Carboxymethyl High-Amylose Starch

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GENETICALLY increasing the proportion of amylose in cornstarch has resulted in a starch with potentially wider uses in such fields as films, fibers, and coatings. In the course of preparation and characterization of carboxymethyl derivatives with degree of substitution (D.S.) 0.1 to 0.9, we have found carboxymethylation effective in producing cold water solubility and in preventing retrogradation of the 57% amylose cornstarch.

Starch used in this study was obtained from corn supplied by the Bear Hybrid Corn Co., Inc., Decatur, Ill., and was isolated in the pilot plant of this laboratory by conventional wet-milling methods (*1, 15*). Analysis of the starch showed an amylose content (by iodine sorption) of 57% and a nitrogen content of 0.12%.

Preparation of carboxymethyl derivatives of cellulose, starch, and inulin is described by Chowdhury (*4*). Others (*11-13*) used the same reactions in aqueous media with subsequent recovery of products by precipitation in alcohol or by a "dry" process (*3*). Filbert (*7*) carboxymethylated potato starch in a medium containing sufficient ethanol to prevent solubilization. This method, or slight variations of it, proved the least troublesome as a laboratory procedure, although it would not necessarily be chosen for large-scale operations.

A series of carboxymethylated high-amylose starches was prepared covering a range of degree of substitution. For a typical preparation, 100 grams (dry basis) of 57% amylose starch was slurried in 250 ml. of absolute ethanol in a 2-liter, 3-necked, round-bottomed reaction flask equipped with a sealed-glass stirrer, reflux condenser, and dropping funnel. The amount of monochloroacetic acid (slightly more than theoretical) found necessary to give the desired D.S. had been previously dissolved in the ethanolic reaction medium. The stirred slurry was brought to reflux temperature and held there throughout the dropwise addition of an ethanolic NaOH solution. Refluxing was continued 15 minutes; then the product was recovered by filtering and washing with 80 to 85% ethanol until free of chloride. The NaOH solution was made by dissolving 2.5 moles of NaOH per mole of $ClCH_2COOH$ used in a small amount of water, then mixing with sufficient absolute ethanol to make a 90% ethanolic solution and filtering. For example, 45 grams of monochloroacetic acid were used with 50 grams of sodium hydroxide dissolved in 400 ml. of 90% ethanol and reacted with 100 grams of starch to give a product with a D.S. about 0.75. Preparations were also made by this procedure from fractionated amylose, ordinary starch, and waxy starch for comparison.

Some preparations were analyzed for D.S. by the method of Eyler, Klug, and Diephuis (*6*), but when the D.S. was above 0.5, the product was too soluble in 80% methanol to permit a reliable determination. An alternative method was devised in which ion exchange was used in place of acid washing to de-ash the sample and, incidentally, to reduce the time required for an analysis. Small samples, approximately 20 mg., were dissolved in about 25 ml. of distilled water by warming or by adding dilute NaOH, if necessary. The solution was then washed through a cation-exchange column holding about 1 gram of Dowex-50 resin in the acid form; effluent was collected in a tared beaker. This solution was evaporated to dryness on a steam bath repeatedly until no chloride ion could be detected; then the beaker and residue were cooled in a desiccator and weighed. The residue was taken up in 0.01*N* NaOH and back-titrated with standardized 0.01*N* HCl to the phenolphthalein end point. Comparison with a blank titration provided a basis for calculation of milliequivalents of acid per gram of sample. From this, D.S. was calculated as follows:

$$\frac{0.162 \times \text{meq. COOH/g.}}{1 - (0.058 \times \text{meq. COOH/g.})} = \text{D.S.}$$

Duplicate determinations agreed well from the lower solubility limit to a D.S. above 1.0.

Polyelectrolytic behavior of the high-amylose derivatives was examined in an Ostwald-Cannon-Fenske viscometer at two levels of substitution and compared with similar data obtained from derivatives of ordinary starch and fractionated amylose. Salt-free solutions of the carboxymethyl derivatives were made by dissolving them in 0.5*N* NaOH at room temperature, then dialyzing against distilled water until equilibrium was reached at pH 6.9 to 7.0. The solution was made up to 0.004*M*. as measured by evaporating an aliquot to dryness and weighing. This stock solution was then mixed with a 0.002*M* NaCl stock solution in various proportions and dilutions; the capillary flow time of each combination was measured at 25.1° ± 0.03° C. From these data, curves were plotted using the logarithmic viscosity number *vs.* the concentration in grams per deciliter. Shown in Figure 1 is a family of these curves for a sample of 57% amylose starch derivative with D.S. 0.94. The flattening of the curve with addition of salt is characteristic of a polyelectrolyte. Carboxymethyl derivatives of amylose and ordinary starch with comparable D.S. gave similar families of curves, as did carboxymethyl high-amylose starches with lower D.S.

