# THE SYNTHESIS OF 3-*O*-β-D-XYLOPYRANOSYL-D-XYLOSE AND THE RECHARACTERIZATION OF SOME BENZYLIDENE DERIVATIVES OF D-XYLOSE[1]

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ABSTRACT

The benzylidene groups of the di-*O*,*O*-benzylidene derivative of D-xylose diethyl dithioacetal have been located at the 2,4:3,5 positions, and *not* at the 2,3:4,5 positions suggested by Zinner, Rembarz, Linke, Ulbricht. Partial hydrolysis yielded the 2,4-*O*-benzylidene compound, from which 3-*O*-β-D-xylopyranosyl-D-xylose (rhodymenabiose) was prepared.

## INTRODUCTION

An attempt was made to synthesize 4-*O*-β-D-xylopyranosyl-D-xylose by condensing tri-*O*-acetyl-α-D-xylopyranosyl bromide under standard Koenigs–Knorr conditions with a D-xylose derivative which had all the hydroxyl groups substituted with the exception of that at $C_4$. It was also desired that this substance be in the open-chain form since experiments have indicated (2) that such reactants give greater yields than those in a ring form due to a lessening of steric hindrance in the Koenigs–Knorr reaction. It was decided that such a derivative might be synthesized using as starting material the mono-*O*,*O*-benzylidene-D-xylose diethyl dithioacetal prepared by Zinner *et al.* (1). This was obtained in low yield by the partial hydrolysis of a di-*O*,*O*-benzylidene-D-xylose diethyl dithioacetal formed by condensing D-xylose diethyl dithioacetal with benzaldehyde in the presence of hydrochloric acid. The above workers (1) assigned the 2,3-linkage to the mono-*O*,*O*-benzylidene compound and gave the 2,3:4,5-linkage to the di-*O*,*O*-benzylidene compound.

Accordingly, the crystalline *O*-benzylidene derivative was prepared and was acetylated to yield a crystalline di-*O*-acetyl-mono-*O*,*O*-benzylidene-D-xylose diethyl dithioacetal, which was then demercaptalated with mercuric chloride and cadmium carbonate in dry methanol, yielding a crystalline di-*O*-acetyl-mono-*O*,*O*-benzylidene-D-xylose dimethyl acetal. Deacetylation of the latter compound produced a crystalline mono-*O*,*O*-benzylidene-D-xylose dimethyl acetal, which was then partially benzoylated using one equivalent of benzoyl chloride in pyridine at low temperature. This reaction yielded a crystalline *O*-benzoyl-*O*,*O*-benzylidene-D-xylose dimethyl acetal. From the reaction sequence, this compound should be 5-*O*-benzoyl-2,3-*O*-benzylidene-D-xylose dimethyl acetal, which has all its hydroxyl groups substituted except that at $C_4$, a position which is sterically unhindered owing to the absence of a hemiacetal ring. Hence, it was a suitable intermediate for the desired synthesis of 4-*O*-β-D-xylopyranosyl-D-xylose.

*O*-Benzoyl-*O*,*O*-benzylidene-D-xylose dimethyl acetal was therefore condensed with tri-*O*-acetyl-α-D-xylopyranosyl bromide under standard Koenigs–Knorr conditions (3). The reaction product was deacetylated and debenzoylated by warming it with sodium hydroxide solution, and benzylidene and acetal groups were subsequently removed by hydrolysis with warm dilute sulphuric acid. There resulted a syrup which, from chromatographic examination, consisted mainly of xylose and a disaccharide with an unexpectedly high mobility on chromatograms. The mixture was separated by chromatography on a cellulose column, and the disaccharide component was isolated as a syrup which crystallized. The disaccharide showed on chromatograms a single spot with an unexpectedly

[1]*Manuscript received March 25, 1960.*
*Contribution from the Department of Chemistry, Queen's University, Kingston, Ontario.*

high mobility ($R_{xylose}$ 0.53, solvent A; 0.64, solvent B), whereas the mobility of the expected 4-*O*-β-D-xylopyranosyl-D-xylose* was much lower ($R_{xylose}$ 0.20, solvent A; 0.38, solvent B). The disaccharide was, in fact, shown to be 3-*O*-D-xylopyranosyl-D-xylose, from its oxidation with sodium periodate and the formation of an osazone. The specific rotation ($-35° \rightarrow -22°$ (constant)) showed the compound to be 3-*O*-β-D-xylopyranosyl-D-xylose. 3-*O*-α-D-Xylopyranosyl-D-xylose has a specific rotation of $+106° \rightarrow +118°$ (constant) (5).

Howard (6) has obtained a xylose-containing disaccharide by partial hydrolysis of a xylan produced by *Rhodymenia palmata*. This disaccharide, "rhodymenabiose", has the same physical constants as the synthetic xylobiose, which is now identified as 3-*O*-β-D-xylopyranosyl-D-xylose. The structure of "rhodymenabiose" was tentatively suggested to be 3-*O*-β-D-xylopyranosyl-D-xylose but no attempts were made to characterize it (6).

