STRUCTURE OF THE STARCH GRANULE

II. Action of Various Amylases on Granular Starches[1]

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ABSTRACT

The dissolving action of various amylases on granular starches has been studied, with particular attention to bacterial alpha-amylase. The following starches are listed in order of increasing resistance to amylase: waxy maize, tapioca, waxy sorghum, sorghum, corn, wheat, rice, sago, arrowroot, potato, heat-moisture-treated potato, high-linear corn. Two general patterns of enzymatic solubilization were observed: A, extensive erosion and fragmentation of corn and sorghum starches and their waxy counterparts, and B, selective granule-by-granule destruction of potato and most of the other starches. Even after 50% solubilization of corn and sorghum starches, the fragmented residues have iodine affinities, intrinsic viscosities, Brabender viscosities, and swelling and solubility patterns similar to those of the parent starches. It is suggested that these cereal starches may have a porous granule structure accessible to the enzyme, while potato starch is less permeable. Other evidence indicates that the starch granule has no restrictive outer membrane, that the starch fractions are distributed uniformly throughout the granule, and that amylase action is not related to considerations of molecular association or crystallization pattern.

A previous publication from this laboratory described the patterns of progressive swelling and solubilization of various granular starches when heated in water (7). The results were indicative of the extent and strength of molecular association (i.e., hydrogen bonding) within the granule. Recent work by Sandstedt (9) and by Badenhuizen (2) suggests that the dissolving action of amylases on granular starches may offer another means for studying granule structure. The extent and mode of enzyme digestion of various starches might provide evidence of physical differences. In addition, comparison of the undissolved residue with the original granular starch might indicate the nature of any skeletal structure.

Materials and Methods

Sources of Starches.

Corn, sorghum, and waxy sorghum ("white milo"), Corn Products Co.
Waxy maize ("Amioca" brand), American Maize-Products Co.
Wheat, Huron Milling Division, Hercules Powder Co.

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Tapioca ("Maris" brand) and rice, Stein Hall & Co.
Potato, Northern Aroostook Starch Co.
Arrowroot, prime St. Vincent, Morningstar-Paisley, Inc.
Unbleached sago flour (kindly supplied by National Starch and Chemical Corp.) was suspended in water, screened through fine bolting cloth, and the starch filtered and air-dried.
High-linear corn starch, phenotype aeae, laboratory-prepared, 11.9% iodine affinity.

Sources of Enzymes.

Bacterial alpha-amylase, analytical grade, SKB value at 30°C. = 16,000, Wallerstein Co., Inc.
"Rapidase" brand bacterial alpha-amylase, SKB value at 30°C. = 10,000, Wallerstein.
Glucamylase, laboratory-prepared fungal amylase from *Aspergillus niger* (12).
Pancreatin, Takamine Laboratory, division of Miles Laboratories, Inc.
Malt diastase, analytical grade, activity = 1,000°L, Wallerstein.
Fungal alpha-amylase, analytical grade, SKB value at 30°C. = 10,000, Wallerstein.
"Mylase" SC brand fungal alpha-amylase from *Aspergillus oryzae,* SKB value = 6,000, Wallerstein.
Beta-amylase, analytical grade, activity = 1,000° L, Wallerstein.

Method of Digestion. The specified amount of enzyme powder (expressed as percentage of dry starch) was added to 400 ml. of an aqueous slurry containing exactly 100 g. of starch (on dry basis). Two milliliters of toluene were added to prevent bacterial or mold action. The slurry was then adjusted to the optimum pH level for the particular enzyme (see Table I), placed in a thermostatted water bath at the desired temperature, and slowly stirred for 24 hours. At the end of this period the mixture was cooled, adjusted to pH 3.0 with hydrochloric acid, and stirred for 15 minutes to inactivate the enzyme. The slurry was then adjusted to pH 6.0 and filtered on a Buchner funnel; the cake was thoroughly washed with distilled water. After air-drying at room temperature, the weight and moisture content of this residue starch were determined, and the percentage of solubilized starch calculated by difference.

Analytical Methods. Intrinsic viscosities (in 1*N* potassium hydroxide) and iodine affinities were determined on the original and enzyme-treated starches by the methods of Lansky *et al.* (6). Brabender viscosities were determined by cooking for 1 hour at 95°C., then cooling to 50°C. and holding 1 hour (8). Swelling and solubility patterns were determined by the method of Leach *et al.* (7). Granule densities were determined in xylene at 30°C., using starches predried for 4 hours in the vacuum oven at 120°C. As an index of precision, average deviation of all density results was ± 0.0049.

