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Synthesis of 1-Glyceritol D-Galactopyranosides

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The so called *iso*floridoside, which has been isolated from *Porphyra umbilicalis* and shown to be an O-α-D-galactopyranosyl-(1 → 1)-glyceritol, has now been reinvestigated. It has been found to be an isomorphous mixture of the diastereoisomeric D- and L-glyceritol derivatives of the structure given above. These compounds and the corresponding β-D-galactopyranosides have been synthesised from glyceritol derivatives of known configuration; the α-glycosides were also isomorphous. The reaction between 2,3-O-*iso*propylidene-D-glyceritol and tetra-O-acetyl-α-D-galactopyranosyl bromide in the presence of silver oxide unexpectedly gave a mixture of D- and L-glyceritol derivatives. In one case a Koenigs-Knorr synthesis using silver oxide gave an unusually high yield of the α-anomer. The infrared spectra of the 1-glyceritol D-galactopyranosides are recorded.

A glyceritol glycoside (glycoside A) found in the red algae *Polysiphonia fastigiata*[1] and *Corallina officinalis*[2] was shown in a preceding paper[2] to be an O-α-D-galactopyranosyl-(1 → 6)-O-β-D-galactopyranosyl-(1 → 1)-glyceritol. This compound was first found in wheat flour lipids by Carter *et al.*[3] who showed it to have the above structure. The occurrence of this glycoside in plant materials of such widely different origin gives greater interest to the possible biochemical role of the glyceritol glycosides in red algae. Those previously found are: O-α-D-galactopyranosyl-(1 → 2)-glyceritol (floridoside)[4–7] O-α-D-mannopyranosyl-(1 → 3)-O-α-D-galactopyranosyl-(1 → 2)-glyceritol [8] and O-α-D-galactopyranosyl-(1 → 1)-glyceritol (*iso*floridoside) [9]. In *iso*floridoside and glycoside A the glyceritol is asymmetrically substituted, and as a possible relationship between the two compounds might have been revealed by the configuration of the glyceritol residues, the synthesis of the four possible 1-glyceritol D-galactopyranosides * was undertaken to provide material for comparison with the natural compounds. As a result of this work the glyceritol residue of glycoside A has been shown to have the D-configuration [2]; the investigation of *iso*floridoside and the synthetic work will be described below.

In an attempt to prepare 1-D-glyceritol β-galactoside, 2,3-O-*iso*propylidene-D-glyceritol (I), which is easily available from 1,2:5,6-di-O-*iso*propylidene-

* For convenience, O-α-D-galactopyranosyl-(1 → 1)-D-glyceritol will be referred to in the following as 1-D-glyceritol α-galactoside *etc.*

D-mannitol[10–12], was allowed to react with tetra-O-acetyl-α-D-galactopyranosyl bromide following Reynolds' and Evans' modification [13] of the Koenigs-Knorr method, but neither the intermediate products nor the expected 1-D-glyceritol β-galactoside crystallised spontaneously. It was thought that the α-anomer of this glycoside might be identical with *iso*floridoside; the hexaacetate of the presumed 1-D-glyceritol β-galactoside was therefore treated with titanium tetrachloride in chloroform [14] to effect anomerisation. Residual β-galactoside in the product after deacetylation was hydrolysed with a β-galactosidase preparation and the carbohydrate mixture obtained was separated on a carbon column. One fraction was chromatographically similar to *iso*floridoside * and crystallised upon seeding with it, but the melting point **, 121—127°, raised to 125—127° after repeated recrystallisations, was much lower than that previously reported for *iso*floridoside [9] (134—135° uncorr.) and the mixed melting point showed a wide range but no depression. However, the melting point of analytically pure *iso*floridoside, as redetermined on a Kofler hot stage, was 132—141°, and a reasonable explanation compatible with the experimental data was thus that both samples were isomorphous mixtures of 1-D- and 1-L-glyceritol α-galactosides. This assumption was confirmed by resolving the mixtures on a carbon column which had previously been found to give very sharp separations. Both *iso*floridoside and the synthetic sample gave the same two components, m. p. *ca* 131—133° and 149—151.5° (*cf.* Table 1); no melting point depression was observed with mechanically mixed samples of these compounds but the mixed fusion technique gave a melting point minimum of 120°.

Table 1. 1-Glyceritol galactopyranosides.

	M. p.	$[\alpha]_D^{20}$	Analysis			
			Per-iodate[a]	Formic acid[b]	C	H
Calculated for $C_9H_{18}O_8$:			3.00	1.00	42.5	7.14
Compound:						
O-α-D-Galactopyranosyl-(1→1)-D-glyceritol	150 —152°	+155°	2.91	0.93	42.8	7.37
O-α-D-Galactopyranosyl-(1→1)-L-glyceritol	131.5—133°	+159°	2.90	0.95	42.9	7.45
O-β-D-Galactopyranosyl-(1→1)-D-glyceritol	140.5—141.5°	−7°	2.89	0.97		
O-β-D-Galactopyranosyl-(1→1)-L-glyceritol[c]	104—107°	+2°	2.89	0.97	42.4	7.45

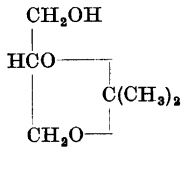
[a] Moles of sodium metaperiodate consumed overnight by 1 mole of glycoside [27].
[b] Moles of formic acid formed from 1 mole of glycoside on periodate oxidation overnight.
[c] The central parts of crystals in a solidified sample melted at 108—110°.

* Kindly supplied by Dr. B. Lindberg.
** Melting points are corrected unless stated otherwise.

The 1-glyceritol β-galactoside preparation obtained above crystallised when seeded with 1-D-glyceritol β-galactoside * from wheat flour lipids [2,3], m. p. 139.5—142°, but the product had m. p. 124—128°, and this value was not appreciably raised by recrystallisation. The low specific rotation, $[\alpha]_D^{20}$ -3°, excludes the presence of α-D-galactopyranosides, and as no melting point depression was observed on admixture with the high-melting glycoside used for seeding, the synthetic preparation was assumed to be an isomorphous mixture of 1-D- and 1-L-glyceritol β-galactosides.

The results show, either that the 2,3-O-*iso*propylidene-D-glyceritol used as starting material in the Koenigs-Knorr reaction was extensively racemised before the reaction or that racemisation took place during the reaction. The first alternative seems very improbable as two different preparations of 2,3-O-*iso*propylidene-D-glyceritol had $[\alpha]_D^{23}$ +14.0° and 14.1°. On the other hand the racemisation could be due to traces of acid liberated in the Koenigs-Knorr reaction and the experiment was therefore repeated using a large excess of silver oxide. However, after alkaline and mild acid hydrolysis and subsequent fractionation of the product on a small carbon column, crystalline preparations were obtained which melted over the ranges 97—116° and 124—129°, indicating that extensive racemisation still took place. This phenomenon remains to be explained.