As the disaccharide obtained in the Koenigs–Knorr synthesis was not that expected, it was decided to re-examine the structure of the *O*-benzoyl-*O*,*O*-benzylidene-D-xylose dimethyl acetal (VII) used in the condensation. Compound VII was methylated using silver oxide and methyl iodide. The compound proved difficult to methylate, but, after three repetitions of the methylation procedure, a crystalline monomethyl ether was isolated. Debenzoylation yielded a crystalline mono-*O*,*O*-benzylidene-*O*-methyl-D-xylose dimethyl acetal which, on acid hydrolysis, was converted into crystalline 3-*O*-methyl-D-xylose (7).

Compounds VI and VII were then treated with toluene-*p*-sulphonyl chloride, and mono-*O*,*O*-benzylidene-di-*O*-tosyl-D-xylose dimethyl acetal and *O*-benzoyl-*O*,*O*-benzylidene-*O*-tosyl-D-xylose dimethyl acetal were formed, respectively. Reaction of the first of these two tosyl derivatives with sodium iodide under standard conditions (8) showed the presence of one primary hydroxyl group, while, under the same conditions, the second compound showed primary hydroxyl groups to be absent.

These data, together with the formation of 3-*O*-methyl-D-xylose, characterize VI as 2,4-*O*-benzylidene-D-xylose dimethyl acetal and VII as 5-*O*-benzoyl-2,4-*O*,*O*-benzylidene-D-xylose dimethyl acetal. Compound VI was not oxidized by sodium metaperiodate under standard conditions (9) and this is consistent with the benzylidene group in the 2,4 position, there being no free hydroxyl groups on adjacent carbon atoms in 2,4-*O*-benzylidene-D-xylose dimethyl acetal. If the benzylidene group were attached in the 2,3 position a consumption of one equivalent of periodate would be expected. Reaction of VII with tri-*O*-acetyl-α-D-xylopyranosyl bromide would therefore be expected to yield a (1 → 3)-linked xylopyranosyl–xylose and not the (1 → 4)-linked disaccharide originally expected in the condensation.

3-*O*-β-D-Xylopyranosyl-D-xylose was obtained in 11% yield, which is less than might be expected from a condensation involving an open-chain sugar (2). However, although the sugar was in the open-chain form, the hydroxyl group involved in the reaction—that at $C_3$—was part of a ring system which involved the benzylidene group, and, thus, reaction was sterically hindered to some extent. That the hydroxyl group at $C_3$ was not very reactive was shown by the difficulties encountered in methylating and tosylating this group.

In order to determine if the mono-*O*,*O*-benzylidene derivatives III, IV, and V had the benzylidene group in the 2,4 position or if the group had wandered during one of the interconversions, the structure of mono-*O*,*O*-benzylidene-D-xylose diethyl dithioacetal

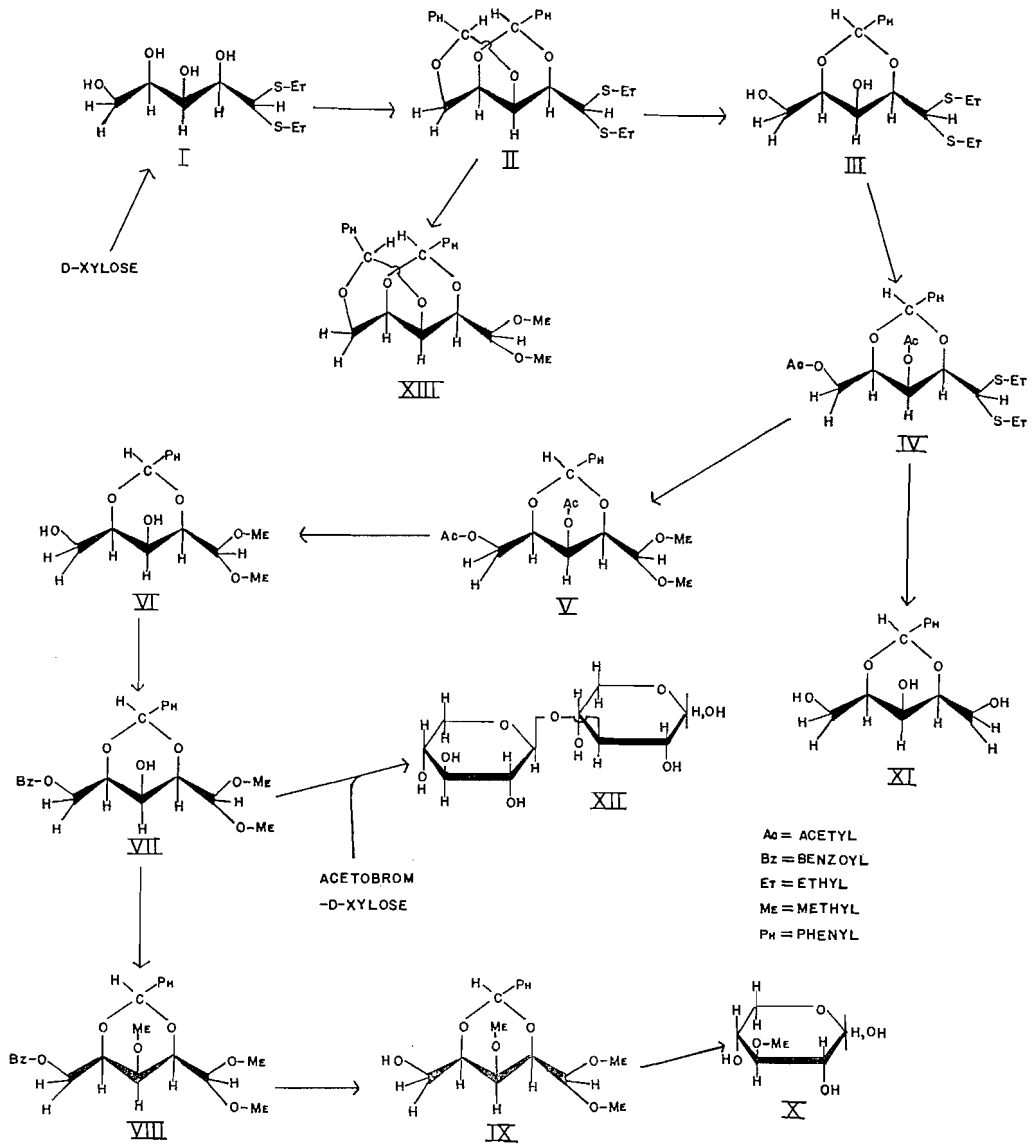
*Prepared by partial hydrolysis of a hemicellulose from aspen (Populus tremuloides) (4).*

