The apparent first-order rate constants for the increase of absorbance (alkaline readings) of the different uracil derivatives are given in Table II. The acid readings showing the decrease of intact nucleoside were in agreement. Typical first-order plots for alkali readings are given in Figure 6.

The stability of the nucleosides and uracils derived from them under the alkaline conditions at room temperature were checked. The compounds FUA, FUL, OHUD, OHDU, IUD, and OHU were not stable. Of these the first two did not degrade in acid. The remainder were assayed using different procedures.

A quantity of 5-iodouridine, sufficient to give a final concentration of 10^{-4} *M*, was weighed into a 100-ml flask and made up to volume with the appropriate concentration of hydrochloric acid, previously equilibrated at the desired temperature. The solutions were maintained at the temperature of study, samples were taken at suitable intervals, and the ultraviolet spectra of these solutions were determined using the Cary spectrophotometer. Apparent first-order rate constants (Tables II and III) were obtained for the decrease in absorbance at the 288 mμ maximum (IUD). Readings at 260 mμ showing the formation of uridine gave similar rates (Figure 1).

Acid solutions containg 1×10^{-4} *M* 5-hydroxyuridine, prepared as above and degraded at the various temperatures (Tables II and III), showed a first-order loss of the 280-mμ maximum with time.

The appearance of a 261-mμ chromophore due to degraded deoxyribose did not interfere significantly with the measurement of absorbance at the 280-mμ maximum of 5-hydroxy-2′-deoxyuridine but interference was obtained from the degradation product OHU (λ_{max} 285 mμ in acid). Two procedures were adopted for following the degradation of this compound. Solutions containing 10^{-4} *M* OHDU were prepared and degraded as described above for 5-iodouridine and the initial rate of decrease of the absorption maximum was followed. The samples were then made alkaline by the addition of a little more than the equivalent quantity of 10 *N* sodium hydroside solution, allowed to stand at room temperature for 6 hr, and the spectra of these alkaline solutions were also obtained. The alkali treatment was shown in a preliminary experiment to degrade all the interfering isobarbituric acid, OHU, and yet hardly affect the absorbance owing to residual OHDU. The rates of loss of OHDU due to acid hydrolysis were comparable when measured by either of these two procedures.

Thin Layer Chromatography.—The plates were prepared with a 0.4-mm layer of silica gel GF_{254} according to Stahl (E. Merck, Darmstadt). A 10^{-2} *M* solution of the nucleoside in 1.0 *N* hydrochloric acid was reacted at 80° and at intervals 0.02 ml of this solution was spotted at the origin of the plate together with suitable standards. Chromatograms were developed for 12 cm generally using a chloroform–isopropyl alcohol (3:1) solvent system. After development, the spots were viewed under short-wave ultraviolet light, 2537 A. A typical thin layer chromatogram is given in Figure 4.

The silica gel corresponding to the R_f value of each spot was scraped off, extracted with water, and the spectra of these extractions were run at acidic and alkaline pH values. Spectral shifts with pH were evident and confirmed the identities of the nucleoside, the pyrimidine, and the degraded sugar in the cases of acid-degraded BDU (R_f 0.34) to give BU (R_f 0.52); CDU (R_f 0.32) to give CU (R_f 0.55); FDU (R_f 0.21) to give FU (R_f 0.36); DLU (R_f 0.24) to give uracil (R_f 0.30); and DU (R_f 0.17) to give uracil. The above R_f values are for chloroform–isopropyl alcohol 3:1 developing solvent. 5-Iodouridine (R_f 0.75 in *n*-butyl alcohol saturated with 0.2 *N* HCl) gave uridine (R_f 0.50 in the same solvent) and OHUD (R_f 0.60) gave OHU (R_f 0.4 also in the *n*-butyl alcohol–HCl solvent system).

Synthesis.—1-(2′-Deoxy-β-D-lyxofuranosyl)uracil was prepared by a method based on that described for 1-(2-deoxy-β-D-lyxofuranosyl)thymine,[27] mp 167°, R_f 0.35 (chloroform–isopropyl alcohol, 3:1). The method was essentially that subsequently described by Horwitz, *et al.*[28] who quote mp 163°.

2′,3′-Dideoxy-3′-iodouridine was prepared according to the method of Pfitzner and Moffatt:[18] mp 171–172° (from chloroform–ethyl acetate); $\lambda_{max}^{0.1\ N\ HCl}$ 263 mμ (ϵ_{max} 11,250); $\lambda_{max}^{0.1\ N\ NaOH}$ 263 mμ (ϵ_{max} 8700); R_f 0.125 (chloroform–ethyl acetate, 1:2), 0.77 (chloroform–isopropyl alcohol, 3:1). Pfitzner and Moffatt[18] quote mp 162° dec (from acetone); $\lambda_{max}^{0.1\ N\ HCl}$ 262 mμ (ϵ_{max} 11,300); $\lambda_{max}^{0.1\ N\ NaOH}$ 263 mμ (ϵ_{max} 8720).

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Amino Derivatives of Starches. Sulfonation Studies on Methyl 3,6-Anhydro-α-D-glucopyranoside and Related Derivatives[1]

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Selective *p*-toluenesulfonation of methyl 3,6-anhydro-α-D-glucopyranoside (VI) gives the 4-*p*-toluenesulfonate IX. The 2-*p*-toluenesulfonate (VII) and 2-methanesulfonate (V) of VI can be prepared by treatment of methyl α-D-glucopyranoside 2,6-di-*p*-toluenesulfonate (IV) and 2,6-dimethanesulfonate (I), respectively, with base. Reductive desulfonation of VII gives VI, and methanesulfonation of V gives II, proving that V and VII are pyranosides, and the structure of VII was further proved by methylation followed by desulfonation to give the known methyl 3,6-anhydro-4-*O*-methyl-α-D-glucopyranoside (XII). The *p*-tolylsulfonyloxy groups in VII and III were found to be remarkably resistant to displacement by hydrazine. A similar resistance to amination by hydrazine displacement was observed with methyl 3,6:3′,6′-dianhydro-2,2′,4′-tri-*O*-(*p*-tolylsulfonyl)-β-maltoside (XIII). *p*-Toluenesulfonated and methanesulfonated derivatives of 3,6-anhydroamylose were prepared, and were found to be likewise resistant to amination by hydrazine.

