which ergosterol had been adsorbed gave an immediate violet coloration on contact with traces of β-chlorovinyldichloroarsine vapor, changing to a deep green in the presence of larger amounts. A similar but much fainter color change was produced by β,β'-dichlorodiethylsulfide only upon prolonged exposure of the adsorbate to air saturated with its vapor. Hydrochloric acid vapor caused the reagent to change to a deep rust brown color.

Experimental

To 15 cc. of purified chloroform was added 50 mg. of ergosterol (Montrose Chemical Company) and the temperature of the mixture raised to the boiling point for one minute. Dry, white silica gel, 80-mesh (Davison Chemical Corporation), 5 g., was added and the mixture allowed to stand for a minute; the solid was then separated from the supernatant liquid and spread out upon a filter paper to dry. As a result of this procedure the silica gel mixture became a pale steel gray; when heat was applied to facilitate the evaporation of the solvent a product varying in color from orange to green was obtained and in such cases the sensitivity of the test was greatly impaired. The dry material was loaded into glass tubes 4 mm. in diameter in columns about 1 cm. long, held in place with glass fiber plugs. Tubes so prepared were evacuated by means of an oil pump and sealed under vacuum. In use, the ends of the tubes were broken open and gas drawn through the impregnated gel with a rubber atomizer bulb, the valves of which had been reversed in such a manner as to provide suction instead of pressure.

A tube prepared and used in this manner indicated the presence of 10 micrograms of β-chlorovinyldichloroarsine in 150 cc. of gas by means of a stripe of readily visible color (violet to green) at the zone of first contact. Ammonia vapor converted this stripe to an orange band, while the unaffected part of the reagent turned straw yellow.

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BETHESDA, MARYLAND

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Some Crystalline Forms of Amylose

BY RALPH W. KERR

Amylose has been reported to crystallize in various forms depending upon conditions, particularly the type of starch from which the amylose fraction is prepared and the solvent medium from which the fraction crystallizes. Schoch[1] noted that when an autoclaved starch solution is treated with an excess of butyl alcohol, a slow precipitation of amylose in the form of minute, semi-crystalline spheroids occurs at the alcohol–water interface. Those from corn starch[2] are described as six segmented spherulites about 15–50 microns in diameter and those from potato as well-formed, six-petalled rosets, 50-80 microns in diameter or occasionally clumps of hair-like needles. Kerr and Severson[3] crystallized that portion of amylose which is leached from starch by warm water, using butyl alcohol–water as the crystallizing medium and obtained rectangular platelets from corn and needles which formed star-like clusters from potato starch. Subsequently, needles in star-like clusters were obtained from tapioca starch by a similar procedure.[4] Wiegel[5] refluxed potato starch with 30% ethanol and obtained needle-like particles which on closer observation appeared to be very thin platelets. When the ethanol was evaporated from the extract and butyl alcohol added to the hot solution, the crystals which separated assumed an hour-glass shape. Aqueous isobutyl alcohol gave typical sphero-crystals.

Kerr and Severson believe that all amyloses are chemically heterogeneous and that the material obtained in a warm water extract of whole starch represents a subfraction of the particular amylose, which is possibly representative of the less branched and lowest degree of polymerization material in the total fraction. Thus, a warm water extract of corn starch contains a predominant proportion of relatively short unbranched amylose chains and these crystallize eventually, after careful purification, as well-formed rectangular platelets. Corresponding subfractions from potato and tapioca crystallize as needles but it is suggested by Kerr[4] that even these may be slightly branched and of greater molecular weight.

It becomes desirable to know whether amylose crystallizes from butyl alcohol–water mixtures in shapes which are largely determined by minor variables of the crystallization technique or whether an amylose, such as corn amylose, can be subfractionated so as to obtain crystal shapes which are inherently related to the predominating structure in these subfractions and its orientation with butyl alcohol. This report proposes a method for subfractionating the total butyl alcohol precipitate of starch and describes the crystal forms which are characteristic of the subfractions obtained from corn amylose.