(III) was examined. The formulation of this as the 2,3-*O*-benzylidene isomer (Zinner *et al.* (1)) rests upon its oxidation with lead tetraacetate, when one equivalent of the oxidizing agent was consumed and formaldehyde (amount not stated) was liberated. This consumption of oxidant was interpreted to mean that the compound contained hydroxyl groups on adjacent carbon atoms. It is known, however (10, 11), that lead tetraacetate oxidation of thioacetals is an unreliable guide to the presence of hydroxyl groups on adjacent carbon atoms, due to the oxidation of the dithioacetal grouping. In particular, Huebner *et al.* (10) have examined the lead tetraacetate oxidation of the analogous mono-*O,O*-benzylidene derivatives of L-arabinose diethyl dithioacetal. With the fully substituted di-*O*-benzylidene-L-arabinose diethyl dithioacetal, one equivalent of oxidant was consumed rapidly (5 minutes) and a further one equivalent was consumed slowly (2 hours). This consumption they assumed to be due to oxidation of the dithioacetal group—a rapid oxidation of one sulphur atom followed by a slower oxidation of the second. With mono-*O,O*-benzylidene-L-arabinose diethyl dithioacetal, a consumption of two equivalents (rapid) plus two equivalents (slow) of lead tetraacetate per mole was observed, which, it was assumed, was due partially to oxidation of the thioacetal groups (one fast plus one slow) and partially to oxidation of hydroxyl groups on adjacent carbon atoms (also, one fast plus one slow). It would, therefore, seem from these observations that the data reported (1) for the lead tetraacetate oxidation of mono-*O,O*-benzylidene-D-xylose diethyl dithioacetal would support the view that this compound does *not* possess hydroxyl groups on adjacent carbon atoms, rather than the reverse.

By demercaptalation of II, Zinner *et al.* (1) obtained a 2,3,4,5-di-*O*-benzylidene-*aldehydo*-D-xylose which, by heating with methanol, they converted into a crystalline 2,3,4,5-di-*O*-benzylidene-D-xylose methyl hemiacetal. They point out that the physical constants (melting point and specific rotation) of this compound are very similar to those of a 2,3,4,5-di-*O*-benzylidene-L-xylose methyl hemiacetal, which was prepared by lead tetraacetate oxidation of a 1,2,3,4-di-*O*-benzylidene-D-glucitol and subsequent treatment with methanol (Wolfe, Hann, and Hudson (12)). However, although the structures of these L-xylose and D-glucitol derivatives were at first in doubt, di-*O,O*-benzylidene-D-glucitol was later shown by Angyal and Lawler (13) to have the benzylidene groups spanning the 1,3 and 2,4 hydroxyl groups. These workers isolated, by partial hydrolysis, 2,4-*O*-benzylidene-D-glucitol, the structure of which was previously known. It therefore follows that the 2,3,4,5-di-*O*-benzylidene-L-xylose methyl hemiacetal must, in fact, have the 2,4:3,5 structure, and the identity of physical constants of this and di-*O,O*-benzylidene-D-xylose methyl hemiacetal would indicate that the latter compound *too* has the 2,4:3,5 structure.

Finally, to characterize compounds II, III, and IV, di-*O*-acetyl-*O,O*-benzylidene-D-xylose diethyl dithioacetal (IV) was demercaptalated, using mercuric chloride and cadmium carbonate in aqueous acetone (14), to yield di-*O*-acetyl-*O,O*-benzylidene-*aldehydo*-D-xylose. This compound was reduced with sodium borohydride in aqueous methanol, and the resulting alkaline solution was heated on a water bath to complete removal of the acetyl groups. The solution was deionized and evaporated to a crystalline residue which was shown to be the well-characterized, symmetrical 2,4-*O*-benzylidene-xylitol (XI) (15, 16). Compound IV is, therefore, 3,5-di-*O*-acetyl-2,4-*O*-benzylidene-D-xylose diethyl dithioacetal, whereas compounds II and III must be 2,4:3,5-di-*O*-benzylidene-D-xylose diethyl dithioacetal and 2,4-*O*-benzylidene-D-xylose diethyl dithioacetal, respectively.