A number of reports[2-4] from this laboratory have been concerned with the preparation and characterization of an aminated derivative of amylose,[2] obtained from a slightly derivatized amylose by conversion into a di-*p*-toluenesulfonated derivative, followed by hydrazinolysis and reduction. Possible reaction pathways in the formation of this aminated derivative have been discussed,[3] leading to the production of residues in the polymer with the 3,6-diamino-3,6-dideoxy-D-altrose, 2,6-diamino-2,6-dideoxy-D-mannose, 3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-D-glucose, and 2-amino-3,6-anhydro-2-deoxy-D-mannose structures. Hydrazinolysis studies on the model compound methyl 2,6-di-*O*-(methylsulfonyl)-α-D-glucopyranoside have indicated[4] that 3,6-diamino-3,6-dideoxy-D-altrose resi-

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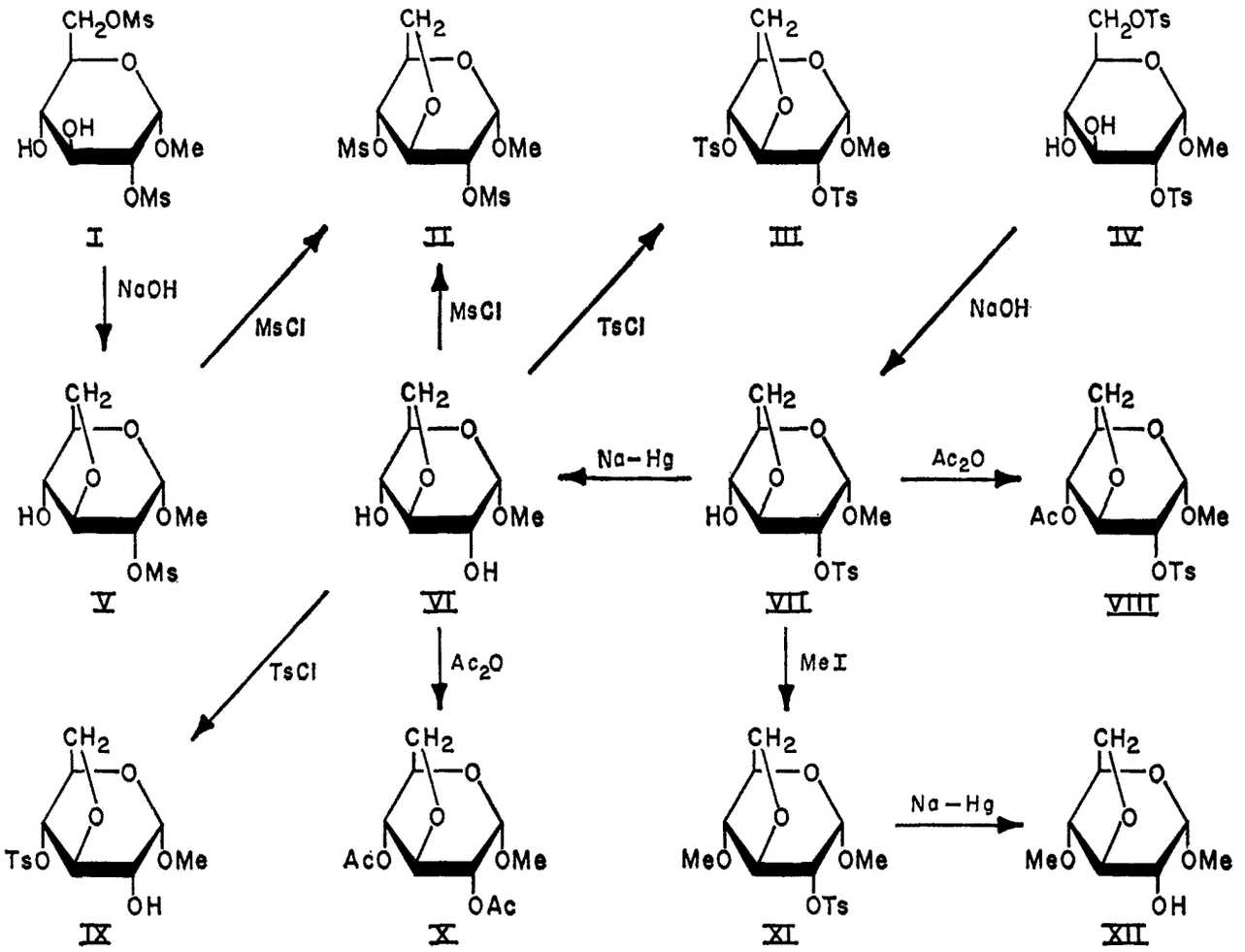
SCHEME I



dues are probably an important constituent of aminated amylose.[2] 2,6-Diamino-2,6-dideoxy-D-mannose has been synthesized[3] as a reference compound, although residues of this sugar are not considered to be significant constituents of aminated amylose prepared by the hydrazinolysis procedure. Since aminated amylose[2] contains 3,6-anhydrohexose residues and contains a small proportion of sulfur which is not removed, even on prolonged treatment with hydrazine, consideration must be given to residues of 3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-D-glucose and 2-amino-3,6-anhydro-2-deoxy-D-mannose, as constituents of aminated amylose. This report is concerned with the synthesis and characterization of methyl 3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII), its 2-*O*-(methylsulfonyl) analog V, and some related derivatives, to furnish model monosaccharide systems for hydrazinolysis studies (see Scheme I). As a disaccharide model system the 2,2′,4′-tri-*p*-toluenesulfonate (XIII) of methyl 3,6:3′,6′-dianhydro-β-maltoside was prepared, and polysaccharide model systems were obtained from 3,6-anhydroamylose by *p*-toluenesulfonation or methanesulfonation. The results indicate that the 2-*p*-tolylsulfonyloxy group is extremely resistant to hydrazinolysis when the 3,6-anhydro bridge is present and suggest therefore that 2-amino-3,6-anhydro-2-deoxy-D-mannose residues are not significant components of aminated amylose. Synthesis of the 2-amino-3,6-anhydro-2-deoxy-D-mannose system is reported in a following paper.[5]

Treatment of methyl 3,6-anhydro-α-D-glucopyranoside[6] (VI) in pyridine with an excess of *p*-toluenesulfonyl chloride gave the 2,4-di-*p*-toluenesulfonate III in excellent yield. The 2,4-di-*p*-bromobenzenesulfonate and the 2,4-dimethanesulfonate II were prepared similarly. Unimolecular *p*-toluenesulfonation of the anhydro glycoside VI gave in low yield a crystalline mono-*p*-toluenesulfonate which was formulated as methyl 3,6-anhydro-4-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (IX) since it differed from a sample of the 2-*p*-toluenesulfonate isomer (VII) which had been synthesized by a definitive route. Substance IX was unstable and turned black on storage, even after careful purification, and it decomposed on melting.