The synthesis of the 1-D-glyceritol α- and β-galactosides was finally accomplished in the following way. 2,2'-O-Methylene-bis-(3-O-benzoyl-D-glycerose)[15], obtained by oxidising II with lead tetraacetate, was reduced catalytically to the crystalline glyceritol derivative (IV) and this was allowed to react with tetra-O-acetyl-α-D-galactopyranosyl bromide under Reynolds' and Evans'



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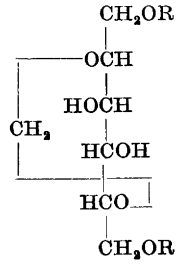
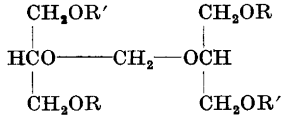


II R = C_6H_5CO
III R = Tetra-O-acetyl-α,β-D-galactopyranosyl



IV R = C_6H_5CO; R' = H
V R = C_6H_5CO; R' = Tetra-O-acetyl-α,β-D-galactopyranosyl
VI R = H; R' = $(C_6H_5)_3C$
VII R = Tetra-O-acetyl-α,β-D-galactopyranosyl; R' = H

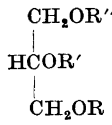
* Kindly supplied by Dr. Carter.

conditions, giving V. After acetolysis and alkaline alcoholysis the product was separated on a carbon column and then gave 17 % of the α- and 36 % of the β-galactoside. As this yield of α-anomer is unexpectedly high the experiment was repeated adding the halide much more rapidly to see whether this would influence the yields. These were now *ca.* 3 % and 28 % for the α- and β-anomer, respectively. The yield of α-galactoside was thus still fairly high.

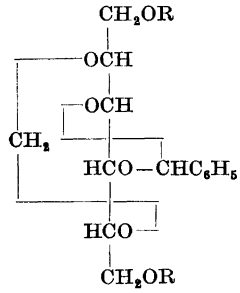
Mixed fusion confirmed the identity of 1-D-glyceritol α-galactoside and the high-melting component of *iso*floridoside.

The anomalous course of the above reaction is probably not due to simple steric hindrance in the aglycone, as a much more hindered molecule, 2,2'-O-methylene-bis-(3-O-trityl-L-glyceritol) (VI), under similar conditions gave 21 % of the theoretical yield of 1-L-glyceritol β-galactoside only; no α-anomer was isolated. VI was prepared from IV by tritylation and debenzoylation.

1-L-Glyceritol β-galactoside was more readily prepared starting from 2,3-O-*iso*propylidene-D-glyceritol (I). Benzylation of I and acid hydrolysis of the benzyl ether gave VIII [16,17] witch was tritylated and the trityl ether (IX) was benzylated to give crystalline 1-O-trityl-2,3-di-O-benzyl-L-glyceritol (X). This was detritylated by heating with aqueous acetic acid and the crude amorphous 2,3-di-O-benzyl-L-glyceritol (XI) obtained was used directly in the Koenigs-Knorr reaction without distillation. The product from this reaction (XII) was deacetylated, the benzyl groups were removed by catalytic hydrogenolysis and the glycoside mixture was separated by recrystallisation and fractionation on a carbon column, giving 1 % and 59 %, respectively,



VIII R,R′ = H; R″ = $C_6H_5CH_2$
IX R = $(C_6H_5)_3C$; R′ = H; R″ = $C_6H_5CH_2$
X R = $(C_6H_5)_3C$; R′, R″ = $C_6H_5CH_2$
XI R = H; R′, R″ = $C_6H_5CH_2$
XII R = Tetra-O-acetyl-α,β-D-galactopyranosyl; R′, R″ = $C_6H_5CH_2$



XIII R = H
XIV R = C_6H_5CO
XV R = Tetra-O-acetyl-α,β-D-galactopyranosyl

of 1-L-glyceritol α- and β-galactosides. The α-anomer had a lower melting point (127—129°) than the low-melting component of *iso*floridoside (m. p. 131.5—133°), but the two samples were shown to be identical by mixed fusion. If the synthetic sample had contained considerable amounts of the isomorphous 1-D-glyceritol α-galactoside a melting point minimum would have been observed, but the presence of a small quantity of this diastereoisomer could not be excluded. This would imply that the 1-L-glyceritol β-galactoside was contaminated with some of the corresponding D-glyceritol derivative, and the rather broad range of the melting point (*ca.* 101—107°) lent some support to this idea. However, chromatography on an efficient carbon column showed that this was most probably not the case. The material was slightly hygroscopic, and a pure sample after intensive drying had m. p. 104—107°. On mixed fusion with 1-D-glyceritol β-galactoside (m. p. 140.5—141.5°) it gave a melting point minimum of 98—99°.

Another possible route to the 1-L-glyceritol galactosides was also considered. 2,5-O-methylene-3,4-O-benzylidene-D-mannitol (XIII), prepared from its easily available dibenzoate (XIV)[15], was allowed to react with tetra-O-acetyl-α-D-galactopyranosyl bromide. This would be expected to give the digalacto-

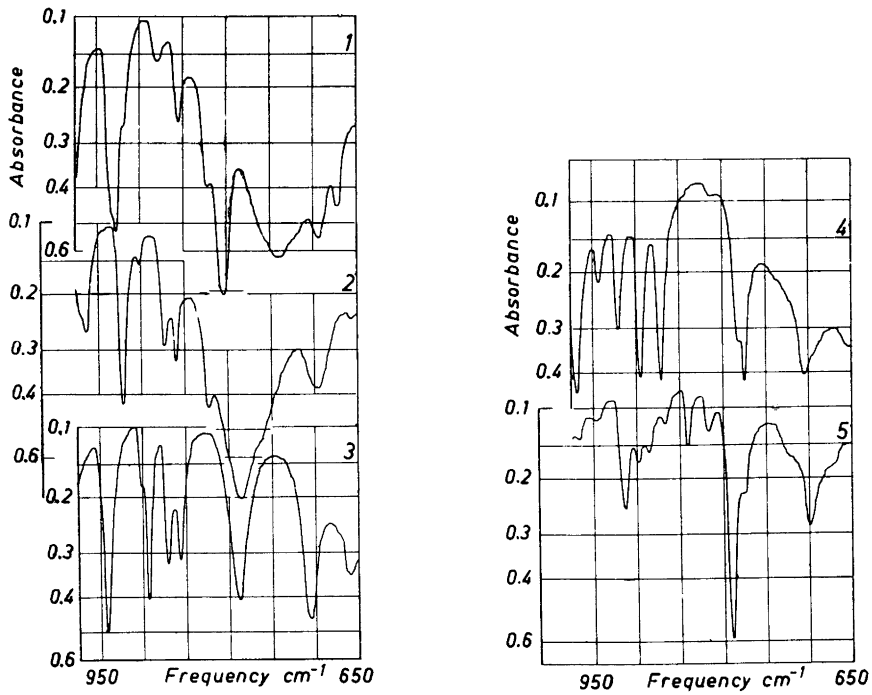


Fig. 1. Infrared absorption curves of 1) O-α-D-Galactopyranosyl-(1 → 1)-D-glyceritol; 2) O-α-D-Galactopyranosyl-(1→1)-L-glyceritol; 3) O-β-D-Galactopyranosyl-(1→1)-D-glyceritol; 4) O-β-D-Galactopyranosyl-(1→1)-L-glyceritol; 5) O-α-D-Galactopyranosyl-(1→6)-O-β-D-galactopyranosyl-(1→1)-D-glyceritol.