The real sequence of reactions in the synthesis of the disaccharide was therefore:




Demercaptalation of 2,4:3,5-di-*O*-benzylidene-D-xylose diethyl dithioacetal (II), using mercuric chloride and cadmium carbonate in dry methanol, yielded crystalline 2,4:3,5-di-*O*-benzylidene-D-xylose dimethyl acetal (XIII). This was shown to be identical with the di-*O*,*O*-benzylidene-D-xylose dimethyl acetal prepared by Breddy and Jones (17) by condensing D-xylose with benzaldehyde and methanol in the presence of hydrochloric acid, and used by them for the estimation of xylose. The structure of this compound was not previously known.

Zinner *et al.* (1), in addition to preparing the mono-*O*,*O*-benzylidene (III) and di-*O*,*O*-benzylidene (II) derivatives of D-xylose diethyl dithioacetal, also prepared, in the same

way, a mono-*O,O*-benzylidene-D-xylose dipropyl dithioacetal, and di-*O,O*-benzylidene derivatives of the methyl, propyl, *n*-butyl, isobutyl, and benzyl dithioacetals of D-xylose. These, like II and III, were assigned the 2,3-linkage (in the case of the mono-*O,O*-benzylidene-D-xylose dipropyl dithioacetal) and the 2,3:4,5-linkage (in the case of the di-*O,O*-benzylidene derivatives of the various D-xylose dithioacetals). No attempt has been made to investigate the structures of these derivatives, but it would seem likely that they, like II and III, have the di-*O,O*-benzylidene groups in the 2,4:3,5 positions and the mono-*O,O*-benzylidene group in the 2,4 position.

## EXPERIMENTAL

Optical rotations were measured at 21° ± 2° C and in water unless otherwise stated. Solutions were concentrated under reduced pressure at 40° C. The following solvent systems (v/v) were used to separate sugars on paper chromatograms: (A) 1-butanol:ethanol:water, 3:1:1; (B) ethyl acetate:acetic acid:water, 9:2:2. Sugars were detected with the *p*-anisidine hydrochloride spray (18). Infrared absorptions were measured either as solutions in chloroform or as a powder (mulled in a potassium bromide pellet) on a Perkin–Elmer Model 21 spectrophotometer.

### D-*Xylose Diethyl Dithioacetal* (I)

D-Xylose (100 g), ice-cold ethanethiol (100 g), and ice-cold hydrochloric acid (100 ml) were shaken together in a stoppered flask and the resulting mixture was left overnight at room temperature. The solution was diluted with water (200 ml) and then deionized by passage through Amberlite IR 400 (OH) resin. The eluate was concentrated to a syrup which was diluted with ether (200 ml) and refrigerated. The product crystallized and was filtered and washed with ether. Yield: 140 g (82%), m.p. 63–64° C.

### *2,4:3,5-Di-O-benzylidene*-D-*xylose Diethyl Dithioacetal* (II)

D-Xylose diethyl dithioacetal (50 g) was dissolved in redistilled benzaldehyde (500 ml) with warming and after being cooled to 5° C in ice, dry hydrogen chloride was bubbled through the solution. After 5 minutes a semisolid mass was formed which was diluted with ethanol (400 ml), shaken, and filtered under suction. The crude crystalline product was dried *in vacuo* and recrystallized from boiling ethanol. Yield: 40.6 g (48%), m.p. 181–182° C, $[\alpha]_D$ −36° (*c*, 4.95, chloroform). Literature (1) gives a melting point of 181–182° C and $[\alpha]_D$ −36.7° (*c*, 2.53, chloroform).

### *2,4-O-Benzylidene*-D-*xylose Diethyl Dithioacetal* (III)

The conditions given by Zinner *et al.* (1) (hydrolysis with ethanol (3 parts) plus 80% acetic acid (4 parts) for 1 hour) were found to produce only a small yield of mono-*O,O*-benzylidene compound. After various hydrolyzing mixtures were tried, the following conditions were found to be most satisfactory. A mixture of di-*O,O*-benzylidene-D-xylose diethyl dithioacetal (80 g), acetone (1800 ml), and 80% acetic acid (1200 ml) was refluxed for 3 hours. The resulting solution was cooled to 10° C in ice water, and unhydrolyzed di-*O*-benzylidene material was removed by filtration. The filtrate was concentrated to a small volume and the acid was neutralized with cold potassium hydroxide solution followed by sodium bicarbonate solution. The aqueous solution was extracted with chloroform (3×100 ml) and the combined extracts were washed once with water, dried ($MgSO_4$), and filtered. Evaporation of the filtrate yielded a crystalline residue which was extracted with boiling water. The aqueous extract was cooled and mono-*O,O*-benzylidene-D-xylose diethyl dithioacetal was removed by filtration. Yield: 5.5 g (43% after recovery

of unchanged di-*O*-benzylidene compound). After recrystallization from water and from ether it had a melting point of 146–147° C and $[\alpha]_D$ −22° (*c*, 2.0, methanol). Literature (1) gives a melting point of 147° C and $[\alpha]_D$ −23.3° (*c*, 2.76, methanol).