Methyl 2,6-di-*O*-(methylsulfonyl)-α-D-glucopyranoside (I),[7] a well-characterized, crystalline compound, was heated with aqueous ethanolic sodium hydroxide to give crystalline methyl 3,6-anhydro-2-*O*-(methylsulfonyl)-α-D-glucopyranoside (V) in good yield. This product was characterized by methanesulfonation to give the 2,4-dimethanesulfonate II identical with that prepared by methanesulfonation of VI. Methyl 2,6-di-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (IV), known[8] only as a syrup, was heated with aqueous ethanolic sodium hydroxide to give crystalline methyl

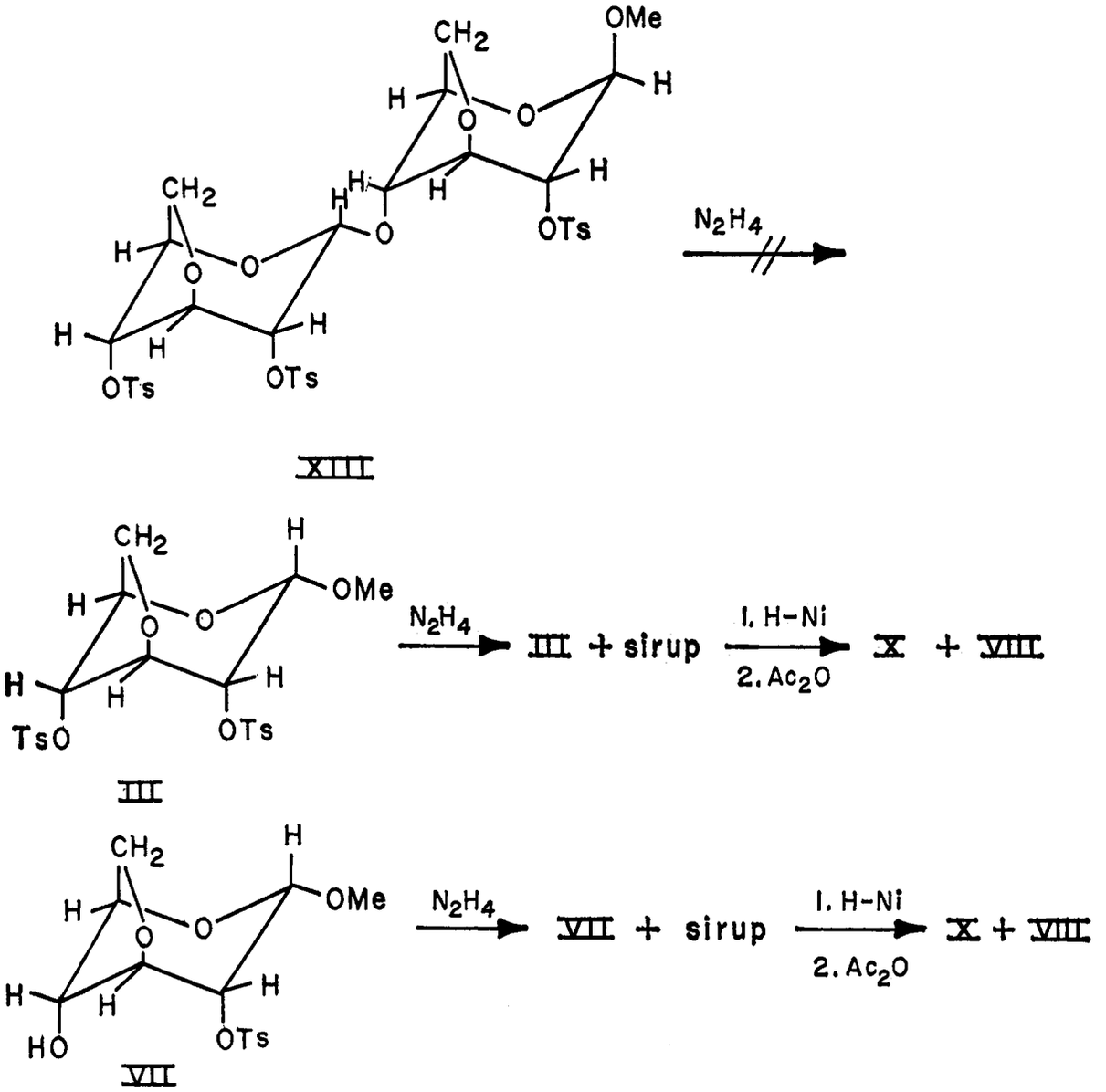
(5) M. L. Wolfrom, P. Chakravarty, and D. Horton, *J. Org. Chem.*, in press.

(6) W. N. Haworth, L. N. Owen, and F. Smith, *J. Chem. Soc.*, 88 (1941).

(7) A. K. Mitra, D. H. Ball, and L. Long, Jr., *J. Org. Chem.*, **27**, 160 (1962).

(8) J. Asselineau, *Bull. Soc. Chim. France*, 937 (1955).

SCHEME II



3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII) in good yield. Its physical constants differed from those given for the isomeric 4-*p*-toluenesulfonate IX; it was stable on storage, and it melted to a clear liquid. Substance VII was further characterized as the 4-acetate VIII. The structure assigned to VII was verified by conversion into methyl 3,6-anhydro-α-D-glucopyranoside (VI) by reductive desulfonation and by methylation to the crystalline 4-methyl ether XI followed by reductive desulfonation to the known[6] methyl 3,6-anhydro-4-*O*-methyl-α-D-glucopyranoside (XII).