side octaacetates (XV) which by catalytic hydrogenolysis to III, lead tetraacetate oxidation and catalytic hydrogenation to VII and subsequent hydrolysis would ultimately give the desired 1-L-glyceritol galactosides. However, while XIII rapidly consumed the theoretical volume of hydrogen in the presence of palladium on charcoal, the product that should have contained XV was not susceptible to hydrogenolysis under similar conditions. Raney nickel catalyst gave some reaction as was evident from the change in optical rotation, but the product finally obtained by following the scheme above was a rather complex mixture. Chromatography on a carbon column gave fractions with the chromatographic properties of a glyceritol galactoside but could not be induced to crystallise. It is possible that some ketal migration analogous to that observed with the 2,3-O-*iso*propylidene-D-glyceritol above takes place in this Koenigs-Knorr reaction.

Infrared spectra of glycoside A and of the 1-glyceritol galactosides were recorded with a Perkin-Elmer double beam spectrometer Model 21 with a sodium chloride prism, using the potassium bromide disc technique. Previous studies of the infrared spectra of carbohydrates have been confined to the frequency range 730—960 cm^{-1} (Ref.[18]); the corresponding sections of the spectra recorded here are shown in Fig. 1. The frequencies of the absorption bands in this region are given in Table 2. Barker *et al.*[19] made the following assignments for some of the characteristic infrared bands of D-galactopyranose derivatives (Nujol mull): Type 2 a, 825 ± 11 cm^{-1}, deformation mode of equatorial hydrogen at the anomeric carbon atom of α-anomers; type 2 b, 895 ± 9 cm^{-1}, deformation mode of axial hydrogen at the anomeric carbon atom of β-anomers; type 2 c, 871 ± 7 cm, deformation mode of equatorial hydrogen at carbon atom 4. The spectra now recorded agree with these interpretations. It is known that with compounds which take up water of crystallisation, the spectra may gradually change as the potassium bromide discs are stored [20]. However, no hydrates of the 1-glyceritol glycosides have been found, and no spectral changes were observed.

Table 2. Infrared bands of 1-glyceritol glycosides in the region 730—960 cm^{-1}. Frequencies * in cm^{-1}.

Frequencies assigned[18]:			Type 2a 825 ± 11		Type 2c 871 ± 7		Type 2b 895 ± 9			
Compound:										
O-α-D-Galactopyranosyl-(1→1)-D-glyceritol	740	802	821	854	879			(918)	927	(932)
O-α-D-Galactopyranosyl-(1→1)-L-glyceritol		783	821	863	871			900	920	
O-β-D-Galactopyranosyl-(1→1)-D-glyceritol		786		855	869		892	(900)	940	
O-β-D-Galactopyranosyl-(1→1)-L-glyceritol	775	(781)	(815)		871		895	921	944	
O-α-D-Galactopyranosyl-(1→6)-O-β-D-Galactopyranosyl-1→1)-D-glyceritol	(775)	789	815	840	866	884	896	914	945	

* Points of inflexion are given in brackets.

EXPERIMENTAL

*2,3-O-*Iso*propylidene-*D*-glyceritol (I).* 2,3-O-*Iso*propylidene-D-glycerose, b. p.$_{20}$ 55°, was prepared from D-mannitol by the method of Baer and Fischer [10,11]. The aldehyde (23 g), which had polymerised to some extent, was dissolved in ethanol and this solution was added with shaking to potassium borohydride (7 g) in 0.2 M disodium phosphate solution (150 ml). After standing overnight the mixture was acidified to pH *ca.* 6 by adding acetic acid and when the evolution of hydrogen had stopped the solution was made alkaline with sodium hydroxide and extracted repeatedly with chloroform. The extract was dried over potassium carbonate and concentrated. The residual oil was distilled through a short Vigreux column. Yield 18.9 g (81 %), b. p.$_{14}$ 82—83°, $[\alpha]_D^{23}$ +14.1° (pure oil), Baer and Kates report [12] $[\alpha]_D^{20}$ +14.0°.

Attempted preparation of 1-D-*glyceritol β-galactoside from I.* 2,3-O-*Iso*propylidene-D-glyceritol (I) (9.1 g), Drierite (100 g), freshly prepared silver oxide (25 g) and dry chloroform (100 ml) were stirred together in the dark for 1 h. A solution of acetobromogalactose (31 g) and iodine (4 g) in chloroform (120 ml) was gradually added with stirring during the next 2 h, and the mixture was then kept with stirring overnight. After filtration the chloroform solution was washed with water, dried over magnesium sulphate and concentrated under reduced pressure to a syrup (*ca.* 36 g). Paper chromatograms run in ethyl ether—dimethyl sulphoxide [21] showed one spot with R_F *ca.* 0.8 which appeared slowly with the silver nitrate—sodium hydroxide reagent and two spots with R_F *ca.* 0.2 and 0.4 which appeared quickly with this reagent. By countercurrent extraction between ethyl ether and dimethyl sulphoxide containing 10 % of water using 5 separating funnels [21], the fastest component was obtained in a chromatographically pure state (14.2 g from 27 g of the crude product). It did not crystallise and was obviously a mixture of diastereoisomers, consisting of 2,3-O-*iso*propylidene-D-glyceritol β-galactoside tetraacetate and the corresponding L-glyceritol derivative. Later a sample was deacetylated with sodium ethoxide in ethanol and hydrolysed (0.02 N sulphuric acid, 20°, 17 h) to give a syrup which crystallised from methanol—ethanol only when seeded with 1-D-glyceritol β-galactoside; m. p. 124—128°, undepressed on admixture with the material used for seeding, $[\alpha]_D^{20}$ −3° (water, c = 2.0). *Cf.* Table 1.