*3,5-Di-O-acetyl-2,4-O-benzylidene*-D-*xylose Diethyl Dithioacetal* (IV)

2,4-*O*-Benzylidene-D-xylose diethyl dithioacetal (29.4 g) was dissolved in dry pyridine (300 ml), and acetic anhydride (250 ml) was added dropwise to the solution while this was cooled and stirred mechanically. After the addition (1 hour) stirring was continued for 1 hour more and the solution was then left at room temperature (16 hours), after which it was poured with stirring into ice water (1200 ml). The product precipitated from solution and was collected, washed with water, and air-dried. After recrystallization from methanol it weighed 35 g (96%) and had a melting point of 92.5–93° C and $[\alpha]_D$ −2° (*c*, 4.35, chloroform). Anal. Calc. for $C_{20}H_{28}O_6S_2$: S, 14.95. Found: S, 14.96.

*3,5-Di-O-acetyl-2,4-O-benzylidene*-D-*xylose Dimethyl Acetal* (V)

3,5-Di-*O*-acetyl-2,4-*O*-benzylidene-D-xylose diethyl dithioacetal (10 g) was dissolved in dry methanol (300 ml). Cadmium carbonate (90 g) was added to the solution, which was stirred vigorously, and a solution of mercuric chloride (90 g) in dry methanol (100 ml) was added portionwise at room temperature. After the addition (1 hour) stirring was continued for a further 15 hours at room temperature and the mixture was then refluxed (40 minutes). The solution was filtered through a Celite pad and the precipitate was washed with methanol. Filtrate and washings were concentrated to ca. 200 ml and this solution was poured into 500 ml each of chloroform and water. The chloroform layer was extracted with water until chloride-free, dried ($MgSO_4$), filtered, and evaporated to a syrup (9.6 g). This crystallized when allowed to stand in the desiccator. The crude product was recrystallized from methanol. Yield: 6.5 g (76%), m.p. 82.5–83.5° C, $[\alpha]_D$ +34° (*c*, 4.3, chloroform). Anal. Calc. for $C_{18}H_{24}O_8$: C, 58.71; H, 6.52; S, nil. Found: C, 58.94; H, 6.34; S, 0.07.

*2,4-O-Benzylidene*-D-*xylose Dimethyl Acetal* (VI)

3,5-Di-*O*-acetyl-2,4-*O*-benzylidene-D-xylose dimethyl acetal (1.45 g) was dissolved in dry methanol (30 ml) and the resulting solution was cooled to 5° C. Methanolic sodium methoxide (0.88 *N*, 1 ml) was added and the solution was then stored at 7° C (22 hours) in a stoppered flask. The solution was neutralized (glacial acetic acid and then sodium bicarbonate solution) and then evaporated to dryness, to yield a white crystalline residue. This was recrystallized from ether. Yield: 0.90 g (81%), m.p. 142–143° C, $[\alpha]_D$ +32° (*c*, 1, methanol). Infrared examination showed no absorption for carbonyl group (1700–1750 $cm^{-1}$). Anal. Calc. for $C_{14}H_{20}O_6$: C, 59.16; H, 7.04. Found: C, 59.41; H, 7.15.

*5-O-Benzoyl-2,4-O-benzylidene*-D-*xylose Dimethyl Acetal* (VII)

The method used was similar to that described by Zinner *et al.* (19). 2,4-*O*-Benzylidene-D-xylose dimethyl acetal (4.8 g) was dissolved in dry pyridine and the solution was cooled to −15° C and stirred mechanically. A solution of redistilled benzoyl chloride (2.5 g) in dry pyridine (10 ml) was then added dropwise while the temperature was kept below −10° C (40 minutes). The solution was stored at −5° C (4 hours), then at 0° C (16 hours), and finally at room temperature (2 hours), after which the product was precipitated by pouring the solution into ice water (700 ml). The mixture was left at room temperature (2 hours) to complete hydrolysis of excess benzoyl chloride and was then filtered. The precipitate was washed with water and after drying *in vacuo* it weighed 4.5 g (69%). The material was recrystallized from methanol and then had a melting point of 120–121° C

and $[\alpha]_D$ +44° (*c*, 4.06, chloroform). Anal. Calc. for $C_{21}H_{24}O_7$: C, 64.94; H, 6.19. Found: C, 65.21; H, 6.40.

