The sirupy 5-deoxy-D-*xylo*-hexose (III) formed a crystalline phenylosazone (V) and phenylosatriazole (VI) identical with those produced from 5-deoxy-D-*threo*-hexose (IV, "5-deoxy-L-sorbose")[11] of established structure. Thus the sirupy free sugar is established as 5-deoxy-D-*xylo*-hexose.

Experimental

5-Deoxy-1,2-*O*-isopropylidene-α-D-*xylo*-hexofuranose (II).—A solution of 3 g. of 5,6-dideoxy-1,2-*O*-isopropylidene-α-D-*xylo*-hexofuran-5-enose (I)[4] and 0.93 g. of lithium borohydride in 50 ml. of tetrahydrofuran was treated dropwise, under a stream of nitrogen gas, with 2.5 g. of concentrated sulfuric acid and the resulting solution stirred for 2 hr. at room temperature. The excess reagent was destroyed by adding methanol and the solution was evaporated under reduced pressure. The residue was dissolved in 50 ml. of chloroform, washed twice with water, dried over sodium sulfate, and the solvent removed under reduced pressure to yield a sirup. The sirup was dissolved in 10 ml. of absolute ethanol, 2 g. of pulverized sodium hydroxide was added and the alkaline ethanol solution was treated dropwise, with stirring, with 1.5 ml. of 30% hydrogen peroxide. After stirring for 1 hr. at room temperature, the mixture was filtered. The filtrate was neutralized with Amberlite[12] IR-120 (H^+) resin and evaporated to a sirup under reduced pressure following removal of the resin by filtration. The sirup was extracted with ether and the solvent removed under reduced pressure to yield a sirup which crystallized from ether–petroleum ether (b.p. 30–60°) yield 0.74 g., m.p. 88–92°. A second recrystallization from the same solvent yielded pure product, m.p. 94–96°, $[\alpha]^{24}D$ −10.6° (*c* 2.0, chloroform). The aforementioned sample and a sample furnished by Professor Overend evidenced identical X-ray powder diffraction patterns[13]: 9.41 vw, 8.30 s (2), 5.68 s (3), 4.72 vs (1), 4.21 vw, 3.92 w, 3.72 w, 3.53 w, 3.29 vw. A mixture melting point of the sample with an authentic sample of 6-deoxy-1,2-*O*-isopropylidene-β-L-idofuranose[6] (m.p. 89–91°) was 70–80°.

5-Deoxy-D-*xylo*-hexose (III).—A solution of 0.5 g. of II in 10 ml. of water was heated on a steam bath for 3 hr. with 2.0 g. of Amberlite[12] IR-120 (H^+) resin. The mixture was then filtered and the solvent removed from the filtrate under reduced pressure to yield a sirup. This sirup was dried by repeated evaporation with methanol under reduced pressure, $[\alpha]^{24}D$ +38° (*c* 2.1, water).[14]

This sirupy free sugar was chromatographed on Whatman No. 1 filter paper using an upper layer of 1-butanol–ethanol–water (4:1:5 by vol.) as developer. A single spot, $R_{glucose}$ 2.59, was obtained by spraying with aniline hydrogen phthalate reagent.[15]

5-Deoxy-D-*threo*-hexose Phenylosazone (V).—A solution of 0.5 g. of II in 5 ml. of 0.1 *N* hydrochloric acid was heated on a steam bath for 1.5 hr. to hydrolyze the isopropylidene group. The solution was neutralized with 5 ml. of 0.1 *N* sodium hydroxide solution. To this solution was added 5 ml. of a solution of phenylhydrazine prepared from 1 g. of phenylhydrazine hydrochloride and 1.5 g. of sodium acetate. The solution was heated on a steam bath for 0.5 hr. and cooled in a refrigerator overnight. The crude osazone was removed by filtration, triturated with chloroform, and dried; yield, 0.12 g. Recrystallization from 60% ethanol gave a pure product, m.p. 151° (lit.[11] m.p. 153°); X-ray powder diffraction pattern[13]: 10.53 m, 8.42 w, 6.92 vw, 5.55 m (3), 4.80 m (2), 4.56 s (1), 4.27 m, 3.87 vw, 3.38 vw, 3.33 m, 3.23 vw, 3.09 w.