Maltose was converted into its methyl β-glycoside[9] by a modified Königs–Knorr type of synthesis, with mercuric cyanide[10] as the condensing agent instead of silver carbonate;[11] the procedure gave a 51% over-all yield in preparations on a 100-g scale. The 6,6′-di-*p*-toluenesulfonate of methyl β-maltoside, prepared by selective *p*-toluenesulfonation, was obtained crystalline in excellent yield, although it proved difficult to purify. The crude 6,6′-di-*p*-toluenesulfonate, on treatment with base, gave the known[11] methyl 3,6:3′,6′-dianhydro-β-maltoside in good yield, and the latter was converted into the amorphous 2,2′,4′-tri-*p*-toluenesulfonate XIII.

3,6-Anhydroamylose[12] was prepared by unimolecular *p*-toluenesulfonation of a slightly derivatized amylose dispersed in pyridine, followed by treatment with methanolic sodium methoxide, according to the general procedure of Whistler and Hirase.[13] This sulfur-free product was *p*-toluenesulfonated in pyridine at room temperature to give a product whose sulfur content indicated that it was 3,6-anhydro-2-*O*-*p*-tolylsulfonylamylose with a degree of substitution (DS) of 0.6 with respect to the *p*-tolylsulfonyloxy group. The 2-*O*-(methylsulfonyl) analog, also of DS 0.6, was similarly prepared. Attempts to prepare a *p*-toluenesulfonated derivative of higher DS by forcing conditions

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(10) B. Helferich and K. F. Wedemeyer, *Ann.* **563**, 139 (1949); R. Kuhn and W. Kirschenlohr, *Ber.*, **86**, 1331 (1953).

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(12) B. J. Bines and W. J. Whelan, *Chem. Ind.* (London), 997 (1960).

(13) R. L. Whistler and S. Hirase, *J. Org. Chem.*, **26**, 4600 (1961).

(hot pyridine) led to the incorporation of chlorine in the molecule; this behavior has been observed[14] in other sugar and polysaccharide systems. A *p*-toluenesulfonated derivative of DS 0.95 was, however, obtained by treatment of 2(?),6-di-*O*-(*p*-tolylsulfonyl)amylose[2] with base.

Efforts to displace the *p*-tolylsulfonyloxy groups by hydrazine in methyl 3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII), in its 2,4-di-*p*-toluenesulfonate analog III, in methyl 3,6:3′,6′-dianhydro-2,2′,4′-tri-*O*-(*p*-tolylsulfonyl)-β-maltoside (XIII), and in 3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)amylose were uniformly unsuccessful, even under the forcing conditions of prolonged reflux in hydrazine, and at best only trace proportions of nitrogen-containing products were observed (see Scheme II). Prolonged hydrazinolysis of VII and III gave, in addition to unchanged starting material, mixtures of syrupy products which, after treatment with hydrogen-saturated Raney nickel followed by acetylation, gave mixtures of methyl 2,4-di-*O*-acetyl-3,6-anhydro-α-D-glucopyranoside (X) and methyl 4-*O*-acetyl-3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VIII), indicating that some cleavage of *p*-tolylsulfonyl groups had taken place, probably during the hydrazinolysis step. Traces only of aminated products were observed by chromatography.

The reluctance of the foregoing derivatives to undergo amination is in contrast with the relatively facile displacement of *p*-tolylsulfonyloxy groups by hydrazine in other carbohydrate systems.[15,16] It is perhaps noteworthy that the systems in the present study are locked in the D-*1C* conformation by the 3,6-anhydro bridge and have an axial oxygen atom antiparallel to the (axial) *p*-tolylsulfonyloxy group at the adjacent carbon atom. It is suggested that this arrangement, in a rigid ring system, is unreactive because of steric hindrance and electronic interactions, between the axial oxygen atom and the incoming nitrogen nucleophile, since the latter must enter from the same side of the ring as the axial oxygen atom. In support of this hypothesis it may be noted that 1,6-anhydro-2,3-*O*-isopropylidene-4-*O*-(*p*-tolylsulfonyl)-β-D-mannopyranose, which likewise has a locked D-*1C* conformation and an oxygen atom (at C-3) antiparallel to the *p*-tolylsulfonyloxy group, is also very resistant to amination by hydrazine or azide ion.[17]

Experimental Section[18]

Methyl 3,6-Anhydro-2,4-di-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (III).—A solution of methyl 3,6-anhydro-α-D-glucopyranoside (VI, 5.00 g), prepared[6] from methyl 6-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside,[19] in dry pyridine (25 ml) was treated at 0° with *p*-toluenesulfonyl chloride (13.55 g, 2.5 molar equiv), and the mixture was kept for 39 hr at room temperature. After cooling to 0°, water (15 ml) was added, and after 15 min the mixture was poured onto ice (500 g). After 1 hr the solid product was filtered, washed with water, and dried: yield 13.75 g (99%). Recrystallization from ethanol–acetone gave pure material, mp 162–163°, $[\alpha]^{21}D$ +28° (*c* 1, chloroform).

Anal. Calcd for $C_{21}H_{24}O_9S_2$: C, 52.05; H, 4.99; S, 13.23. Found: C, 52.32; H, 5.22; S, 13.10.

Methyl 3,6-Anhydro-2,4-di-*O*-(*p*-bromophenylsulfonyl)-α-D-glucopyranoside.—This compound was prepared by a route essentially identical with that used for the di(*p*-tolylsulfonyl) analog III: yield 90%, mp 144–145° (from ethanol–acetone), $[\alpha]^{17}D$ +31° (*c* 1, chloroform).

Anal. Calcd for $C_{19}H_{18}Br_2O_9S_2$: C, 37.21; H, 2.96; Br, 26.06; S, 10.46. Found: C, 37.70; H, 3.09; Br, 26.10; S, 10.60.

Methyl 3,6-Anhydro-2,4-di-*O*-(methylsulfonyl)-α-D-glucopyranoside (II). A. From Methyl 3,6-Anhydro-α-D-glucopyranoside (VI).—This compound was prepared by a route essentially identical with that used for the di(*p*-tolylsulfonyl) analog III: yield 70%, mp 160–161° (from acetone–ethanol), $[\alpha]^{22}D$ +66° (*c* 1, chloroform).

Anal. Calcd for $C_9H_{16}O_9S_2$: C, 32.54; H, 4.85; S, 19.29. Found: C, 32.97; H, 4.78; S, 19.40.