To circumvent the difficulty of extrapolating the curves to zero concentration to find intrinsic viscosity, the same data were also plotted according to Fuoss and Strauss (*8*) (Figure 2). The ratio of specific viscosity to square root of molar concentration *vs.* square root of molar concentration gave a family of straight lines in which the slope decreased with increased salt concentration. Extrapolation of these curves showed convergence on the *Y*-axis (zero concentration) at a point below zero. $\eta_{sp}/(C)^{1/2} = A + B(C)^{1/2}$, where *A* is the intercept on the *Y*-axis and *B* is the slope of the curve. *A* can be plus, minus, or zero according to Jones and Dole (*10*) and Dolian and Briscoe (*5*), but negative values have no significance, according to Harned and Owen (*9*). Thus, our negative values for the *Y*-intercept give no real values for intrinsic viscosity, although the characteristics of the curves are those of a polyelectrolyte. As in the case of logarithmic viscosity number *vs.* concentration, the carboxymethyl derivatives with D.S. 0.1 and 0.4 gave families of curves similar to those shown for D.S. 0.9.

Temperature-viscosity relationships were investigated by means of the Brabender Amylograph-Viscograph. Carboxymethyl derivatives of the high-amylose starch, of ordinary cornstarch (27% amylose), and of fractionated corn amylose were run at various concentrations and D.S. The heating cycle started at 25°, rose 1.5° per minute to 90°, was held 15 minutes at 90°, and then fell 1.5° per minute to 25°. Viscosity was continuously recorded in Brabender units, and the curves were later redrawn on a more convenient scale. When salt-free 2% solutions were compared in this way, two derivatives of high-amylose starch (D.S. 0.94 and

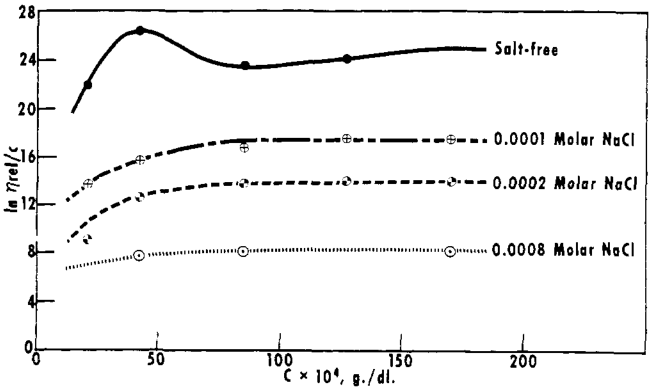


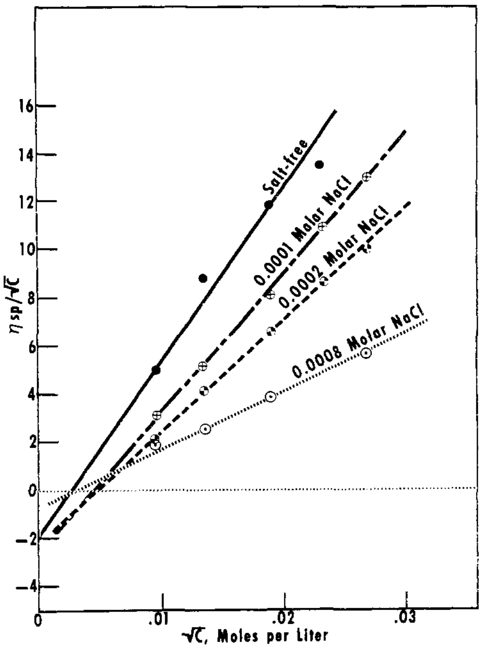
Figure 1. Logarithmic viscosity number vs. concentration of carboxymethyl high-amylose starch

Degree of substitution 0.94

⟶

Figure 2. Specific viscosity–concentration quotient vs. concentration of carboxymethyl high-amylose starch

Degree of substitution 0.94



0.25) and one of amylose (D.S. 0.81) showed a much lower viscosity than similar derivatives of cellulose and ordinary starch. The carboxymethyl amylose curve was lowest, varying between 9 and 14 Brabender units. The curves of high-amylose starch derivatives were only slightly above the carboxymethyl amylose curve, and the fact that this difference is not greater probably reflects the great influence exerted by the larger proportion of amylose in this new type of starch.

The effect of D.S. on viscosity is shown in Figure 3. These preparations were used without treatment after isolation; therefore, the solutions had a pH of 11 or higher. The preparations were stable to this alkalinity, for all regained their original viscosity on cooling. The low modification (D.S. 0.1) was not cold water-soluble but required a temperature of 50° to 60° for dispersion. Not shown is a sample with D.S. 0.05 that was not soluble in boiling water. The curve for the high modification (D.S. 0.9) lies between those for the low and intermediate modifications—no ready explanation is available.

Although the solutions used in the Amylograph appeared to be thoroughly dispersed, some settling out was noted prior to measurement of setback (Table I). Settling volume was calculated as the ratio of depth of translucent layer (beneath a transparent layer) to total depth of liquid in a parallel-sided vessel. Setback was measured by replacing the solution in the cup of the Amylograph and operating the instrument at 25° long enough to get a stable tracing. In most instances a slight correction was made in the reading to compensate for some transfer losses of solution.