Results

Comparative Activities of Various Enzymes. Preliminary tests were run to select the enzyme and digestion conditions which would give the desired solubilization of granular starch. Corn and potato starches were used as substrates, because they exhibit very wide differences in physical properties and granule organization. Table I shows the percentage of starch solubilized during 24 hours' digestion by each of the enzymes at the specified temperature and enzyme level. At 30°C., corn starch is solubilized to approximately the same extent (i.e., 18-22%) by 1% of bacterial alpha-amylase, pancreatin, glucamylase, or malt diastase. At 50°C. and 0.5% enzyme, the bacterial alpha-amylase is clearly the most active. The beta-amylase and fungal alpha-amylase showed low activity at 30°C. and were pre-

TABLE I

DIGESTION OF GRANULAR CORN AND POTATO STARCHES BY VARIOUS ENZYMES

ENZYME	pH	STARCH	ENZYME DOSAGE[a]	TEMPERATURE	STARCH SOLUBILIZED
			%	°C	%
Bacterial alpha-amylase	6.5	Corn	1.0	30	20.5
		Corn	0.5	50	49.5
		Potato	1.0	50	21.8
"Rapidase"	6.5	Corn	1.0	30	21.5
		Corn	0.5	50	48.6
Glucamylase	4.5	Corn	1.0	30	18.4
		Corn	0.5	50	36.4
		Potato	3.0	50	0.5
Pancreatin	7.0	Corn	1.0	30	20.7
		Corn	0.5	50	29.9
		Potato	1.0	50	12.5
Malt diastase	4.5	Corn	1.0	30	18.4
		Corn	0.5	50	6.0
		Potato	1.0	50	3.5
Fungal alpha-amylase	5.0	Corn	1.0	30	8.3
		Corn	0.5	50	4.9
		Potato	1.0	50	0.8
"Mylase" SC	5.0	Corn	1.0	30	8.4
		Corn	0.5	50	6.7
		Potato	1.0	50	1.5
Beta-amylase	4.5	Corn	1.0	30	8.3
		Corn	0.5	50	0.0

[a] Enzyme dosage calculated on dry starch basis.

sumably inactivated at 50°C.

In all cases (and particularly with glucamylase), granular potato starch is very much more resistant to enzyme action than is corn starch. Similar conclusions have been reached by Balls and Schwimmer (3), by Gates and Sandstedt (5), and by Badenhuizen (2). The greater digestibility of corn starch by enzyme appears to be an inherent property of this starch, and is not due to granule damage caused by drying the corn or by factory milling. As evidence, the following two starches showed substantially the same susceptibility toward bacterial alpha-amylase as commercial corn starch: 1) laboratory-prepared starch from dough-stage dent corn (35% moisture) used directly without drying, and 2) prime corn starch from the floury endosperm, obtained from the factory stream prior to buhr-milling. In contrast, Alsberg and Griffing (1) and more recently Blish, Sandstedt, and Kneen (4) have attributed differences in the enzyme susceptibilities of wheat flours in large part to granule damage during dry-milling.

The soluble fractions from the various 50°C. conversions of corn starch were qualitatively chromatographed to indicate the probable identity of the sugar products. Bacterial and pancreatic amylases gave principally d-glucose, with decreasing amounts of maltose, maltotriose, and maltotetraose. Glucamylase gave only d-glucose, and malt diastase gave principally maltose with a trace of the triose. The fungal alpha-amylases gave only d-glucose, probably indicating the presence of glucamylase.

Conditions for Digestion with Bacterial Alpha-Amylase. Since the analytical grade of bacterial alpha-amylase had the highest solubilizing action on both corn and potato starches, further studies were made to establish the optimum temperature, digestion time, and enzyme level. The solubilization of both corn and potato starches increased as the temperature was raised from 30° to 50°C.:

	Starch Solubilized	
Digestion temperature, °C.	*Corn starch +0.5% enzyme*	*Potato starch +3% enzyme*
	%	%
30	17.2	4.7
40	28.1	12.6
50	49.5	29.7

At 50°C. neither starch showed any evidence of granule swelling, or any enlargement or loss of birefringence at the hilum. As shown in Fig. 1, both starches were solubilized at a rapid rate during the