5-Deoxy-D-*threo*-hexose Phenylosotriazole (VI).—A solution of crude 5-deoxy-D-*threo*-hexose phenylosazone (0.1 g.) in 6 ml. of isopropyl alcohol was mixed with a solution of 0.3 g. of cupric sulfate pentahydrate in 9 ml. of water and refluxed for 1 hr. on a steam bath. The reaction mixture was treated with a small amount of carbon and filtered while warm. The filtrate was concentrated, under reduced pressure, to a volume of about 3 ml. and kept in a refrigerator overnight. The brown precipitate obtained on filtration was dissolved in a minimal amount of hot water, decolorized with carbon, and again placed in the cold. Colorless crystals were obtained upon filtration; yield, a few mg.; m.p. 140–140.5°; X-ray powder diffraction pattern[13]: 13.00 w, 7.53 m, 6.49 s (3), 4.93 s (1), 4.53 w, 4.33 w, 4.05 w, 3.88 vw, 3.60 vw, 3.44 s (2), 3.23 m, 2.96 vw, 2.88 vw, 2.69 vw, 2.53 vw, 2.44 w. These physical constants are identical with those of the phenylosotriazole produced from a sample of 5-deoxy-D-*threo*-hexose (5-deoxy-L-sorbose) kindly provided by Dr. P. Regna.

Acknowledgment.—It is a pleasure to acknowledge the assistance of Dr. G. Fraenkel and Mr. Byron Bossenbroek of this department in obtaining and interpreting the n.m.r. spectrum. The assistance and counsel of the late Dr. Alva Thompson is also acknowledged.

(11) P. P. Regna, *J. Am. Chem. Soc.*, **69**, 246 (1947).

(12) A product of the Rohm and Haas Co., Philadelphia, Pa.

(13) Interplanar spacing, Å., CuKα radiation. Relative intensities, estimated visually; s, strong; m, medium; w, weak; v, very. Strongest lines numbered, 1 strongest.

(14) This value supersedes that previously reported[1a] although since the product was a sirup no high accuracy is claimed.

(15) S. M. Partridge, *Nature*, **164**, 443 (1949).

Amino Derivatives of Starches. Amination of Amylose[1]

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Received June 13, 1963

It was of interest to perform chemical modifications on the amylose molecule with a view to the replacement of secondary hydroxyl by amino groups while maintaining polymeric structure. Such a cationic polymer might be expected to possess physical and chemical properties which could offer possibilities for increased utilization of starch. A polymer modified by amination at C-2 might possess the high stability toward hydrolysis exhibited by chitosan; the acetamido analog would be analogous to chitin, a polysaccharide whose high degree of intermolecular hydrogen bonding[2] affords great physical stability. The present work describes the conversion of a sulfonylated amylose by hydrazinolysis, followed by reduction, to give a product which in all probability has had a considerable proportion of its secondary hydroxyls replaced by amino groups.

Amylose was treated portionwise with a total of 2.2 molar equivalents of *p*-toluenesulfonyl chloride in pyridine to yield a *p*-toluenesulfonate ester derivative with a degree of substitution of 1.7. Esterification of amylose with *p*-toluenesulfonyl chloride has been shown[3] to take place selectively and readily at the C-6 position of the D-glucose units. Further reaction would presumably occur selectively, though probably not exclusively, at the C-2 hydroxyl group.[4]

The 2(3),6-di-*O*-*p*-tolylsulfonylamylose was then refluxed with hydrazine and the resulting hydrazino derivative was reduced by Raney nickel to yield an aminated derivative with a degree of substitution of 1.4. The amino groups of the aminated amylose were selectively acetylated with aqueous acetic anhydride. The

(1) Supported by Contract No. 12-14-100-5760(71) (Research Foundation Project 1301) from the U. S. Department of Agriculture, Northern Utilization Research Bureau, Peoria 5, Ill. The opinions expressed in this article are those of the authors, and not necessarily those of the sponsoring agency.

(2) S. E. Darmon and K. M. Rudall, *Discussions Faraday Soc.*, **9**, 251 (1950).