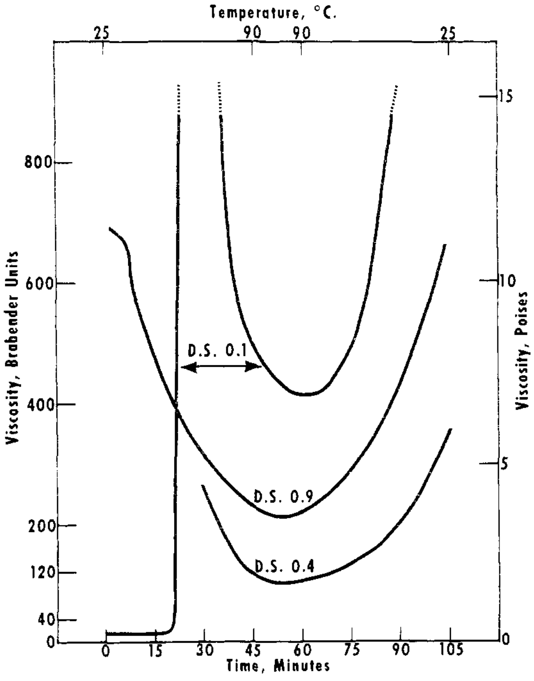


Figure 3. Viscosity–degree of substitution relationship for carboxymethyl high-amylose starch

10% concentration

POTENTIAL APPLICATION

Soil-suspending power of sodium carboxymethyl high-amylose and ordinary starches was evaluated by a method based on that of Bayley and Weatherburn (*2*) and of Weatherburn, Rose, and Bayley (*14*). The best high-amylose starch derivative (D.S. 0.7) had 66% of the soil-suspending power of a commercial carboxymethylcellulose and was considerably above any of the ordinary starch derivatives tested.

Carboxymethyl high-amylose starch at three levels of modification was evaluated as a beater additive in paper sizing. Tests run on hand sheets showed no improvement in paper quality. Paper was coated with the high-amylose

Table I. Amylograph Viscosity of Carboxymethyl High-Amylose Starch

D.S.	% Solids	Amylograph Viscosity, Brabender Units 90°	55°	25°	Setback[a]	Settling Volume
0.9	2	15	20	25	Nil	...
0.9	5	65	100	180	200	0.7
0.9	10	220	360	660	...	0.9
0.4	5	40	60	95	100	0.96
0.4	10	105	165	360	360	1.0
0.1	5	115	145	300	...	1.0
0.1	10	415	630	1000	...	1.0

[a] At approximately 2 months.

starch derivatives at D.S. 0.3 and 0.9 from 5% neutralized solutions and then dried and evaluated. Tear factor and wet breaking length were decreased, but bursting strength was increased about 50%, dry breaking length over 20%, and folding endurance was 2 to 3 times that of the uncoated paper.

DISCUSSION

High-amylose cornstarch was readily soluble in hot water when carboxymethylated (CM) to 1 substituent group per 10 anhydroglucose units (AGU); it was cold water-soluble at 2 or more CM groups per 10 AGU. The product exhibited typical polyelectrolyte properties, and its viscosity in solution was markedly reduced by salts. The viscosity was generally less than that of similar derivatives of ordinary starch or cellulose, but good stability was noted over the heating and cooling range of the Brabender Amylograph in both neutral and highly alkaline solutions. Some settling out of solution was noted, but setback was negligible.

Although unsatisfactory as a paper size, the carboxymethyl high-amylose starches had fairly good soil-suspending power. The inherent film-forming property of a high-amylose content material was displayed by the increase in tensile and bursting strength and in folding endurance of paper coated with this derivative.

ACKNOWLEDGMENT

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Density and Viscosity of Anhydrous Hydrazine at Elevated Temperatures

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THE VISCOSITY and density of hydrazine were determined experimentally from 288.16° and 296.24° K., respectively, to 449.83° K. The published data now cover a temperature range which extends into the autodecomposition region. Predictions of the density and viscosity of hydrazine, within 0.1% accuracy, may be made over the temperature range of the present investigation from correlations of the experimental data.

The advancement of hydrazine in recent years from the role of laboratory curiosity to a compound of increasing importance in the commercial and scientific world necessitated broadening of the temperature range over which the physical properties are known. The previously reported maximum temperatures at which the density and viscosity of hydrazine were known were 323.2° K. (*7*) and 344.3° K. (*6*), respectively.

This article presents the results of experimental measurements of the density and viscosity of hydrazine to temperatures extending into the autodecomposition region. A very careful application of standard laboratory methods enabled measurements of high accuracy. All experimental work was done in a completely inert atmosphere, to minimize the alteration of hydrazine purity by absorption of water and reaction with oxygen or carbon dioxide.

EXPERIMENTAL

Hydrazine used was of a purity greater than 99.6% by weight. The hydrazine used in the density determinations at 310.26° and 338.73° K. was of 99.9% purity at the start of the series, but picked up sufficient water to reduce purity to 99.6%. This sample had been prepared by stirring commercial-grade (about 97%) hydrazine over barium oxide for several hours, vacuum distilling, and twice passing the condensate through a packed column of Linde Molecular Sieves, Type 5A. Hydrazine used in the remainder of the