B. From Methyl 3,6-Anhydro-2-*O*-(methylsulfonyl)-3,α-glucopyranoside (V).—A solution of 100 mg of methyl D-6-anhydro-2-*O*-(methylsulfonyl)-α-D-glucopyranoside (V, see below) in dry pyridine (0.1 ml) was treated at 0° with methanesulfonyl chloride (0.1 ml), then kept for 30 hr at room temperature. The solution was poured into water at 0°, whereupon the product crystallized: yield 80 mg (61%). Recrystallization from acetone–ethanol gave pure material, mp 161–162°, identical by mixture melting point and infrared spectrum with material prepared by method A.

Methyl 3,6-Anhydro-4-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (IX).—A solution of methyl 3,6-anhydro-α-D-glucopyranoside (VI, 1.0 g) in pyridine (5 ml) and chloroform (25 ml) was treated at 0° with *p*-toluenesulfonyl chloride (1.08 g, 1.0 molar equiv) added portionwise during 40 min. The solution was kept for 2 days at room temperature, then water (5 ml) was added, and after 30 min the mixture was stirred with ice (100 g). The chloroform layer was separated, washed with water (40 ml), dried (magnesium sulfate), and evaporated to a syrup, which crystallized from ethanol: yield 0.2 g (11%); mp 116° (with blackening); $[\alpha]^{30}D$ +32° (*c* 1, chloroform); λ_{max}^{KBr} 3.00 (OH), 6.23 μ (aryl C=C).

Anal. Calcd for $C_{14}H_{18}O_7S$: C, 50.90; H, 5.49; S, 9.71. Found: C, 50.66; H, 5.28; S, 9.66.

This compound, even when carefully purified, darkened on standing at room temperature, and became black after a few weeks.

Methyl 3,6-Anhydro-2-*O*-(methylsulfonyl)-α-D-glucopyranoside (V).—Methyl 2,6-di-*O*-(methylsulfonyl)-α-D-glucopyranoside[7] (I, 1.0 g) in ethanol (10 ml) was treated with 1 *N* aqueous sodium hydroxide (5 ml) for 1 day at room temperature and then for 1 hr at 60°. The solution was neutralized with solid carbon dioxide and evaporated, and the residue was extracted with boiling acetone. Evaporation of the extract and crystallization from ethanol gave a chromatographically homogeneous product, yield 0.80 g (55%), mp 118–119°. Recrystallization from ethanol gave material, mp 119–120°, $[\alpha]^{20}D$ +87° (*c* 2.2, chloroform).

Anal. Calcd for $C_8H_{14}O_7S$: C, 37.79; H, 5.55; S, 12.61. Found: C, 38.44; H, 5.75; S, 12.82.

Methyl 3,6-Anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII).—Methyl α-D-glucopyranoside (20 g) was converted[8] into syrupy methyl 2,6-di-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (IV), and a solution of this product in ethanol (200 ml) was treated with 1 *N* aqueous sodium hydroxide (100 ml) for 1 day at 60°. The solution was neutralized with solid carbon dioxide and evaporated, and the residue was extracted with hot acetone. Evaporation of the extract gave a crystalline product, yield 11 g (55%), which was recrystallized twice from ethanol: mp 122°

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(15) K. Freudenberg and F. Brauns, *Ber.*, **55**, 3233 (1922).

(16) M. L. Wolfrom, F. Shafizadeh, R. K. Armstrong, and T. M. Shen Han, *J. Am. Chem. Soc.*, **81**, 3716 (1959); M. L. Wolfrom, F. Shafizadeh, and R. K. Armstrong, *ibid.*, **80**, 4885 (1958); D. Horton, M. L. Wolfrom, and A. Thompson, *J. Org. Chem.*, **26**, 5069 (1961); M. L. Wolfrom, J. Bernsmann, and D. Horton, *ibid.*, **27**, 4505 (1962).

(17) A. K. Chatterjee, D. Horton, and J. S. Jewell, to be published.

(18) Melting points were determined with a Hershberg-type apparatus [A. Thompson and M. L. Wolfrom, *Methods Carbohydrate Chem.*, **1**, 517 (1962)]. Specific rotations were determined in a 2-dm. polarimeter tube. Elemental analyses were made by W. N. Rond. X-Ray powder diffraction data give interplanar spacings, in angstroms, for Cu Kα radiation. Relative intensities were estimated visually: s, strong; m, moderate; w, weak; v, very. The strongest lines are numbered (1, strongest), multiple numbers indicate approximately equal intensities. Thin layer chromatography was performed with Desaga equipment, and silica gel G (E. Merck, Darmsadt, Germany) activated at 110° as the adsorbent, with indication by sulfuric acid. For column chromatography, silica gel Davison, Grade 950, 60–200 mesh (Davison Division of the W. R. Grace Co., Baltimore, Md.), was used.

(19) F. Cramer, H. Otterbach, and H. Springmann, *Ber.*, **92**, 384 (1959).

(without decomposition); $[\alpha]^{26}D$ +63° (*c* 1, chloroform); X-ray powder diffraction data, 11.63 w, 8.59 s (1), 6.97 s (2), 5.68 vw, 5.40 m, 5.10 m, 4.58 s (3), 3.95 m, 3.75 s, 3.62 vw, 3.14 w, 2.98 w.

Anal. Calcd for $C_{14}H_{18}O_7S$: C, 50.90; H, 5.49; S, 9.71. Found: C, 51.11; H, 5.66; S, 10.00.

This product was stable on storage.

Thin layer chromatography of the mother liquor, with ethyl acetate as developer, revealed the presence of eight components.

Desulfonylation of Methyl 3,6-Anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII).—A solution of VII (1.0 g) in ethanol (24 ml) was treated[15] portionwise with 2% sodium amalgam (20 g), stirred for 24 hr at room temperature, neutralized with solid carbon dioxide, and evaporated. The residue was extracted with boiling acetone, and then with ethyl acetate. The extracts were evaporated to give a crystalline residue, yield 0.46 g (86%), which after recrystallization from ethyl acetate had mp 110–112°. The product was indistinguishable by mixture melting point and X-ray powder diffraction pattern from an authentic sample of methyl 3,6-anhydro-α-D-glucopyranoside (VI).