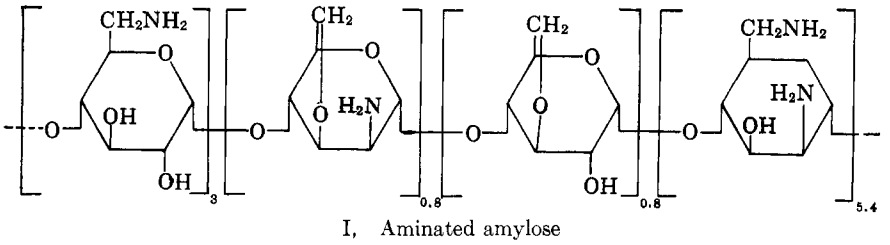
(3) R. L. Whistler and S. Hirase, *J. Org. Chem.*, **26**, 4600 (1961).

(4) J. F. Mahoney and C. B. Purves, *J. Am. Chem. Soc.*, **64**, 9 (1942); T. S. Gardner and C. B. Purves, *ibid.*, **64**, 1539 (1942); A. N. DeBelder, B. Lindberg, and O. Theander, *Acta Chem. Scand.*, **16**, 2005 (1962).

acetamido derivative was less dextrorotatory (+50.5°) than the di-*O*-*p*-tolylsulfonyl precursor (+91.5°), a fact which may be due to the expected inversion at C-2(3) during the hydrazinolysis reaction.[5]

The solubility behavior of these two modified polysaccharides is of interest. The aminated amylose was soluble only in dilute acids and no true solvent could be found for it. The *N*-acetylated aminated amylose, on the other hand, could be dispersed in water and was soluble also in dimethyl sulfoxide although not in *N*,*N*-dimethylformamide. The water solubility of the acetamido derivative would suggest that the hydrophilic amide groups are located in sterically favorable positions for solvation of the polysaccharide, in contrast to chitin where intermolecular hydrogen bonding[2] promotes insolubility.

Periodate oxidation of *N*-acetyl aminated amylose showed that approximately 30% of the pyranose residues contain contiguous hydroxyl groups at C-2 and C-3. Determination of the 5-(hydroxymethyl)-2-furaldehyde formed during acid degradation of the polymer, by the method of O'Neill and co-workers,[6] demonstrated that about 16% of the pyranose units contain the 3,6-anhydro ring. Such a ring would be formed by base-catalyzed elimination of the *p*-tolylsulfonyloxy group on C-6. Closure through a reaction involving a sulfonate ester on C-3 and a free hydroxyl on C-6 would be sterically impossible if inversion is involved and improbable since there is good evidence[3] that all the C-6 hydroxyl groups would be sulfonated. A chain structure of ten monomeric units which would accommodate the known facts is shown in I. In actuality the sequence of units in I could be expected to be random.



I, Aminated amylose

Work is in progress in this laboratory to substantiate the preceding formulation by degradative procedures.

Experimental

2(3),6-Di-O-*p*-tolylsulfonylamylose.—Amylose (Superlose, HAA-11-HV, High Viscosity, Control No. 12215, Stein-Hall and Co., Inc., New York, N. Y.), 324 g., was dissolved in 95% aqueous pyridine (2 l.) and water was removed azeotropically at 60° under reduced pressure. To the pyridine-swollen amylose was added *p*-toluenesulfonyl chloride (850 g., 2.2 equiv.) portionwise during 2 hr. The mixture was maintained at room temperature for 24 hr., then agitated with ice and methanol in a blender, and the resultant white powder was washed several times with water and dried; average yield, 720 g., $[\alpha]^{20}D$ +91.5° 2.1, dimethyl sulfoxide).

Anal. Calcd. for $[C_6H_7O_2(OH)_{1.3}(OSO_2C_6H_4CH_3)_{1.7}]_n$: C, 49.97; H, 4.75; S, 12.78. Found: C, 50.06; H, 4.51; S, 12.10.

Aminated Amylose.—2(3),6-Di-*O*-*p*-tolylsulfonylamylose (50 g.) was refluxed with anhydrous hydrazine (700 ml.) under nitrogen for 7 days. Excess hydrazine was removed under reduced pressure, the residue added to water (500 ml.), and the solution stirred with Raney nickel (20 g.) until the evolution of ammonia ceased (24 hr.). Stirring was continued and the temperature was slowly raised to 100°. The catalyst was filtered. The filtrate was concentrated to 200 ml., dialyzed against tap water for 3 days, and then against distilled water for 3 days. Lyophilization of the solution produced the aminated amylose as a white, nonhygroscopic powder; yield, 9.5 g. This product was readily soluble in dilute acids and was insoluble in water, dimethyl sulfoxide, and *N*,*N*-dimethylformamide.