Experimental

Nine grams of sphero-crystals of corn amylose (8.4 g. dry basis) which had been obtained from defatted corn starch by the method of Schoch[2] and by recrystallization from an aqueous butyl alcohol solution, was added with stirring to 183 ml. of ethylenediamine in a closed vessel at room temperature. After several days, the amylose had completely dissolved. Absolute ethyl ether was added dropwise with shaking at 25° until a distinct cloudiness developed. This required adding 53 ml. of ether. The flask was shaken intermittently for two days, whereupon the upper phase, richer in ethyl ether, was removed and poured into 500 ml. of absolute ether. The precipitate was washed by long standing in two additional 500-ml. portions of ether and decanting and then air dried to evaporate the ether. Then 300 ml. of warm water was added, the *p*H carefully adjusted to 6.2 with hydrochloric acid, the solution heated to 80°, filtered, saturated with butanol and slowly cooled over a period of several days. The crystals obtained were rectangular platelets.[6] These were centrifuged at 2000 r. p. m. and washed by standing

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[1] T. J. Schoch, *Cereal Chem.*, **18**, 121 (1941).

[2] T. J. Schoch, THIS JOURNAL, **64**, 2957 (1942).

[3] R. W. Kerr and G. M. Severson, *ibid.*, **65**, 193 (1943).

[4] R. W. Kerr, "Chemistry and Industry of Starch," Academic Press, Inc., New York, N. Y., 1944, p. 147.

[5] E. Wiegel, *Kolloid-Z.*, **102**, 145 (1943).

[6] See ref. 4 for figure illustrating platelet-shaped crystals of corn amylose.

for twenty-four hours each in two portions of 200 ml. of water saturated with butanol. The crystals were dehydrated and freed of butyl alcohol by prolonged standing in five successive portions of 200 ml. of absolute methanol, filtered, and the methanol evaporated in a vacuum desiccator. The yield was 2.14 g.

The subfraction procedure was repeated by adding 100 ml. of ethylenediamine to the sirupy, residual phase, poorer in ether and then adding 32 ml. of ether. The upper phase which separated was removed and the amylose crystals prepared as indicated above. The yield was 1.53 g. of thick platelets.

In the third step of the subfractionation procedure, 90 ml. of ethylenediamine was added followed by 29 ml. of ether. The yield was 1.45 g. of blunt needles.

Finally, the residual, lower phase from the third, phase separation procedure was treated with a large excess of ether to precipitate the remaining amylose. After crystallization from water–butyl alcohol, this subfraction formed well-shaped, thin needles which arranged themselves in star-like clusters and appeared very similar in shape to those described for the crystalline amyloses of tapioca and potato starches obtained by the addition of butyl alcohol to warm water extracts of the starches.[4] This yield was 1.82 g.

The subfraction technique was repeated several times and the results were the same as reported above. In one of these duplicate subfractionations, a small amount of needles (in water–butyl alcohol from the first butyl alcohol crystallization of the fourth subfraction) was added as seed to a solution of the first subfraction in water–butyl alcohol. On standing, however, platelet shaped crystals formed as usual. The complementary experiment was performed wherein platelets (the first subfraction) were added to a crystallization of the fourth subfraction. Needle-shaped crystals formed as usual.

Qualitatively the solution viscosity in ethylenediamine of the platelets, obtained by subfractionation of corn amylose sphero-crystals and from a warm water extraction of corn starch are of the same order of magnitude; also, those of the needles obtained from water extracts of tapioca starch and from the subfractionation of corn amylose are the same. The viscosity of the total butyl alcohol precipitate of corn starch in ethylenediamine is intermediate to those two ranges.

An analysis of the various subfractions obtained from corn amylose by the use of ethylenediamine—ether phase separation as well as a study of the application of the ethylenediamine–ether phase separation method as a fractionation procedure for whole starch is proceeding in our laboratories.

RESEARCH LABORATORIES
CORN PRODUCTS REFINING COMPANY
ARGO, ILLINOIS
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α-Methoxycrotonic Acid

BY KARL PFISTER, C. A. ROBINSON AND MAX TISHLER

The recent publication of L. N. Owen[1] on the "Reactivity of α-Bromo-crotonic Acid" prompts us to report at this time our results of a similar study. As indicated in our report, the conclusions are on the whole in agreement with those of Owen.