Methyl 4-*O*-Acetyl-3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VIII).—Methyl 3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII, 1.0 g) was treated with pyridine (5 ml) and acetic anhydride (1 ml) for 24 hr at room temperature, and the mixture was poured onto ice to give a crystalline solid, yield 1.12 g (99%), mp 162–163°. Recrystallization from ethanol–petroleum ether (bp 36–60°) gave pure material: mp 162°; $[\alpha]^{21}D$ +70° (*c* 1, chloroform); X-ray powder diffraction data, 11.95 s (3), 8.59 w, 8.19 s (1), 7.20 m, 6.46 m, 5.61 s, 5.54 m, 5.31 m, 4.72 m, 4.48 m, 4.15 s (2).

Anal. Calcd for $C_{16}H_{20}O_8S$: C, 51.64; H, 5.41; S, 8.61. Found: C, 51.79; H, 5.42; S, 8.68.

Methyl 2,4-Di-*O*-acetyl-3,6-anhydro-α-D-glucopyranoside (X).—Methyl 3,6-anhydro-α-D-glucopyranoside (VI, 0.80 g) in pyridine (8 ml) was treated with acetic anhydride (2 ml) for 24 hr at room temperature, the solution was poured into ice and water, and the product was extracted with chloroform. The dried (magnesium sulfate) extract was evaporated, the residue was codistilled with toluene to remove pyridine and then crystallized from ethanol–petroleum ether (bp 30–60°): yield 1.06 g (90%). Recrystallization from the same solvents gave a product: mp 131–132°; $[\alpha]^{22}D$ +106° (*c* 1, chloroform); X-ray powder diffraction data, 8.51 s (1), 6.92 m, 5.61 m, 4.90 s (2), 4.72 vw, 4.53 s (3), 4.25 vw, 4.10 m, 3.39 m, 3.71 s, 3.45 m, 2.95 m.

Anal. Calcd for $C_{11}H_{16}O_7$: C, 50.76; H, 6.19. Found: C, 50.93; H, 6.21.

Methyl 3,6-Anhydro-4-*O*-methyl-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (XI).—Methyl 3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII, 2.0 g) was dissolved in dry *N*,*N*-dimethylformamide (20 ml) at 0°, methyl iodide (2 ml) and freshly prepared silver oxide (3 g) were added, and the mixture was shaken in the dark for 28 hr. The solution was filtered, the filter was washed repeatedly with chloroform, and the combined filtrate and washings were filtered again to remove a white solid. The filtrate was evaporated and the residue was crystallized from ethanol–petroleum ether (bp 30–60°): yield 1.85 g (90%), mp 86–87°. Recrystallization from the same solvents gave pure material, mp 86–87°, $[\alpha]^{22}D$ +43° (*c* 1, chloroform).

Methyl 3,6-Anhydro-4-*O*-methyl-α-D-glucopyranoside (XII).—Methyl 3,6-anhydro-4-*O*-methyl-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (XI, 0.50 g) was dissolved in ethanol (12 ml), water (3 ml) was added, and the mixture was stirred with 2% sodium amalgam (10 g), added in portions. After 24 hr at room temperature the solution was neutralized with solid carbon dioxide and evaporated, and the residue was extracted with chloroform. Evaporation of the extract gave a crystalline product, which was recrystallized from acetone–ether: yield 0.205 g (74%), mp 149–152°. Further recrystallization from acetone–ether gave pure product, mp 152°, $[\alpha]^{23}D$ +24° (*c* 1.1, water).

Haworth and co-workers[8] gave mp 152°, $[\alpha]^{17}D$ +24° (*c* 1.1, water), for methyl 3,6-anhydro-4-*O*-methyl-α-D-glucopyranoside (XII).

Hydrazinolysis of Methyl 3,6-Anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VII).—Methyl 3,6-anhydro-2-(*O*-*p*-tolylsulfonyl)-α-D-glucopyranoside (VII, 2.5 g) was refluxed with hydrazine[20] (25 ml) for 3 days under nitrogen. The mixture was evaporated and water was added to the syrup. A white crystalline precipitate formed and was filtered, yield 0.50 g, which was shown to be unchanged starting material (VII). The filtrate was stirred for 2 days with an excess of Raney nickel,[21] and then filtered and evaporated. Paper chromatography of the syrup, with 5:5:3:1 pyridine–ethyl acetate–water–acetic acid[22] as developer and indication with ninhydrin, revealed two ninhydrin-positive components, R_f 0.19 and 0.47, but in very small proportion. The dried syrup (1.8 g) was treated with pyridine (5 ml) and acetic anhydride (5 ml) for 24 hr at room temperature. Ice and water were added; after 2 hr the solution was exhaustively extracted with chloroform and the extract was dried with magnesium sulfate and evaporated. Thin layer chromatography of the resultant syrup, with ethyl acetate as developer, revealed a major component, R_f 0.65, a minor component, R_f 0.77, and two trace components, R_f 0.12 and 0.22. The major component, R_f 0.65, was separated on silica gel plates having a 1.5-mm coating. The zone was marked by iodine vapor and was collected, eluted with chloroform, and crystallized from ethanol–petroleum ether (bp 30–60°): yield 0.70 g, mp 133–134°, $[\alpha]^{20}D$ +106° (*c* 1, chloroform). This substance was identical by mixture melting point, specific rotation, X-ray powder diffraction pattern, and infrared spectrum with an authentic sample of methyl 2,4-di-*O*-acetyl-3,6-anhydro-α-D-glucopyranoside (X).

The product, R_f 0.77, was chromatographically indistinguishable from authentic methyl 4-*O*-acetyl-3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VIII). The identities of the other two components were not determined.