Anal. Calcd. for $[C_6H_7O_2(NH_2)_{1.4}(-O-)_{0.5}(OH)(OSO_2C_6H_4CH_3)_{0.1}]_n$: N, 11.90; S, 1.94. Found: N, 11.48; S, 1.72.

N-Acetyl Aminated Amylose.—Aminated amylose (9.0 g.) was suspended in water (100 ml.) and stirred while acetic anhydride (50 ml.) was added. Stirring was continued until the solution became clear (1 hr.). After standing at room temperature overnight, the reaction mixture was dialyzed against tap water for 3 days and then against distilled water for 3 days. The solution was concentrated to 200 ml. and lyophilized to give a white, hygroscopic powder; yield, 11.5 g.; $[\alpha]^{20}D$ +50.5° (*c* 0.7, dimethyl sulfoxide). The *N*-acetyl aminated amylose was soluble in dimethyl sulfoxide and in dilute acids and swelled into water to give a clear colloidal solution. It was insoluble in *N*,*N*-dimethylformamide.

Anal. Calcd. for $[C_6H_7O_2(NHCOCH_3)_{1.4}(-O-)_{0.5}(OH)_{1.1}]_n$: N, 8.93, CH_3CO; 26.84. Found: N, 9.39; CH_3CO, 26.52; S, 0.1.

Periodate Oxidation of *N*-Acetyl Aminated Amylose.—To a sample of *N*-acetyl aminated amylose (0.1 g.) in water was added 5 ml. of an aqueous solution of sodium metaperiodate (0.3 *M*, 3.5 equiv.) and the volume was brought to 100 ml. A blank determination was prepared by omitting the sample. The solutions were maintained at room temperature in the dark and aliquots were analyzed at intervals for periodate consumption by the method of Neumuller and Vasseur.[7] Samples (5 ml.) were added to a mixture of phosphate buffer (25 ml., pH 6.98) and 20% potassium iodide (2 ml.). The resulting iodine was titrated with 0.01 *N* sodium thiosulfate, using starch as indicator. The periodate consumption in moles per saccharide unit was (time in hr., moles of oxidant consumed per hexose unit): 0.5, 0.30; 2, 0.31; 5, 0.32; 7, 0.35; 12, 0.37; 24, 0.44.

Determination of 3,6-Anhydrohexose Units in N-Acetyl Aminated Amylose.[8]—Ten samples (10 mg. each) of *N*-acetyl aminated amylose were mixed with 0.15 *N* sulfuric acid (2 ml.) and hydrolyzed in sealed tubes at 100°. Tubes were removed at intervals during a 24-hr. period, the contents were neutralized with barium carbonate, filtered into 50-ml. volumetric flasks, and diluted to 50 ml. Optical densities were read at 2850 Å. To correct for the first-order decomposition[8] of 5-(hydroxymethyl)-2-furaldehyde to formic acid and levulinic acid during the hydrolysis, the logarithm of the optical density was plotted against time and the linear portion of the curve was extrapolated to zero time. The 5-(hydroxymethyl)-2-furaldehyde detected corresponded to 11.8% of the polysaccharide or 16.4% of 2-acetamido-3,6-anhydro-2-deoxyhexopyranose units.

(5) R. U. Lemieux and P. Chu, *J. Am. Chem. Soc.*, **80**, 4745 (1958); M. L. Wolfrom, F. Shafizadeh, R. K. Armstrong, and T. M. Shen Han, *ibid.*, **81**, 3716 (1959).

(6) A. N. O'Neill, *ibid.*, **77**, 2837 (1955); D. B. Smith, A. N. O'Neill, and A. S. Perlin, *Can. J. Chem.*, **33**, 1352 (1955).

(7) G. Neumüller and E. Vasseur, *Arkiv. Kemi*, **5**, 235 (1953).

(8) H. P. Teunissen, *Rec. trav. chim.*, **49**, 784 (1930); **50**, 1 (1930).

A Self-Condensation Reaction of 2- and 4-Hydroxymandelamines[1]

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Received July 18, 1963

Octopamine (I) [norsympatol, norsynephrine, (α-aminomethyl)-4-hydroxybenzyl alcohol] has been