Part of the crude acetate (9 g) from the Koenigs-Knorr reaction was deacetylated with sodium ethoxide in ethanol, a small quantity of water was added and the solution was saturated with carbon dioxide, filtered and concentrated. On paper chromatograms run in ethyl acetate—acetic acid—water (3:1:1) * and then sprayed with periodate—benzidine [22], the deacetylated material gave a rather strong and somewhat trailing spot with R_F *ca.* 0.7. Some much slower spots were also observed. The deacetylated material was dissolved in water and added to a carbon—Celite mixture (100 g + 100 g) contained in a Büchner funnel. On batch-wise elution with aqueous ethanol (1, 2, 4, 6, 8, 10, 15, 25, 35 and 50 %, 500 ml of each) a fraction (2.8 g of dry material) was obtained which on paper chromatograms gave one main spot with R_F 0.7. It did not crystallise, but was assumed to be a mixture of *2,3-O-*iso*propylidene-*D*-glyceritol β-galactoside* and the corresponding L-glyceritol derivative, as mild hydrolysis (0.02 N sulphuric acid at 20° overnight) gave a homogeneous product (paper chromatographically) which when seeded with 1-D-glyceritol β-galactoside (from wheat flour lipids) gave crystals from methanol—ethanol having m. p. 120—124° and $[\alpha]_D^{18}$ −3° (water, c = 2.3). *Cf.* values for pure 1-D- and 1-L-glyceritol-galactosides, Table 1. On paper chromatograms run in ethyl acetate—acetic acid—water the 1-glyceritol β-galactosides travel with the same rate and slightly faster than galactose.

In a second run a mixture of I (0.72 g), silver oxide (4 g), Drierite (10 g) and chloroform (10 ml) was stirred for 1 h and then a solution of acetobromogalactose (2.5 g) and iodine (0.3 g) in chloroform (12 ml) was added with stirring over a period of 3 h. After stirring overnight the chloroform solution was washed with water, dried over magnesium sulphate and concentrated. The residue was deacetylated with sodium ethoxide in ethanol, and then hydrolysed for 30 min at 50° in 0.1 N hydrochloric acid. After treatment with Amberlite resin IR 4B and concentrating, the aqueous solution was added to a

* This solvent system was used throughout unless stated otherwise.

carbon—Celite column (1:1, 24 × 3 cm). On gradient elution with aqueous ethanol (2 000 ml), the concentration of which was raised linearly from 1 to 30 %, material with low optical rotation and with the paper chromatographic properties of a 1-glyceritol β-galactoside was obtained as an apparently homogeneous fraction. The first and last halves of this were collected and worked up separately. The first gave material (0.5 g) which crystallised from methanol—ethanol when seeded with 1-D-glyceritol β-galactoside. Yield 0.39 g; m. p. 124—129°, $[\alpha]_D^{18}$ −4° (water, c = 2.4). The second half (0.4 g) similarly gave 0.20 g, m. p. 110—116°, $[\alpha]_D^{18}$ −3° (water, c = 2.1). It is thus evident that some separation of the 1-D- and 1-L-glyceritol β-galactosides was achieved.

Anomerisation of D,L-glyceritol β-galactoside [14]. Purified 2,3-O-*iso*propylidene-D,L-glyceritol β-galactoside tetraacetate (4.0 g) from the preparation described above was refluxed for 20 min with 2 N sulphuric acid (2 ml) in 60 % aqueous ethanol (100 ml) to remove the *iso*propylidene group. The solution was treated with Amberlite resin IR 4B and concentrated to dryness under reduced pressure. The residue so obtained was acetylated with acetic anhydride and pyridine to give 1-D,L-glyceritol β-galactoside hexaacetate (3.9 g). This was dissolved in dry chloroform (100 ml), titanium tetrachloride (1.2 ml) was slowly added and the mixture was refluxed for 20 h with the exclusion of moisture. Pyridine (4 ml) and an excess of water were then added, the mixture was filtered with Celite and the chloroform phase was washed with sodium carbonate and water, dried over magnesium sulphate and concentrated under reduced pressure. To deacetylate the residual syrup (3.8 g) it was dissolved in absolute ethanol (100 ml) to which sodium (0.2 g) had previously been added and was then left at room temperature overnight. The solution obtained on dilution with water (100 ml), was filtered through Amberlite resins IR 120 H and IR 4B, concentrated under reduced pressure to a small volume, treated with decolorising carbon, filtered and concentrated to give the mixed α- and β-galactosides as a syrup (1.75 g).

To hydrolyse the remaining β-galactoside the syrup was incubated at 30° with a β-galactosidase solution [2,23] (5 ml) in 0.1 N sodium acetate buffer (pH 4.6, 100 ml). When the optical rotation of the solution had reached a constant value (after 60 h) acetone (120 ml) was added and after centrifugation the solution was further purified by precipitation with aqueous lead acetate, filtration with Celite, saturation with hydrogen sulphide and filtration. The solution obtained was concentrated under reduced pressure to a small volume (30 ml) and was then added to a carbon—Celite column (1:1, 24 × 3 cm). Gradient elution with aqueous ethanol (1—30 %, 2 000 ml) gave three separate fractions which were shown by paper chromatography to contain glyceritol, galactose and 1-glyceritol galactosides, respectively. The glycoside fraction crystallised from a methanol—ethanol mixture on seeding with *iso*floridoside (m. p. 132—141°, $[\alpha]_D^{21}$ +157°). Yield 0.51 g; m. p. 121—127°, $[\alpha]_D^{21}$ +157° (water, c = 2.0). After three further recrystallisations the product (*ca.* 0.2 g) had m. p. 125—137°, undepressed on admixture with *iso*floridoside.

The synthetic glycoside mixture and its concentrated mother liquors were combined (0.6 g in total), dissolved in water (10 ml) and the solution was added to the top of an efficient carbon—Celite column (1:1, 43 × 3.5 cm). Aqueous ethanol (4 000 ml), the concentration of which was raised linearly from 3 % to 6 %, was used as eluant. The optical rotation of the eluate showed two strong peaks which were only partly separated. The optically active part of the eluate was cut into 3 fractions. Fraction 1, corresponding to the first peak and fraction 2, corresponding to the second peak, were separated at the point of minimum rotation; the "tail" of the slowest component was collected separately to give fraction 3. As some overlapping could be expected, fraction 2 was run through the column again in the same way. However, there were no indications of the presence of any material from fraction 1 in the eluate.

Fraction 1 was concentrated to dryness and the residue (0.24 g) was recrystallised from methanol—ethanol. Yield 0.21 g; m. p. 131.5—133°, $[\alpha]_D^{20}$ +159° (water, c = 1.9). The properties did not change on further recrystallisation (analysis: see Table 1).

Fraction 2 similarly gave crystalline material (0.15 g), m. p. 149—151.5°, $[\alpha]_D^{20}$ +152° (water, c = 2.0).

Fraction 3 (35 mg) gave a small crop of crystals with m. p. 143—149°.

