy than the results from the proton spectra, owing to the nuclear Overhauser effect. The mixture obtained with the triflyl derivatives had a ratio of α to β anomer of 1:3, that with the tosyl derivative of 3:1.

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