

*Procedure.* A solution of 38 g NaOH (0.95 mole) in 1 200 ml water is saturated with  $\text{CO}_2$ . The resulting solution of  $\text{NaHCO}_3$  is slowly added to a solution of 136 g (0.80 mole)  $\text{AgNO}_3$  in 400 ml water under vigorous shaking. This operation and the following must take place under exclusion of day-light (red light is permissible). The  $\text{Ag}_2\text{CO}_3$  precipitate is separated from the  $\text{NaNO}_3$ -solution by decantation and afterwards washed four times with 500 ml water which is also removed by decantation. The remaining, moist  $\text{Ag}_2\text{CO}_3$  is transferred to a platinum dish where 47 g 40 % HF (0.94 mole) is added in small portions under good stirring. When the initial, rapid evolution of  $\text{CO}_2$  has ceased the reaction is finished by heating for thirty minutes on a steam-bath. The AgF-solution is separated from excess  $\text{Ag}_2\text{CO}_3$  by filtering through ash-free filter-paper on a paraffined funnel into a paraffined beaker and afterwards placed in the (weighed) platinum dish again. The water is evaporated on a steam-bath under continued stirring until the weight of the sample is 115 g. The contents of the dish are now a mixture of about 90 g AgF (partly separated as crystals) and about 25 g water. In order to remove the water rapidly and completely 100 ml anhydrous methanol is added and, after thorough stirring, removed by decantation together with most of the water. The treatment with methanol is repeated twice and followed by a similar treatment with 100 ml anhydrous ethyl ether three times. The silver fluoride (moist with ether) is now quickly transferred to a round-bottomed glass flask with a side tube through which most of the ether is removed by a water aspirator. The last trace is evaporated by heating to 60–70° C in a water-bath and applying an oil pump. Yield: 75 g (70–80 g) of a light-brown powder.

The ether, used at the decantation, is added to the methanol together with 300 ml extra ether. A nice, yellow precipitate is separated from the mother liquor by decantation and washed twice with 25 ml anhydrous methanol and three times with 50 ml anhydrous ether. The last ether is removed as above. Yield: 15 g (14–16 g). The two crops are almost of the same quality although they differ in colour. Total yield: 90 g (0.72 mole) or 89 %.

*Properties.* AgF is highly hygroscopic and very sensitive to day-light. It attacks glass only very slowly at room temperature. It is stored in dark, paraffined vessels. The purity of the preparation was checked by determining the silver and the

fluorine equivalent weights. For the brown and the yellow sample the silver equivalent weight was 126.4 and 127.3, respectively, (theory 126.9) while the fluorine determinations gave 124.4 and 122.0. 1 000 ml anhydrous methanol dissolves 14.5–15 g AgF at room temperature. In the dark and stored on a platinum container the solution is stable for at least 24 hours.

The method for the preparation of AgF here indicated is preferable to removing the water from an aqueous solution in a desiccator, a procedure which does not result in a well-defined product within a reasonable period.

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## The Question of Furanosidic Bonds in Starch

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In a recent study of the hydrolysis of starch with acids and with  $\beta$ -amylase, Blom and Schwartz<sup>1</sup> find that the change in specific rotation per mole hydrolysed bond,  $[\text{S}_p]_D$ , is 166–187° at the beginning of hydrolysis, the mean value for total hydrolysis 232° and the values for maltose and isomaltose 280° and 221°, respectively. They conclude that as the value for the first bonds hydrolysed is much lower than the value for maltose, these bonds must be of a quite different type, and assume, therefore, that they are furanosidic (1 : 4), and that the next glucose unit in the starch molecule is linked by a 1 : 5-bond.

The authors appear, however, to have overlooked the great differences between terminal and central bonds with respect to their contribution to the rotation. If, in a linear polysaccharide containing uniform bonds every second bond ( $n/2$  bonds) is

hydrolysed,  $n/2$  molecules of the corresponding biose is obtained. If now the biose is hydrolysed, the net result is a total hydrolysis of the polysaccharide, and it is evident that the mean value of  $[\text{Sp}]_D$  is the mean of  $[\text{Sp}]_D$  for the central bond and that for the biose. For starch the  $[\text{Sp}]_D$  for central bonds is thus calculated as  $184^\circ$ , in good agreement with the value found by Blom and Schwartz. In the series cellobiose to celloheptaose<sup>2</sup> and xylobiose to xylohexaose<sup>3</sup> there are also great differences between the rotation contributions of central and terminal bonds. The plot of  $[\text{M}]_n/n$  against  $n-1/n$ , where  $[\text{M}]_n$  is the molecular rotation and  $n$  the degree of polymerization, yields a straight line, however, which according to Freudenberg<sup>4</sup> means that the linkages in the series are uniform. In the maltose series only the next member, maltotriose, is known<sup>5</sup>, but from the molecular rotation of this substance ( $+807^\circ$ ), of maltose ( $+470^\circ$ ), and of glucose ( $+95^\circ$ ), the  $[\text{Sp}]_D$  for a central bond is calculated as  $204^\circ$ .

Thus Blom and Schwartz have merely confirmed experimentally the expected value of  $[\text{Sp}]_D$  for central bonds in the starch molecule. They have not adduced any evidence supporting the occurrence of furanosidic bonds in starch. If the mere fact that the  $[\text{Sp}]_D$  of a central bond is different from that of a terminal bond should constitute an argument for this assumption, it could equally well be applied to all tri- and higher saccharides.

1. Blom, J., and Schwartz, B. *Acta Chem. Scand.* 6 (1952) 697.
2. Wolfrom, M. L., and Dacons, J. C. *J. Am. Chem. Soc.* 74 (1952) 5331.
3. Whistler, R. L., and Chen-Chuan Tu. *J. Am. Chem. Soc.* 74 (1952) 4334.
4. Freudenberg, K. "Tannin, Cellulose and Lignin", J. Springer, Berlin 1933, p. 104.
5. Sugihara, J. M., and Wolfrom, M. L. *J. Am. Chem. Soc.* 71 (1949) 3357.

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## Crystalline Rhodanese

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Recently we reported a method for the partial purification of rhodanese<sup>1</sup> from beef liver. The enzyme has now been further purified and obtained in the crystalline state.

The method of purification was as follows: Beef liver was extracted and the extract fractionated with ammonium sulfate at pH 3.8 and 7.8 as described before<sup>1</sup>. The enzyme was then dialyzed against 0.01 *M* sodium acetate and the pH of the dialyzed solution adjusted to 4.9. Fractionation with acetone was then carried out at  $-5^\circ\text{C}$  and the precipitate appearing between 30 and 50 % by volume acetone was collected and dissolved in cold 0.01 *M* sodium acetate. The remaining acetone was removed by dialysis against 0.01 *M* sodium acetate. The pH of the solution was then adjusted to 4.5, the enzyme precipitated with ammonium sulfate at 40 % saturation and the precipitate dissolved in 0.01 *M* sodium acetate. When a 0.5 % solution of this preparation was examined in the "Spinco" ultracentrifuge of this department, only one homogeneous boundary with  $S_{20} = 2.76 \cdot S$  was observed. The solubility of a similar preparation in ammonium sulfate of varying concentration was then studied at room temperature and pH 5. The logarithm of solubility for the enzyme was found to be proportional to the concentration of ammonium sulfate. An amorphous precipitate was obtained, but when those supernatants, which still contained enzymatic activity were brought to  $+2^\circ\text{C}$ , a pronounced prethixotropy slowly developed. No crystals were, however, visible in the microscope, but later experiments showed, that rhodanese could be crystallized from ammonium sulfate at