Fractionation of iso*floridoside on carbon.* A sample of *iso*floridoside [9] (0.25 g), m. p. 132—141°, $[\alpha]_D$ +157° from which the low-melting component had been partially removed by extensive recrystallisation, was fractionated in the same way as the synthetic α-galactosides and then gave fraction 1′ (0.06 g), m. p. 130.5—132°, $[\alpha]_D^{18}$ +155° (water, c = 1.9) and fraction 2′ (0.12 g), m. p. 149.5—151.5°, $[\alpha]_D^{18}$ +156° (water, c = 1.7).

As the melting point of mechanically mixed samples of the high- and low-melting glycoside showed no depression, but a very wide range (*ca.* 130—150°), further comparisons were made by the mixed fusion technique [24] using a Kofler hot stage. The samples to be compared were placed on each side of and close to a *ca.* 1 mm wide splinter of a cover glass which was placed on the glass slide of the hot stage. Heating was applied gradually until the molten material spreading from each side under the cover glass had met, if possible so that a few crystals of the most high-melting component were left. If necessary a seed crystal was added. The temperature was then adjusted so that the crystals grew at a maximum rate and the process was followed by polarised light. When all had solidified the new melting points were determined.

On mixed fusion of fraction 1′ and 2′ the crystallisation of the latter was found to be fairly fast up to the boundary between the samples. The zone of mixing was passed only very slowly, but then the rate of the crystal growth increased again and when all had solidified the crystals appeared perfectly homogeneous. On renewed heating a narrow channel indicating the zone of mixing formed at *ca.* 120°; fraction 1′ melted at 129—132° and fraction 2′ at 149—151°. When compared in the same way, 1-L-glyceritol α-galactoside (m. p. 127—129°; from 2,3-di-O-benzyl-L-glyceritol, XI) and fraction 1 and 1′ behaved as one compound, though the melting points of the solidified samples were slightly different, 126—129°, 131—134° and 129—132°, respectively. 1-D-Glyceritol α-galactoside (m. p. 150—152°; from 2,2′-O-methylene-bis-(3-O-benzoyl-D-glyceritol), (IV) was in the same way found to be identical with fraction 2 and 2′; the melting points of the solidified samples were 150—152°, 149—151° and 149—151°, respectively.

*2,2′-O-Methylene-bis-(3-O-benzoyl-*D*-glyceritol) (IV).* 1,6-Di-O-benzoyl-2,5-O-methylene-D-mannitol (II) was prepared from D-mannitol by the procedure of Ness *et al.*[15] As the synthesis was carried out on about 25 times the scale used by these authors, special care was taken to avoid overheating; the overall yield of a crude product, m. p. 117—121°, was 69 %. A sample was recrystallised from methanol, m. p. 119.5—121°, $[\alpha]_D^{22}$ −66° (chloroform, c = 2.0).

Finely powdered lead tetraacetate (44 g) was added slowly with stirring to a solution of II (40 g) in dry ethyl acetate (300 ml) and after standing for an hour the precipitated lead salts were filtered off. The filtrate was washed successively with water, aqueous sodium bicarbonate and water, and was then dried for a short time over sodium sulphate, filtered and concentrated to a smaller volume (*ca.* 100 ml). Carefully washed Raney nickel W-2 catalyst [25] (*ca.* 10 ml) was added and the mixture was hydrogenated at 70° and 130 atm pressure for 3 h. When the cooled hydrogenation vessel was opened the product had partly crystallised. It was brought into solution by gentle heating, the solution was filtered and an equal volume of light petroleum (b. p. 45—65°) was added to the combined filtrate and washings. After the mixture had been kept cold for a while the crystals (28 g, m. p. 104.5—106°) were filtered off, the mother liquors were concentrated under reduced pressure and the residue was recrystallised from carbon tetrachloride, giving a second crop (4 g, m. p. 101—105°). The combined material was recrystallised from chloroform—carbon tetrachloride. Yield 30 g (75 %), m. p. 105—106.5° $[\alpha]_D^{20}$ −58° (chloroform, c = 2.0). After recrystallisation from aqueous ethanol a sample had m. p. 106.5—108° and $[\alpha]_D^{18}$ −60° (chloroform, c = 2.3). (Found: C 61.9; H 6.00. Calc. for $C_{21}H_{24}O_8$: C 62.4; H 5.98.)

In order to test the stability of IV to silver oxide, a sample (m. p. 105—107°; 80 mg) was shaken with freshly prepared silver oxide (200 mg) in chloroform (4 ml) for 120 h at room temperature. The specific rotation of the samples remained unchanged and afterwards the filtered chloroform solution, when concentrated, gave a crystalline residue with m. p. 103—106° without further purification. Thus no significant racemisation took place.

*1-*D*-Glyceritol α- and β-galactosides.* A mixture of 2,2′-O-methylene-bis-(3-O-benzoyl-D-glyceritol) (IV) (10 g), freshly prepared silver oxide (17 g), Drierite (75 g) and dry chloro-

form (150 ml) was stirred for 1 h and then a solution of acetobromogalactose (22.5 g) and iodine (3 g) in chloroform (100 ml) was added dropwise with stirring during 2 h. Care was taken to protect the reaction mixture from light. After 2 days in a dark place, inorganic material was removed by filtration, the filtrate was washed with water, dried over magnesium sulphate and concentrated under reduced pressure. To split the methylenedioxy-bridge of V, the residual syrup was dissolved in a mixture of acetic anhydride (70 ml), acetic acid (30 ml) and sulphuric acid (1 ml). After 1 h at room temperature anhydrous sodium acetate (4 g) and water (100 ml) were added and when the anhydride had decomposed, the mixture was diluted with more water and extracted with chloroform. The chloroform solution was washed with aqueous sodium carbonate and water, dried over magnesium sulphate and concentrated. The residue was treated with a solution of sodium (4 g) in absolute ethanol (600 ml) at room temperature overnight. Water (600 ml) was then added, the solution was filtered through Amberlite resins IR 120 H and IR 4B and the filtrate was concentrated under reduced pressure. When dissolved in methanol (10 ml), the residue crystallised spontaneously but the product (7.3 g) had m. p. 121—136° and $[\alpha]_D^{21}$ +38° (water, c = 2.0) and was probably a mixture of the α- and β-galactosides. Recrystallisation from methanol gave 1-D-glyceritol β-galactoside (2.1 g) m. p. 138—140°, $[\alpha]_D^{21}$ ±0 (water, c = 2.0), and 1-D-glyceritol-α-galactoside (0.2 g), m. p. 150—152°, $[\alpha]_D^{18}$ +155° (water, c = 1.8), (analysis: See Table 1.)