Hydrazinolysis of Methyl 3,6-Anhydro-2,4-di-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (III).—Methyl 3,6-anhydro-2,4-di-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (III, 5.0 g) was refluxed with hydrazine[20] (50 ml) for 5 days under nitrogen. The mixture was evaporated and water was added to the residue. The white, crystalline solid which precipitated was filtered, yield 2.5 g; it was shown to be unchanged starting material. The filtrate was stirred for 2 days with an excess of Raney nickel[21] and then filtered and evaporated. Paper chromatography of the syrup, with 5:5:3:1 pyridine–ethyl acetate–water–acetic acid[22] as developer, revealed two minor ninhydrin-positive components, R_f 0.20 and 0.48. The dried residue was acetylated as in the preceding experiment, and thin layer chromatography of the product, with ethyl acetate as developer, revealed a major component, R_f 0.66, a minor component, R_f 0.8, and two trace components, R_f 0.11 and 0.20. The first two components were separated by preparative thin layer chromatography as in the preceding experiment. Crystallization of the component, R_f 0.66, from ethanol–petroleum ether (bp 30–60°) gave methyl 2,4-di-*O*-acetyl-3,6-anhydro-α-D-glucopyranoside (X), yield 0.20 g, mp 133–134°, $[\alpha]^{20}D$ +106° (*c* 1, chloroform). Crystallization of the component, R_f 0.80, from ethanol–petroleum ether (bp 30–60°) gave methyl 4-*O*-acetyl-3,6-anhydro-2-*O*-(*p*-tolylsulfonyl)-α-D-glucopyranoside (VIII), yield 0.18 g, mp 162°, $[\alpha]^{20}D$ +70° (*c* 1, chloroform). Both crystalline compounds were identical with authentic samples by mixture melting point, specific rotation, X-ray powder diffraction pattern, and infrared spectrum. The identities of the trace components were not determined.

Preparation of Methyl Hepta-*O*-acetyl-β-maltoside.—Maltose (100 g) was suspended in acetic acid (500 ml) and acetic anhydride (250 ml). Perchloric acid (2.4 ml) was added to the cooled, stirred mixture. After 1 hr the clear solution was cooled to 0°, and a 40% solution of hydrogen bromide in acetic acid (400 ml) was added. The mixture was allowed to warm to room temperature and was then poured into 2 l. of ice and water and stirred for 15 min. The precipitate was then filtered, washed well with water, and dissolved in chloroform. The chloroform solution was washed once with water, once with aqueous sodium bicarbonate, and once more with water, dried over magnesium sulfate, filtered, and concentrated to a syrup, yield 174 g. This hepta-*O*-acetyl-α-maltosyl bromide (amorphous) was shaken in benzene (450 ml) and methanol (90 ml) in the presence of mercuric cyanide (90 g) for 3 hr. 1,2-Dichloroethane (1.5 l.) was added and the resulting solution was washed with water until the washings gave no precipitate with silver nitrate solution. The solution was then dried with magnesium sulfate, filtered, and concentrated to a syrup which crystallized upon the addition of

(20) Technical hydrazine, anhydrous 95+%. Olin Mathieson Chemical Corp., New York 22, N. Y.

(21) Raney nickel catalyst No. 28, The Raney Catalyst Division of the W. R. Grace Co., Inc., Chattanooga, Tenn.

(22) F. G. Fischer and H. J. Nebel, *Z. Physiol. Chem.*, **302**, 10 (1955).

ethanol. The product was recrystallized from ethanol: yield 110 g (58% over-all yield from maltose), mp 122–124°, $[\alpha]^{23}D$ +53° (*c* 3.2, chloroform); lit.[11] mp 123–124°, $[\alpha]^{20}D$ +53.5° (*c* 3.1, chloroform).

Preparation of Methyl β-Maltoside Monohydrate.—Methyl hepta-*O*-acetyl-β-maltoside (110 g) was refluxed for 5 hr in dry methanol (750 ml) containing *n*-butylamine (15 ml). The solution was concentrated and allowed to crystallize: yield 55 g, mp 101–105°. The product was recrystallized from ethanol: yield 53 g (84% or 51% over-all yield from maltose), mp 108–109°, $[\alpha]^{23}D$ +78° (*c* 2, water); lit.[11] mp 110–111°, $[\alpha]^{19}D$ +81° (*c* 2, water).

Methyl 6,6′-di-*O*-*p*-Tolylsulfonyl-β-maltoside.—An azeotropically dried solution of methyl β-maltoside (2.0 g) in dry pyridine (10 ml) was treated at 0° with *p*-toluenesulfonyl chloride (2.1 g, 2 molar equiv). After 24 hr at 0° and 1 hr at 25°, water (2 ml) was added to the solution, and after 30 min the solution was poured onto ice (150 g). The product was extracted with chloroform, and the dried (magnesium sulfate) extract was evaporated to a syrup which crystallized after nucleation: yield 2.8 g (86%). Nuclei were obtained by chromatography of the syrup (1 g) on a column of silica gel[18] (100 g) with a series of solvents of increasing polarity in the order benzene, dichloromethane, ether, ethyl acetate, acetone; crystalline material (0.5 g) was obtained from fractions eluted with acetone. The crude product was recrystallized six times from 95% ethanol: yield 1.6 g (50%), mp 124–126°, $[\alpha]^{23}D$ +45° (*c* 3.3, chloroform).

Anal. Calcd for $C_{25}H_{32}O_{15}S_2$: C, 47.16; H, 5.07; S, 10.07. Found: C, 47.59; H, 5.68; S, 10.20.

Preparation of Methyl 3,6:3′,6′-Dianhydro-β-maltoside.—Methyl 6,6′-di-*O*-*p*-tolylsulfonyl-β-maltoside (crude, 50 g) was refluxed for 3 hr in ethanol (1 l.) containing 2.5 *N* aqueous sodium hydroxide (200 ml). Solid carbon dioxide was added to neutralize the base, the precipitated sodium carbonate was filtered, and the filtrate was concentrated to a thin syrup which crystallized on nucleation. Recrystallization from ethanol gave pure material: yield 18 g (71%), mp 100.5–102.0°, $[\alpha]^{20}D$ −68.5° (*c* 2.5, water); lit.[11] mp 95–101°, $[\alpha]^{17}D$ −66° (*c* 2.4, water).

Methyl 3,6:3′,6′-Dianhydro-2,2′,4′-tri-*O*-*p*-tolylsulfonyl-β-maltoside (XIII).—Methyl 3,6:3′,6′-dianhydro-β-maltoside (1 g) in pyridine (5 ml) was treated with *p*-toluenesulfonyl chloride (3 g, ~5 molar equiv) for 12 hr at room temperature. A few drops of water were added, and after 30 min the solution was poured into ice and water (50 ml). The precipitate was filtered, washed with water, and dried to give an amorphous solid, yield 2.2 g (90%). The product was chromatographically homogeneous on silica gel by thin layer and column techniques.

Anal. Calcd for $C_{34}H_{38}O_{15}S_3$: C, 52.16; H, 4.89; S, 12.28. Found: C, 52.26; H, 5.39; S, 11.96.