Our investigation began with an attempt to prepare tetrolic acid by the action of methanolic potassium hydroxide on crotonic acid dibromide under conditions described by Reimer[2] for the preparation of phenylpropiolic acid from cinnamic acid dibromide.

Under these conditions, crotonic acid dibromide was converted to α-methoxycrotonic acid[3] in 80% yield (lustrous rods, m. p. 60–62°; calcd. for $C_5H_8O_3$: C, 51.72; H, 6.95; neut. equiv., 116.1. Found: C, 51.65; H, 6.83; neut. equiv., 115.2). The *p*-bromophenacyl ester derivative melts at 76–77°; calcd. for $C_{13}H_{13}O_4Br$: C, 49.84; H, 4.18. Found: C, 50.02; H, 4.10. α-Methoxycrotonic acid decolorizes bromine and potassium permanganate rapidly. It is very soluble in water, ether and carbon disulfide but sparingly soluble in ligroin. Catalytic hydrogenation (palladium Norite catalyst) converted it into α-methoxybutyric acid which was isolated and characterized by its *p*-bromophenacyl ester derivative (m. p. 69–70°; calcd. for $C_{13}H_{15}O_4Br$: C, 49.53; H, 4.80. Found: C, 49.26; H, 4.54). That the last mentioned compound is not the *p*-bromophenacyl ester of β-methoxybutyric acid was established by preparing the same ester of an authentic sample of β-methoxybutyric acid[4] and comparing the two. The *p*-bromophenacyl ester of β-methoxybutyric acid melts at 59–60° (found: C, 49.35; H, 4.72) and depresses the melting point of the isomeric α-methoxy compound.

Identical treatment of α-bromoisocrotonic acid with methanolic potassium hydroxide gave α-methoxycrotonic acid in 87% yield. However, a much longer reaction time was required to completely remove the halogen from α-bromocrotonic acid (70% yield of α-methoxycrotonic acid). The difference in reaction rates of these two compounds may be due to the limited solubility of the potassium salt of the *trans* acid under the conditions of our experiments and may not reflect the relative reactivities of the halogen atoms in α-bromocrotonic and α-bromoisocrotonic acids.

As one would expect, α-methoxycrotonic acid is hydrolyzed by boiling with dilute hydrochloric acid to α-ketobutyric acid. The latter was identified by conversion to the known phenylhydrazone (m. p. 149–150°). The hydrolysis together with the preparation of the α-alkoxy-α,β-unsaturated acids may find use as a convenient synthesis of aliphatic α-keto acids.

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[1] L. N. Owen, *J. Chem. Soc.*, 385 (1945).

[2] Reimer, THIS JOURNAL, **64**, 2510 (1942).

[3] In connection with this reaction, it is of interest to note that treatment of diethyl α,α'-dibromosuccinate with sodium ethylate in ethanol yields some diethyl ethoxyfumarate. *Cf.* Michael and Maisch, *J. prakt. Chem.*, [2] **46**, 235 (1892); Michael and Bucher, *Ber.*, **29**, 1792 (1896).

[4] Prepared by the method of Purdie and Marshall, *J. Chem. Soc.*, 468 (1891).

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Electrolyte Catalysis in the Ammonolysis of 9-Phenyl-9-chlorofluorene by Liquid Ammonia, Alkaline-Earth Nitrates

BY WILBUR F. ROPER,[1] ROBBIN C. ANDERSON AND GEORGE W. WATT

It has been shown that the ammonolysis of 9-phenyl-9-chlorofluorene by liquid ammonia is an autocatalytic reaction, the ammonium chloride produced acting as catalyst,[2] and, further, that this is not specific "acid catalysis"; but that electrolytes in general seem to catalyze the re-

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[1] Present address: C. A. S. Co. 96, U. S. Army.

[2] Williamson, Anderson and Watt, THIS JOURNAL, **65**, 49 (1943).