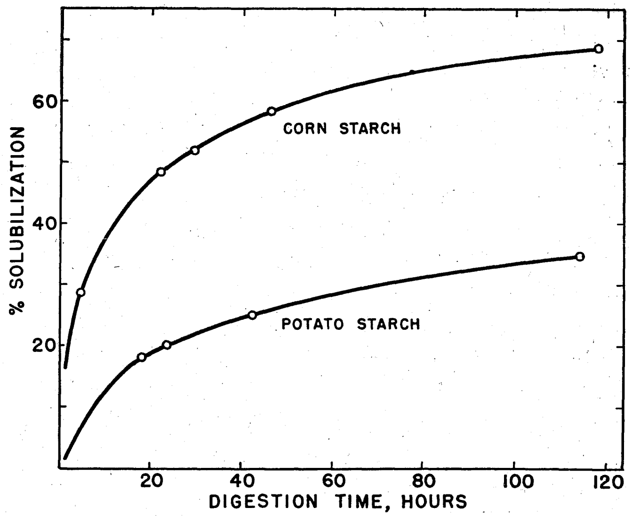

Fig. 1. Rate of solubilization of granular corn and potato starches by 0.5% bacterial alpha-amylase at 50°C.

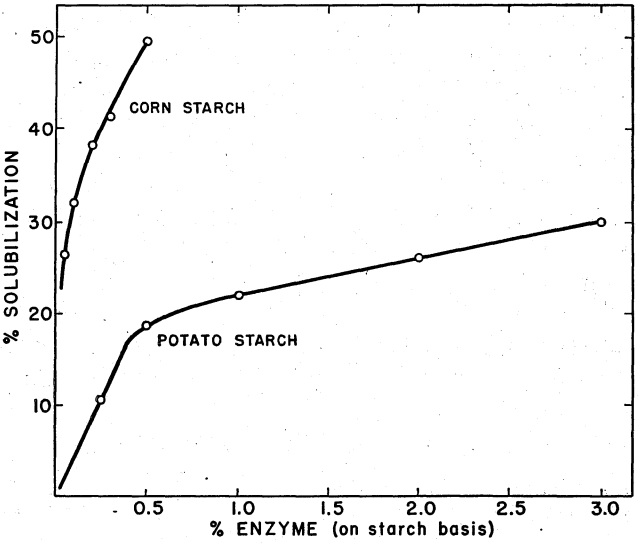

Fig. 2. Effect of dosage of bacterial alpha-amylase on the solubilization of granular corn and potato starches (24 hours' digestion at 50°C.).

initial 24 hours, and more slowly thereafter. The decreased rate of digestion after 24 hours may be due partly to enzyme inactivation and partly to inhibition by the sugar products. For example, 0.2% bacterial alpha-amylase at 50°C. solubilized 100 g. of corn starch to the extent of 38.1% in 24 hours; a similar run in the presence of 30 g. of added d-glucose decreased the solubilization to 27.1%. In Fig. 2, 0.5% enzyme appeared to be an efficient level for solubilization of potato starch. Hence subsequent studies on various granular starches were conducted with 0.5% bacterial alpha-amylase (analytical grade) for 24 hours at 50°C.

Comparative Susceptibility of Various Starches. Eleven different unmodified starches were digested with bacterial alpha-amylase under the above specified conditions (i.e., 24 hours, 50°C., 0.5% enzyme). Also included was heat-moisture-treated potato starch, prepared by refluxing unmodified potato starch in boiling 70% diacetone alcohol for 4 hours (7). Table II lists the various starches in order of increasing resistance to alpha-amylase action. Waxy maize is the most susceptible (57.8% solubilized in 24 hours), and heat-moisture-treated potato starch and high-amylose corn starch are least attacked (15.5 and 14.0%, respectively). In no case was there any preferential attack on either the linear or the branched starch fraction, since the iodine affinity of each residue starch was substantially the same as that of the parent starch. In most instances, there was some decrease in both intrinsic viscosity and Brabender viscosity; however, this effect is considered relatively minor since all the residue starches are still essentially "thick-boiling" in character.

Neither corn nor potato starch appears to adsorb any bacterial alpha-amylase from aqueous solution, and hence their different susceptibilities cannot be ascribed to different adsorptive affinities for the enzyme. For example, 40 g. of corn starch were stirred for 30 minutes in 200 ml. of bacterial alpha-amylase solution at room temperature and pH 6.5. At enzyme concentrations in the range of 0.5 to 4.0 mg. per ml., the alpha-amylase activity of the aqueous substrate was not changed by contact with the starch, as determined spectrophotometrically by a modified Wohlgemuth assay. In a similar series of experiments with potato starch, the activity of the substrate was markedly increased by contact with the starch. This is due to the fact that granular potato starch preferentially adsorbs water from the surrounding enzyme solution, thus increasing the concentration or apparent activity of the latter.