*Condensation of 5-O-Benzoyl-2,4-O-benzylidene-*D*-xylose Dimethyl Acetal with Tri-O-acetyl-α-*D*-xylopyranosyl Bromide*

5-*O*-Benzoyl-2,4-*O*-benzylidene-D-xylose dimethyl acetal (8 g) was dissolved in dry, alcohol-free chloroform (80 ml) and to the solution were added Drierite (10 g) and silver oxide (10 g). The solution was shaken in the dark to dry the reagents. After 1 hour tri-*O*-acetyl-α-D-xylopyranosyl bromide (20) (14.4 g) in dry, alcohol-free chloroform (50 ml) and a few crystals of iodine were added and the shaking was continued at room temperature. After 48 hours a test for ionizable bromine was negative. After a further 48 hours the solution was filtered through a Celite pad, and evaporated to a syrup which was dissolved in a solution of water (4 parts) plus methanol (1 part) (200 ml) containing sodium hydroxide (8 g). The solution was warmed on a water bath at 70° C (4 hours), after which it was cooled and deionized by passage through columns of Amberlite IR 400 (OH) and IRC 50 (H) resins in series; the former resin also removed the bulk of the free xylose. The neutral eluate was concentrated to a syrup (5.4 g) which was dissolved in aqueous *N*/100 sulphuric acid, and the resulting solution was warmed at 70° C (5 hours). The solution was cooled and deionized by passage through Duolite A-4 (OH) resin. Concentration of the eluate yielded a syrup which was shown by paper chromatography (solvent B) to consist, in addition to xylose, of a main component with $R_{xylose}$ 0.64 and smaller components with $R_{xylose}$ 1.23 and 0.38.

The syrup was partially fractionated by chromatography on a column of Dowex 50W resin (94×3.3 cm) (21) and fractionation and concentration of the eluate yielded a syrup (2.3 g) which was enriched with the chromatographically slower-moving components. Part of this (0.9 g) was fractionated by chromatography on a cellulose column (40×3 cm) (22) using as eluent 1-butanol half saturated with water.

*Fraction 1*: A syrup (0.62 g) which chromatographic examination showed to consist mainly of xylose, plus a much smaller component with $R_{xylose}$ 1.23 (solvent B). This latter was presumed to be a partially unhydrolyzed xylose acetal or ester.

*Fraction 2*: A syrup (0.255 g) which crystallized on standing. Chromatography showed a single spot with $R_{xylose}$ 0.53 (solvent A) and 0.64 (solvent B).

*Fraction 3*: A syrup (0.04 g) which chromatographic examination showed to consist of a major component with $R_{xylose}$ 0.26 (solvent A) and 0.38 (solvent B) and a minor component with $R_{xylose}$ 0.53 (solvent A) and 0.64 (solvent B). This fraction was not examined further.

Fraction 2 was recrystallized from methanol and then had a melting point of 192–193° C and $[\alpha]_D$ −35° (4 minutes) → −22° (constant) (*c*, 2.5).

Acid hydrolysis (*N* sulphuric acid at 80° C for 12 hours) showed only xylose to be present. Anal. Calc. for $C_{10}H_{18}O_9$: C, 42.53; H, 6.43. Found: C, 42.71; H, 6.56.

*Periodate Oxidation of Fraction 2* (XII)

Portions (ca. 10 mg) of fraction 2 were oxidized with aqueous sodium metaperiodate using standard procedures, (*a*) in acid pH unbuffered conditions, and (*b*) in solution buffered at pH 8. The periodate uptake was estimated by the method of Neumüller and Vasseur (9) and the formic acid liberation by that of Andrews *et al.* (23).

The molar periodate consumptions per mole of disaccharide were as follows: (*a*) in unbuffered solution, 1.38 (after 14 minutes), 1.75 (39 minutes), 2.36 (1.2 hours), 2.41 (2 hours), 2.78 (4 hours), 2.97 (7.5 hours), and 3.04 (21 hours); (*b*) in solution buffered at

pH 8, 1.48 (after 13 minutes), 2.88 (38 minutes), 7.11 (4 hours), and 8.73 (21 hours). The molar production of formic acid in unbuffered solution was 0.47 (after 39 minutes), 0.58 (1.2 hours), 0.82 (2 hours), 0.99 (4 hours), 1.20 (7.5 hours), and 1.33 (21 hours). Extrapolation of the flat part of the curve to zero time gave values for the experiment in unbuffered solution: uptake of metaperiodate, 2.95 moles per mole of sugar; formic acid production, 1.05 moles per mole of sugar.

The theoretical results for oxidation of a xylobiose in acid pH unbuffered conditions are: for (1 → 2) and (1 → 3)-linked disaccharides, 3 moles of periodate consumed per mole of sugar and 1 mole of formic acid liberated per mole of sugar, and for (1 → 4)- and (1 → 5)-linked disaccharides, 4 moles of periodate per mole and 2 moles of formic acid per mole assuming hydrolysis of formyl ester is slow. The extrapolated values obtained therefore indicated that the glycosidic linkage was either (1 → 2) or (1 → 3), and the overoxidation values showed that it was in fact the latter. There was very little overoxidation in unbuffered solution (only 3.05 moles of periodate were consumed after 21 hours) and this is typical of (1 → 3)-linked disaccharides where initial oxidation produces a formyl ester-containing residue which is stable under the acid pH which results in unbuffered conditions. With (1 → 3)-linked sugars overoxidation is rapid in alkaline solution as the formyl ester is unstable under these conditions, and fraction 2 showed a rapid overoxidation in solution buffered at pH 8. It is known (25) that with (1 → 2)-linked disaccharides overoxidation is rapid even in acid conditions because the residue resulting from initial oxidation does not contain a formyl ester and hence is unstable.