The combined mother liquors were concentrated, the residue was dissolved in water and the solution was added to the top of a carbon—Celite column (1:1, 50 × 5 cm). Gradient elution with aqueous ethanol (2—8 %) gave two fractions containing material with the R_F value of a 1-glyceritol galactoside. The fractions were not completely separated, as judged by spot tests, but the first of them was easily distinguished by its high positive optical rotation. On concentration and recrystallisation of the residue from methanol—ethanol this fraction gave 1-D-glyceritol α-galactoside (1.9 g), m. p. 150—151.5°. The next fraction similarly gave 1-D-glyceritol β-galactoside (2.4 g), m. p. 138.5—140.5°. A sample was recrystallised from methanol and then had m. p. 140.5—141.5°, $[\alpha]_D^{20}$ −7° (water, c = 2.0), (analysis: See Table 1.)

The total yield of the α- and β-anomer was thus 2.1 g and 4.5 g or 17 % and 36 % calculated on IV, respectively.

The synthesis was repeated on half the scale described above; one quarter of the acetobromogalactose solution was added at once and the rest during *ca.* 40 min. The yield of α-anomer (m. p. 150—152°) and β-anomer (m. p. 137—140°) was now only 3 % and 28 % of the theoretical, respectively.

2,2'-O-Methylene-bis-(3-O-trityl-L-glyceritol) (VI). A solution of 2,2'-O-methylene-bis-(3-O-benzoyl-D-glyceritol) (IV) (8.1 g) and trityl chloride (13 g) in dry pyridine (60 ml) was kept at 100° for 4 h and was then poured into ice-water. The mixture was extracted with ether and the ether phase was washed successively with water, sodium bicarbonate and water and was then dried over sodium sulphate. After filtration the ether solution was concentrated and the remaining amorphous *2,2'-O-methylene-bis-(1-O-trityl-3-O-benzoyl-D-glyceritol)* was dissolved in ethanol (50 ml) containing potassium hydroxide (6 g). After heating on a water bath for 1 h the mixture was diluted with water and extracted with chloroform. The chloroform extract was washed with water, dried over sodium sulphate and concentrated under reduced pressure and the residue was recrystallised from carbon tetrachloride. Yield 11.3 g (83 %); m. p. 142—146°. A pure sample obtained by recrystallisation from an ethanol—ethyl acetate mixture had m. p. 146—147.5° and $[\alpha]_D^{18}$ +58° (chloroform, c = 2.0). (Found: C 79.0; H 6.39. Calc. for $C_{45}H_{44}O_6$: C 79.4; H 6.51.)

1-L-Glyceritol β-galactoside from VI. A mixture of 2,2'-O-methylene-bis-(3-O-trityl-L-glyceritol) (VI) m. p. 142—146°; 5.0 g), freshly prepared silver oxide (7.4 g), Drierite (29 g) and dry chloroform (40 ml) was stirred for 1 h and then a solution of acetobromogalactose (6.7 g) and iodine (0.9 g) in chloroform (30 ml) was added with stirring during 3 hours. The mixture was kept with stirring in a dark place until a sample of the chloroform solution no longer gave a positive bromide test with silver nitrate in ethanol (12 days).

The reaction product was isolated and subjected to acetolysis and deacetylation, essentially as described above for V in the preparation of the 1-D-glyceritol galactosides. Most of the tritanol which was liberated in the acetolysis, could be filtered off after the excess of acetic anhydride had been destroyed by the addition of water.

The deacetylated material gave strong spots for galactose and glyceritol on paper chromatograms. It was separated on a carbon—Celite column (1:1, 50 × 5 cm) by the gradient elution technique. Aqueous ethanol (1—8 %, 8 000 ml) was used as eluant and the eluate was investigated polarimetrically and by paper chromatography. One fraction (1.0 g) had the chromatographic properties of a 1-glyceritol galactoside and crystallised from a methanol—ethanol mixture when seeded with 1-D-glyceritol β-galactoside. Yield 0.78 g, (21 % calc. on VI), m. p. 97—100°. After recrystallisation from the same solvent it still had m. p. 97—100° and $[\alpha]_D^{18}$ +1° (water, c = 2.5). These values are somewhat lower than those observed with the same compound prepared from 2,3-di-O-benzyl-L-glyceritol (see Table I).

1-L-Glyceritol α-galactoside travels a little faster than the corresponding β-galactoside on carbon columns, but when the part of the eluate which would contain the α-galactoside was concentrated, the residue (0.16 g) had a low specific rotation ($[\alpha]_D$ +16°) and could not be induced to crystallise.

3-O-Benzyl-L-glyceritol (VIII) has previously been prepared by treating the sodium salt of 2,3-O-*iso*propylidene-D-glyceritol (I) with benzyl bromide and then removing the *iso*propylidene group by mild hydrolysis [16,17]. The benzylation procedure of Zemplén *et al.*[26] was used in the present investigation and seems to give a higher yield.

A mixture of 2,3-O-*iso*propylidene-D-glyceritol (18.9 g), finely powdered potassium hydroxide (115 g) and dioxane (250 ml) was placed in a three-necked flask equipped with a mechanical stirrer, a reflux condenser and a dropping funnel. The flask was immersed in an oil bath which was kept at 80° and freshly distilled benzyl chloride (100 ml) was added through the funnel over a period of 45 min with vigorous stirring. The reaction mixture was kept at 80—90° with stirring overnight. After cooling it was filtered, the filter cake was dissolved in water and the solution was extracted with benzene. The dioxane solution and the benzene extract were combined, dried over magnesium sulphate, filtered and concentrated under reduced pressure (to *ca.* 100 ml). This residue was distilled *in vacuo*, giving a fore-run (54 g) of unreacted benzyl chloride, b. $p._{13}$ 65—66° and then a fraction (43 g), b. $p._{0.3}$ 90—96° and α_D^{21} +13.7° (1 dm, pure oil), containing *1-O-benzyl-2,3-O-*iso*propylidene-D-glyceritol.* In a pure state this compound has b. $p._{0.3}$ 95—97° and α_D +17.8° (1 dm, pure oil) [16]; the theoretical yield should be only 31.8 g. The crude product was probably contaminated with dibenzyl ether, but it was used in the next step without further purification.

The crude benzyl ether obtained above (43 g) was dissolved in methanol (300 ml). Water (100 ml) and 2 N sulphuric acid (20 ml) were added and the mixture was heated to the boiling point and then left at room temperature overnight. The methanolic solution was extracted with five 50 ml portions of light petroleum (b. p. 45—65°) to remove any dibenzyl ether; the light petroleum extracts were washed successively with two 50 ml portions of 80 % aqueous methanol to retain any VIII carried away. The combined methanolic solutions were concentrated under reduced pressure to remove most of the methanol and the residual aqueous solution (*ca.* 150 ml) was extracted with chloroform. The chloroform extract was washed with aqueous sodium carbonate and water, dried over sodium sulphate, filtered and concentrated under reduced pressure and the residual syrup (25.4 g) was distilled *in vacuo* through a Vigreux column. A small fore-run (1.7 g) b. $p._{0.15}$ 123°, n_D^{21} 1.5450, was discarded; VIII then distilled at 128—133° and ca 0.05 mm pressure. Yield 23.1 g (88 % calc. on I; n_D^{21} 1.5322, α_D^{21} +6.6° (1 dm, pure oil). Sowden and Fischer [16] give b. $p._{0.3}$ 138—139°, n_D^{16} 1.5342, α_D +6.1° (1 dm, pure oil).