Successive Enzyme Digestions. It was of interest to compare the residues from corn and potato starches at equally high levels of digestion. Corn starch was given three successive 24-hour digestions with 0.1% bacterial alpha-amylase at 50°C.; after each digestion the residue starch was filtered and dried to determine yield, then resuspended and treated with fresh enzyme. To compensate for its lower

TABLE II

SOLUBILIZATION OF VARIOUS GRANULAR STARCHES WITH BACTERIAL ALPHA-AMYLASE, AND PROPERTIES OF THE RESIDUE STARCHES

(0.5% enzyme, 24 hours' digestion, 50°C.)

STARCH	STARCH SOLUBILIZED	SAMPLE	INTRINSIC VISCOSITY	IODINE AFFINITY	DENSITY	BRABENDER VISCOSITY[a]			
						Concentration	At Peak	At 95°C.	At 50°C.
	%			%		g/500 ml	B u	B u	B u
Waxy maize	57.8	Untreated	1.58	....	1.480	30	760	160	220
		Residue	1.36	....	1.516	30	305	65	120
Tapioca	55.7	Untreated	2.03	3.15	1.521	30	590	150	290
		Residue	1.86	3.20	1.528	30	370	120	225
Waxy sorghum	55.7	Untreated	1.43	....	1.490	30	545	120	180
		Residue	1.33	....	1.522	30	205	45	85
Sorghum	50.2	Untreated	1.43	4.90	1.500	40	665	490	710
		Residue	1.31	4.97	1.504	40	375	230	440
Corn	49.5	Untreated	1.61	4.83	1.517	35	485	280	815
		Residue	1.51	4.72	1.511	35	370	180	545
Wheat	48.5	Untreated	1.25	5.30	1.542	55	530	145	>1000
		Residue	1.14	5.30	1.523	55	715	310	>1000
Rice	32.6	Untreated	1.42	4.70	1.510	40	670	735	>1000
		Residue	1.40	5.07	1.489	40	690	700	>1000
Sago	27.0	Untreated	1.60	4.98	1.494	35	590	190	420
		Residue	1.60	5.07	1.533	35	580	175	480
Arrowroot	20.8	Untreated	1.85	4.45	1.510	35	885	470	780
		Residue	1.85	4.65	1.526	35	850	530	960
Potato	18.4	Untreated	2.44	4.42	1.511	30	>1000	240	470
		Residue	1.53	4.65	1.541	30	>1000	180	310
Heat-moisture-treated potato	15.5	Untreated	1.86	4.45	1.507	35	>1000	750	930
		Residue	0.86	4.40	1.526	35	>1000	570	835
High-linear corn	14.0	Untreated	1.09	11.9					
		Residue	....	11.3					

[a] Brabender viscosities are given at the pasting peak, after holding 1 hour at 95°C., and after cooling and then holding 1 hour at 50°C.

TABLE III

PROPERTIES OF THE RESIDUES FROM CORN AND POTATO STARCHES AFTER EXTENSIVE SOLUBILIZATION WITH BACTERIAL ALPHA-AMYLASE

STARCH	SOLUBLES	INTRINSIC VISCOSITY	IODINE AFFINITY	BRABENDER VISCOSITIES[a]		
				AT PEAK	AT 95°C.	AT 50°C.
	%		%	*B u*	*B u*	*B u*
Corn						
Original	....	1.61	4.83	500	330	830
Enzyme-digested	58.0	1.47	4.55	375	250	530
Potato						
Original	....	2.44	4.42	>1000	240	470
Enzyme-digested	56.6	1.70	4.85	>1000	260	405

[a] Brabender viscosities were run at 35 g. per 500 ml. on the corn starch samples, and at 30 g. on the potato starch samples.

susceptibility, potato starch was given four successive digestions, and the enzyme level was increased to 1.0%. Results are shown in Table III. While Brabender viscosities and intrinsic viscosities are somewhat lower, the residues from both starches are still considered as essentially thick-boiling, even after conversion of 56–58% of the original starch substance.