*3-O-β-*D*-Xylopyranosyl-*D*-xylose Phenyl Osazone*

The sugar (50 mg) was dissolved in water (0.5 ml) and acetic acid (0.2 ml) plus phenyl hydrazine (0.15 ml) were added. The solution was warmed at 70° C (1.5 hours) and then cooled and diluted with water (3 ml). An oil separated which crystallized on standing. This was recrystallized from hot water and then had a melting point of 194–196° C and $[\alpha]_D$ +47° (*c*, 1.25, pyridine). Anal. Calc. for $C_{22}H_{28}O_7N_4$: N, 12.18. Found: N, 12.28.

*5-O-Benzoyl-2,4-O-benzylidene-3-O-methyl-*D*-xylose Dimethyl Acetal* (VIII)

5-*O*-Benzoyl-2,4-*O*-benzylidene-D-xylose dimethyl acetal (1.19 g) was dissolved in methyl iodide (25 ml). Silver oxide (2 g), Drierite (2 g), and a few crystals of iodine were added to the solution, which was then shaken in the dark (21 hours) and finally boiled under reflux (5 hours) with exclusion of moisture. The solution was filtered and the residue was washed with acetone. Filtrate and washings were evaporated to dryness, yielding a partially crystalline residue which was finally dried *in vacuo*. It then weighed 1.17 g and, on infrared analysis, showed appreciable amounts of hydroxyl group to be still present. The above methylation procedure was repeated twice (the materials were boiled under reflux for a total time of 70 hours), after which a crystalline product (1.17 g, total yield i.e. recovery, 96%) was obtained which, when subjected to infrared analysis, showed only a small peak corresponding to free hydroxyl group. The product was dissolved in warm ethanol and cooling the resulting solution induced partial crystallization. The crystalline material was filtered, washed with ethanol, and dried. It weighed 0.37 g and had a melting point of 137–138° C. Further cooling of the solution produced a second crop of crystals (0.16 g, m.p. 137–138° C), after which a third crop was obtained which had a melting point of 90–98° C and was presumably a mixture of methylated and nonmethylated materials. The material with a melting point of 137–138° C was combined (0.53 g, 43%) and recrystallized from ethanol. It then had a melting point of 138–139° C and $[\alpha]_D$ +40° (*c*, 2.5, chloroform). Infrared examination showed there to be no peak for

free hydroxyl group present. Anal. Calc. for $C_{22}H_{26}O_7$: OMe, 23.14. Found: OMe, 23.32.

*2,4-O-Benzylidene-3-O-methyl-*D*-xylose Dimethyl Acetal* (IX)

5-*O*-Benzoyl-2,4-*O*-benzylidene-3-*O*-methyl-D-xylose dimethyl acetal (0.34 g) was dissolved in a solution of ethanol (10 ml) plus aqueous *N*/10 sodium hydroxide solution (10 ml) and the materials were then warmed on a water bath at 70° C (2 hours). The solution was cooled and concentrated to dryness and yielded a residue which was dissolved in chloroform (20 ml). The resulting solution was washed twice with water, dried ($MgSO_4$), filtered, and evaporated to dryness. A crystalline residue was obtained (0.22 g, 88%) which after recrystallization from ether had a melting point of 134–135° C and $[\alpha]_D$ +9° (*c*, 1.55, methanol). Anal. Calc. for $C_{15}H_{22}O_6$: OMe, 31.2. Found: OMe, 31.0.

*3-O-Methyl-*D*-xylose* (X)

2,4-*O*-Benzylidene-3-*O*-methyl-D-xylose dimethyl acetal (0.22 g) was added to aqueous *N*/60 sulphuric acid (10 ml) and sufficient methanol was then added to dissolve the material. The solution was warmed at 70° C (5 hours), cooled, and deionized by passage through Duolite A-4 resin (OH form). Concentration of the eluate yielded a colorless syrup (0.12 g 95%), which crystallized completely after nucleation with authentic 3-*O*-methyl-D-xylose. After recrystallization from acetone the material had a melting point of 96° C and mixed melting point with authentic 3-*O*-methyl-D-xylose 95–96° C, $[\alpha]_D$ +57° (4 minutes) → +19° (constant) (*c*, 1.6). Literature gives a melting point of 103–104° C and $[\alpha]_D$ +52.2° → +14.8° (constant) (7), m.p. 95° C, and $[\alpha]_D$ +45° → +19° (constant) (26). The infrared spectra of this material and of authentic 3-*O*-methyl-D-xylose were identical.

*2,4-O-Benzylidene-3,5-di-O-tosyl-*D*-xylose Dimethyl Acetal*

2,4-*O*-Benzylidene-D-xylose dimethyl acetal (1.07 g) was dissolved in dry pyridine (4 ml) and the solution was cooled to 10° C. A solution of toluene-*p*-sulphonyl chloride (1.6 g) in pyridine (3 ml) was then added and the material was left in a stoppered flask at room temperature (2 hours). Excess tosyl chloride was hydrolyzed by the slow dropwise addition of water, and the mixture was then poured into an excess (100 ml) of water. The resulting solution was extracted with chloroform and the combined chloroform extracts were washed successively with solutions of dilute sulphuric acid and sodium bicarbonate, and finally with water. The solution was dried, filtered, and evaporated to a hard glass (1.97 g). Infrared examination of this showed a slight peak for free hydroxyl group so the tosylation procedure was repeated; a colorless glass (1.93 g, 86%) was thus obtained. This showed no peak corresponding to free hydroxyl group on infrared examination, $[\alpha]_D$ +14° (*c*, 3.4, chloroform). Anal. Calc. for $C_{28}H_{32}O_{10}S_2$: S, 10.77. Found: S, 10.80.