1-O-Trityl-2,3-di-O-benzyl-L-glyceritol (X). A solution of 3-O-benzyl-L-glyceritol (VIII) (23.1 g), and trityl chloride (35.4 g) in dry pyridine (150 ml) was kept at room temperature for 2 days and then heated at 60° for 4 h. It was then stirred into ice-water and the aqueous mixture was extracted with ether. The ether phase was washed several times with water, then as quickly as possible with cold 0.5 N sulphuric acid and finally with aqueous sodium carbonate and water. After drying over sodium sulphate the ether-

eal solution was filtered and concentrated under reduced pressure to give crude *1-O-trityl-3-O-benzyl-*L*-glyceritol* (IX) as a syrup which, however, did not crystallise.

Freshly distilled benzyl chloride (60 ml) was added gradually with vigorous stirring to a mixture of the crude trityl compound and finely powdered potassium hydroxide (50 g) in dioxane (150 ml), while the temperature was kept at 80°. Stirring was continued overnight at this temperature. The insoluble material was then filtered off, the filter cake was dissolved in water and the aqueous suspension obtained was extracted with benzene and the benzene extract was added to the residue remaining after the dioxane filtrate had been concentrated under reduced pressure. The benzene solution was washed with sodium bicarbonate and water, dried over sodium sulphate, filtered, concentrated and finally taken to dryness at *ca.* 0.3 mm pressure and 150° (bath temperature). The residue crystallised when stirred with light petroleum. The crude product was ground to a paste with light petroleum—*iso*propyl ether (1:1, 50 ml), filtered and then recrystallised from benzene—ethanol. After recovering a few grams from the mother liquors, the yield of pure X was 52 g (80 % calc. on VIII); m. p. 84.5—86°, $[\alpha]_D^{18}$ −9° (chloroform, c = 2.5). (Found: C 83.6; H 6.69. Calc. for $C_{36}H_{34}O_3$: C 84.0; H 6.66.)

*2,3-Di-O-benzyl-*L*-glyceritol (XI).* A solution of 1-O-trityl-2,3-di-O-benzyl-L-glyceritol (X) (48,8 g) in acetic acid (340 ml) was heated under reflux and water (240 ml) was added at a moderate rate so that no oil separated. The mixture was refluxed for a further hour and then kept in a refrigerator overnight. Tritanol (23.5 g or 95 %) was filtered off and the mother liquors were concentrated under reduced pressure to give crude XI as a colourless oil (26.7 g). After drying azeotropically with benzene, it was used in the next step without further purification. A sample was distilled *in vacuo*. It had b.p.$_{0.05}$ *ca.* 150°, n_D^{25} 1.5490, d_4^{23} 1.106 and $[\alpha]_D^{23}$ +3.8° (pure oil).

*1-*L*-Glyceritol α- and β-galactosides from XI.* A mixture of crude, dry 2,3-di-O-benzyl-L-glyceritol XI (11 g), freshly prepared silver oxide (19 g), Drierite (50 g) and dry chloroform (100 ml) was stirred for 1 h and then a solution of acetobromogalactose (18 g) and iodine (2.4 g) in dry chloroform (100 ml) was added during 3 h. The reaction mixture was shielded from light. After stirring for a further 20 h a sample of the chloroform solution gave no reaction with ethanolic silver nitrate solution. Insoluble material was then filtered off, the filtrate was washed with water, dried over sodium sulphate and concentrated under reduced pressure. The residue was treated overnight with absolute ethanol (500 ml) to which sodium (0.75 g) had been added, the alkali was neutralised by adding 2 N sulphuric acid (16 ml) and after filtration the ethanolic solution was concentrated to a small volume. 2 N sodium carbonate solution (5 ml) was added to the residue and the di-O-benzylglyceritol galactosides were extracted with three 200 ml portions of ethyl acetate. The extracts were filtered successively through a cellulose column (10 × 3.5 cm), pre-equilibrated with water-saturated ethyl acetate, and then concentrated under reduced pressure. The amorphous residue (16 g) was free from reducing sugars. It was dissolved in methanol (200 ml) and hydrogenolysed in the presence of palladium on charcoal catalyst (2 g) containing 10 % of Pd. The uptake of hydrogen was 2 000 ml in 4 h; theory requires 1 940 ml. After filtration the methanolic solution was concentrated under reduced pressure and the residue was dissolved in a small quantity of water. Some crystalline material (0.2 g, m. p. 80—94°), probably triphenylmethane, was filtered off and the filtrate was concentrated under reduced pressure. The residual syrup was dissolved in methanol—ethanol and seeded with 1-D-glyceritol β-galactoside, giving crystalline 1-L-glyceritol β-galactoside (5.0 g), m. p. 101—103°, $[\alpha]_D^{20}$ +2° (water, c = 3.0).

The residue (3.0 g) obtained on concentration of the mother liquors was separated on a carbon—Celite column (50 × 5 cm) as described previously, using aqueous ethanol (2—8 %, 8 000 ml) as the eluant. Glyceritol was present in the first part of the eluate as indicated on paper chromatograms. After a large empty fraction there followed a fraction with a low positive optical activity and immediately after this another which gave a spot for a 1-glyceritol galactoside on paper chromatograms but was optically inactive.

The optically active fraction was concentrated under reduced pressure giving a residue (0.12 g) which crystallised from methanol—ethanol. Recrystallisation from the same solvent gave 1-L-glyceritol α-galactoside (70 mg), m. p. 127—129°, $[\alpha]_D^{18}$ +156° (water, c = 1.8). As described above, this compound was shown by mixed fusion to be identical with the low-melting component of *iso*floridoside.

The residue (1.5 g) from the next fraction gave 1-L-glyceritol β-galactoside (1.0 g), m. p. 102—107°, $[\alpha]_D^{20}$ +2° (water, c = 2.0). The total yield of this compound was thus 6.0 g (59 %). The melting points of various preparations were somewhat irregular and a sample (400 mg) was therefore added to an efficient carbon—Celite column (1:1, 43 × 3.5 cm) and then eluted with aqueous ethanol (3.5—6.5 %, 4 000 ml). 0.5 ml aliquots of the eluate were oxidised with sodium metaperiodate and the excess of oxidant was determined iodometrically [27] after 4 h. The periodate consumption showed only one peak; the two successive halves of the corresponding part of the eluate were worked up separately giving two crystalline preparations which both had m. p. 102.5—105°. They were combined and recrystallised but the melting point was unchanged except that a few crystals remained up to 107°. However, when a sample was dried intensively and then immediately transferred to the hot stage so that contact with atmospheric moisture was avoided, the m. p. was raised to 104—107°; $[\alpha]_D^{18}$ +2° (water, c = 2.0), (analysis: See Table 1.) Mixed fusion with 1-D-glyceritol β-galactoside gave a melting point minimum of 98—99°, the solidified D- and L-glyceritol derivatives melted at 140—141.5° and 105—110°, respectively. About two days were required for the melt to crystallise at 85°; the crystals appeared homogeneous.