Attempted Hydrazinolysis of Methyl 3,6:3′,6′-Dianhydro-2,2′,4′-tri-*O*-(*p*-tolylsulfonyl)-β-maltoside[23] (XIII).—Methyl 3,6:-3′,6′-dianhydro-2,2′,4′-tri-*O*-(*p*-tolylsulfonyl)-β-maltoside (XIII, 10 g) was refluxed with hydrazine[20] (100 ml) for 4 days under nitrogen. The solution was evaporated and the residue was poured into water (500 ml). A white solid precipitated, yield 5.2 g, which was shown to be unchanged starting material (XIII). The filtrate was stirred for 2 days with an excess of Raney nickel. Evaporation of the solution gave, on trituration with methanol, further crops of solid, apparently starting material XIII. The residual syrup (1.2 g) was shown by paper chromatography to contain only a small proportion of ninhydrin-positive component.

Pyridine-Swollen Amylose.—Samples (1 g) of slightly derivatized amylose[24] were treated at room temperature with aqueous pyridine (25 ml). The material was completely dissolved by 60% aqueous pyridine in 5–10 min, 90% aqueous pyridine required 30 min, 95% aqueous pyridine required 2 hr, and the material was insoluble in anhydrous pyridine. For subsequent conversions the slightly derivatized amylose[24] (18 g) was stirred for 2 hr with a 95:5 pyridine–water mixture (450 ml), the clear viscous solution was evaporated, and water was removed by repeated codistillation with anhydrous pyridine until the distillate no longer became turbid when benzoyl chloride was added. The solvent was then removed completely to leave a residue of pyridine-swollen amylose.

Preparation of 6-*O*-*p*-Tolylsulfonylamylose.[13]—Pyridine-swollen amylose (9 g) was stirred with anhydrous pyridine (180 ml) for 30 min at 35°, *p*-toluenesulfonyl chloride (21 g) was added, and the mixture was stirred for 1 hr at 35°. The resulting thick, brown solution was poured slowly, with vigorous stirring, into an excess of methanol containing 20% of water, and the gummy solid which separated was dispersed in water with a blender. The product was filtered, washed well with water, and dried to give a light brown powder, yield 15.5 g.

Anal. Calcd for $[C_6H_{9.1}O_{4.1}(OSO_2C_7H_7)_{0.9}]_n$: C, 49.10; H, 5.12; S, 9.58. Found: C, 49.22; H, 5.15; S, 9.66.

Preparation of 3,6-Anhydroamylose.[13]—6-*O*-*p*-Tolylsulfonylamylose (19 g) was suspended in absolute methanol (300 ml) in which sodium (3.3 g) had been dissolved, and the mixture was stirred under nitrogen for 3 days at 35°. The solid was filtered, washed with methanol, aqueous methanol, and dispersed in methanol with a Blender. The product was filtered and washed thoroughly with methanol until the washings were neutral to phenolphthalein, and then dried: yield 6.6 g of a slightly brown powder. The product contained no sulfur.

3,6-Anhydro-2-*O*-(methylsulfonyl)amylose.—To a stirred suspension of 3,6-anhydroamylose (3.6 g) in dry pyridine (40 ml) was added methanesulfonyl chloride (5.7 g) dropwise during 3 hr, and the mixture was stirred under nitrogen for 7 days at room temperature. The dark brown mass was dialyzed against running distilled water for 4 days. The solid was filtered, washed successively with water, methanol, and ether, and then dried: yield 4.25 g.

Anal. Calcd for $[C_6H_{7.4}O_{3.4}(OSO_2CH_3)_{0.6}]_n$: C, 41.50; H, 4.80; S, 10.06. Found: C, 40.69; H, 4.95; S, 9.94.

3,6-Anhydro-2-*O*-(*p*-tolylsulfonyl)amylose. A. From 3,6-Anhydroamylose in Hot Pyridine.—3,6-Anhydroamylose (3 g), *p*-toluenesulfonyl chloride (11.8 g, 3 molar equiv), and dry pyridine (90 ml) were refluxed for 10 hr, and the cooled, dark solution was slowly poured into 80% aqueous methanol (540 ml) with constant stirring. The fine, light brown precipitate was filtered, washed with 80% aqueous methanol until free from chloride ion, and dried: yield 4.5 g.

Anal. Calcd for $[C_6H_7O_3(OSO_2C_7H_7)_{0.75}(Cl)_{0.25}]_n$: C, 51.11; H, 4.63; Cl, 3.41; S, 9.22. Found: C, 51.53; H, 5.26; Cl, 3.59; S, 9.16.

B. From 3,6-Anhydroamylose in Cold Pyridine.—The mixture of reactants used in the preceding experiment was shaken for 12 days at room temperature. The resulting solution was dialyzed against running distilled water for 4 days, and the solid was washed successively with water, methanol, and ether, and then dried: yield 5.0 g of a light brown powder.

Anal. Calcd for $[C_6H_{7.4}O_{3.4}(OSO_2C_7H_7)_{0.6}]_n$: S, 8.12. Found: S, 8.42.

C. From 2(?),6-Di-*O*-(*p*-tolylsulfonyl)amylose.—Sodium (6.1 g) was dissolved in methanol (530 ml), 2(?),6-di-*O*-(*p*-tolylsulfonyl)amylose[2] (42 g) was added, and the mixture was stirred under nitrogen for 7 days. The brown product was filtered, washed thoroughly with methanol, and dried: yield 23 g.

Anal. Calcd for $[C_6H_{7.1}O_{3.1}(OSO_2C_7H_7)_{0.9}]_n$: S, 10.19. Found: S, 9.88.

Hydrazinolysis of 3,6-Anhydro-2-*O*-(*p*-tolylsulfonyl)amylose.—The *p*-tolylsulfonyl derivative (prepared by method B or C) was refluxed in hydrazine[20] for periods up to 14 days, the hydrazine was removed by concentration under reduced pressure, and the residue was dialyzed for 2 days against running distilled water. The starting material was recovered, apparently unchanged, and analysis indicated that it contained no nitrogen. A similar lack of reactivity was observed when the material was refluxed in *N*,*N*-dimethylformamide with sodium azide or sodium amide.

(23) This experiment was performed by Dr. M. I. Taha.

(24) "Superlose," Stein-Hall HAA-11-HV, High Viscosity, Control No. 12215, Stein-Hall and Co., Inc., New York, N. Y.