*Reaction of 2,4-O-Benzylidene-3,5-di-O-tosyl-*D*-xylose Dimethyl Acetal with Sodium Iodide*

The conditions used were those recommended by Tipson (8). 2,4-*O*-Benzylidene-3,5-di-*O*-tosyl-D-xylose dimethyl acetal (0.23 g) plus a 10% solution of sodium iodide in acetone (2.3 ml) were sealed in a glass tube, which was then placed in a boiling water bath (2 hours). After being cooled to room temperature the materials were filtered and the precipitated sodium toluene-*p*-sulphonate was washed with acetone. Yield: 0.03 g, which, allowing for the solubility of sodium toluene-*p*-sulphonate in acetone (8), represents 44% reaction from one tosyl group.

*5-O-Benzoyl-2,4-O-benzylidene-3-O-tosyl-*D*-xylose Dimethyl Acetal*

5-*O*-Benzoyl-2,4-*O*-benzylidene-D-xylose dimethyl acetal (0.30 g) was treated with

tosyl chloride using the procedure described above. After three repetitions of the tosylation process a colorless glass (0.41 g, 97%) was obtained which showed no peak corresponding to free hydroxyl group on infrared examination. It had $[\alpha]_D$ +4° (*c*, 4.62, chloroform). Anal. Calc. for $C_{28}H_{30}O_9S$: S, 5.90. Found: S, 5.88.

The material was heated with sodium iodide in acetone as described for 2,4-*O*-benzylidene-di-*O*-tosyl-D-xylose dimethyl acetal. After 2 hours in the boiling water bath no sodium toluene-*p*-sulphonate was produced.

*2,4-O-Benzylidene-xylitol* (XI)

3,5-Di-*O*-acetyl-2,4-*O*-benzylidene-D-xylose diethyl dithioacetal (IV) (5.15 g) was dissolved in a solution of acetone (4 parts) plus water (1 part) (80 ml). Cadmium carbonate (12 g) was added to the solution, which was stirred vigorously while a solution of mercuric chloride (12.3 g) in acetone (40 ml) was added portionwise at room temperature. After the addition (3 hours) stirring was continued for a further 15 hours at room temperature and the mixture was then boiled under reflux (40 minutes). The product was worked up as described for 2,4-*O*-benzylidene-3,5-di-*O*-acetyl-D-xylose dimethyl acetal, a clear syrup (3.7 g) being obtained.

The above crude 3,5-di-*O*-acetyl-2,4-*O*-benzylidene-aldehydo-D-xylose (3.0 g) was dissolved in 50% aqueous methanol (100 ml) and to the cooled solution was added an excess (2.2 g) of sodium borohydride. The solution was left at room temperature (3 hours) after which excess sodium borohydride was destroyed with glacial acetic acid. The solution was deionized by passage through a column of Amberlite IR-400 (OH) resin and the acidic eluate was shaken with Amberlite IR-120 (H) resin in the cold. The neutral solution was evaporated to dryness and boric acid was removed as methyl borate by codistillation with methanol (4×40 ml). A residue remained (1.05 g, 46% over-all yield) which crystallized on standing and was recrystallized from water. It had a melting point of 142–143° C and showed no optical rotation. Literature (16) gives a melting point of 143–144° C. The material formed a crystalline triacetate which had a melting point of 94–95° C and showed no optical rotation. Literature (16) gives a melting point of 94–95° C.

*2,4:3,5-Di-O-benzylidene-D-xylose Dimethyl Acetal* (XIII)

2,4:3,5-Di-*O*-benzylidene-D-xylose diethyl dithioacetal (5 g) was demercaptalated using mercuric chloride and cadmium carbonate in dry methanol as described for 3,5-di-*O*-acetyl-2,4-*O*-benzylidene-D-xylose diethyl dithioacetal (IV). The product was extracted in the same way yielding a white crystalline residue (3.1 g, 72%) which was recrystallized from chloroform/light petroleum. It had a melting point of 210–211° C and $[\alpha]_D$ −9° (*c*, 2.2, chloroform).

Di-*O*-benzylidene-D-xylose dimethyl acetal was also prepared as described by Breddy and Jones (17) by allowing D-xylose to react with methanol and benzaldehyde in the presence of hydrochloric acid. The white crystalline product was recrystallized from chloroform/light petroleum and had a melting point of 210–211° C undepressed by admixture with 2,4:3,5-di-*O*-benzylidene-D-xylose dimethyl acetal and $[\alpha]_D$ −9° (*c*, 2.25, chloroform). Literature (17) gives a melting point of 211° C and $[\alpha]_D$ −9° (*c*, 1.2, chloroform). The infrared spectrum of this material and that of 2,4:3,5-di-*O*-benzylidene-D-xylose dimethyl acetal were identical.

ACKNOWLEDGMENTS

The authors wish to thank the National Research Council for grants which made this work possible.

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