1,6-Di-O-benzoyl-2,5-O-methylene-3,4-O-benzylidene-D-mannitol (XIV). Ness *et al.*[15] prepared XIV in 83 % yield by treating 1,6-di-O-benzoyl-2,5-O-methylene-D-mannitol (II) with 5 parts of benzaldehyde in the presence of 1 part of fused zinc chloride at room temperature. This procedure was somewhat modified to facilitate the preparation of XIV on a larger scale.

Powdered anhydrous zinc chloride (110 g) was added with stirring to freshly distilled benzaldehyde (1 100 ml). When all had dissolved the solution was cooled to room temperature and diluted with dry *iso*propyl ether (1 100 ml). 1,6-Di-O-benzoyl-2,5-O-methylene-D-mannitol (225 g) was added with stirring and when completely dissolved the mixture was allowed to stand at room temperature overnight. The benzylidene compound soon started to crystallise. After keeping in a refrigerator for a day the mixture was filtered and the filter cake was carefully washed with water, aqueous sodium bicarbonate and water and was then dried. Yield 180 g; m. p. 153—156°. A further quantity (60 g), m. p. 153—156° was obtained from the mother liquors by washing with water and aqueous sodium bicarbonate, drying over sodium sulphate, concentrating under reduced pressure and recrystallising the residue from ethyl acetate—methanol (1:3). The total yield of XIV was thus 88 %; $[\alpha]_D^{20}$ +59° (chloroform, c = 2.0). Ness *et al.*[15] record m. p. 151—152° and $[\alpha]_D^{20}$ +61.2°.

2,5-O-Methylene-3,4-O-benzylidene-D-mannitol (XIII) was prepared by refluxing its dibenzoate (XIV) (240 g) with a solution of sodium hydroxide (45 g) in ethanol (1 200 ml) and water (500 ml) for 90 min. Most of the ethanol was distilled off under reduced pressure, water (500 ml) was added and after cooling the crystals were filtered off and recrystallised from water. Yield 108 g (82 %); m. p. 129.5—132°. After concentration under reduced pressure the mother liquors gave a second crop of crystals. A sample was recrystallised from ethyl acetate, m. p. 130—132°, $[\alpha]_D^{18}$ +28° (chloroform, c = 2.3). (Found: C 59.7; H 6.49. Calc. for $C_{14}H_{18}O_6$: C 59.6; 6.43.) On treatment of XIII with benzoyl chloride in pyridine the expected dibenzoate (XIV) was regenerated.

Attempted preparation of 1-L-glyceritol β-galactoside from XIII. A mixture of 2,5-O-methylene-3,4-O-benzylidene-D-mannitol (XIII) (1.0 g), freshly prepared silver oxide (2.7 g), Drierite (10 g) and dry chloroform (10 ml) was stirred for 1 h and then a solution of acetobromogalactose (3.4 g) and iodine (0.4 g) in dry chloroform (15 ml) was added during the next 2 h. After the mixture had been stirred overnight the chloroform solution was free from reactive bromide. Insoluble material was filtered off and the filtrate was washed, dried and concentrated in the usual way giving a syrup (3.7 g), presumed to be XV, which was only moderately soluble in ethanol. A very small quantity of crystals, m. p. 235—237°, which separated from an ethanolic solution, gave a positive formaldehyde test with chromotropic acid.

Part of the syrup (1.8 g) was dissolved in ethanol (40 ml) and hydrogenated at room temperature for 87 h in the presence of palladium on charcoal catalyst (0.5 g) containing 10 % of Pd. However, the hydrogen uptake was of the order 2 ml/h and could be attri-

buted to losses by diffusion. The theoretical volume of hydrogen required for removal of a benzylidene group from XV is *ca.* 80 ml. The hydrogenolysis of a comparable amount (0.54 g, 2 mmoles) of XIII using the same catalyst (0.5 g) was complete in 1 h, giving 2,5-O-methylene-D-mannitol [15], m. p. 176—177.5°, $[\alpha]_D^{20}$ −50° (water, c = 2.0). The presence of catalyst poisons seems to be excluded by the fact that subsequent addition of XIII to the hydrogenation mixture containing the product from the Koenigs-Knorr reaction, resulted in a rapid hydrogen uptake. Either the hydrogenolysis of XV is sterically hindered or XIII rearranges before the Koenigs-Knorr reaction takes place, to give a less reactive molecule.

As an experiment showed that XIII could be hydrogenolysed using Raney nickel as the catalyst, the product (3.4 g, from 3.4 mmoles of XIII) from a Koenigs-Knorr reaction, carried out as described above, was dissolved in ethyl acetate (20 ml) and then Raney nickel catalyst W-2[26] (3 ml) was added. Treating the mixture with hydrogen at 95° and 150 atm, the optical rotation of the solution read in a 1 dm tube was +0.76° at the start and then reached a constant value of −0.78° in 24 h. The catalyst was filtered off and lead tetraacetate was gradually added to the filtrate. *Ca.* 1.0 g of oxidant was rapidly consumed; calculated on the amount of XIII used as starting material the theoretical consumption should be 1.5 g. The excess oxidant was destroyed by adding ethylene glycol, the mixture was filtered and the filtrate was washed with water and aqueous sodium bicarbonate, dried over magnesium sulphate and concentrated under reduced pressure. The residue was hydrogenated for 15 h at 75° and 140 atm in the presence of Raney nickel catalyst (1 ml) in ethyl acetate (15 ml). The hydrogenated product (*cf.* VII) was acetylated and deacetylated as described for V in the preparation of 1-D-glyceritol galactosides. The final product (0.66 g) on paper chromatograms gave an elongated spot with about the R_F value of galactose as well as two slow fairly strong spots and traces of faster spots (periodate—benzidine spray [22]). The mixture was separated on a small carbon column in the usual way giving two adjoining fractions (75 mg and 180 mg) which had the chromatographic properties of 1-glyceritol galactosides but could not be induced to crystallise.

Acetylation of pure samples of the 1-glyceritol galactosides described above, using acetic anhydride in pyridine, gave amorphous products. The paper chromatography of the acetates in dimethyl sulphoxide solvent systems has been described in a previous paper [21].

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