Microscopic Examination. As observed under the polarizing microscope, the various starches show two distinctly different patterns of enzyme attack: A, extensive granule erosion and fragmentation with corn and sorghum starches and their waxy counterparts, and B, selective-granule digestion with potato and the other starches studied. (Wheat starch is intermediate in behavior.) In the typical case of corn starch, the enzyme appears to attack the hilum first, with the loss of birefringence in that region. Schwimmer (11) has likewise observed this type of enzyme action on granular corn and wheat starches. The hilum appears to be the most susceptible part of the granule, since it is attacked in the early stages of enzyme digestion, and since it is likewise the first region to show loss of birefringence when the granule swells in hot water. As enzyme digestion proceeds most of the corn starch granules become highly eroded and fragmented (Fig. 3, A). However, there are always a few granules (i.e., 5–10% of the original starch) which show no evidence of any erosion, even after 50% solubilization. These resistant granules are of both the angular and the round types (i.e., from the horny and floury endosperm, respectively).

In contrast, potato starch shows no evidence of granule erosion or decrease of birefringence even after 56% solubilization (Fig. 3, B). During enzyme digestion, the microscope will show an occasional

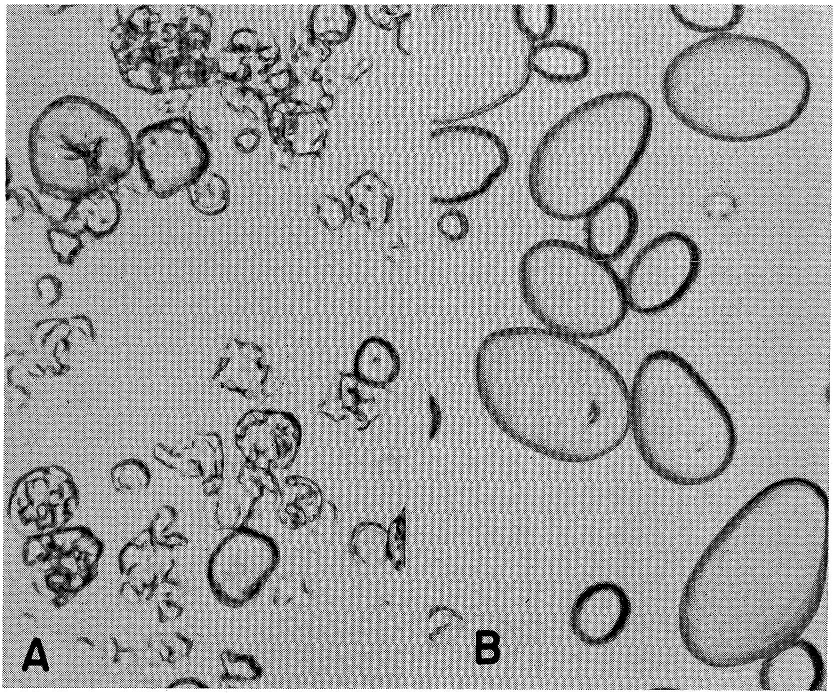

Fig. 3. Microscopic appearance of granular corn (A) and potato (B) starches after approximately 50% solubilization by bacterial alpha-amylase.

"ghost granule" undergoing rapid dissolution. Hence the enzyme attacks and completely destroys a few granules at a time, though the reason for this selectivity is not apparent. The enzyme shows no preference for either the large or the small starch granules. To prove this point, the parent potato starch and an enzyme-treated residue (namely, 56.6% solubilized by four successive digestions with 1% bacterial alpha-amylase) were each photomicrographed, and the granule diameters (i.e., average of long and short dimensions) were manually measured on approximately 400 granules of each sample. Size distribution curves were then obtained by plotting granule diameters against percentage frequency. The curves for the parent starch and the enzyme-digested residue were virtually identical, and the "number-average" and "weight-average" granule diameters (10) did not differ substantially:

	Number-Average Granule Diameter	*Weight-Average Granule Diameter*
	μ	μ
Untreated potato starch	20.9	37.1
Enzyme-digested residue	18.4	34.3

TABLE IV

SWELLING AND SOLUBILITY PATTERNS OF TREATED POTATO STARCHES

PASTING TEMPERATURE	SWELLING POWER[a]			SOLUBILITY		
	A Unmodified	B Enzyme Residue	C Water-Digested	A Unmodified	B Enzyme Residue	C Water-Digested
°C.	*times*	*times*	*times*	%	%	%
60	42.9	2.4	2.0	12.2	0.5	0.3
65	91.2	9.8	23.6	19.8	3.2	10.2
70	141.	26.3	36.1	28.0	6.8	11.9
75	200.	31.1	44.5	29.0	9.0	11.9
80	261.	41.8	61.5	34.3	10.0	15.1

[a] Corrected for solubility.

Effect of Exhaustive Drying on Enzyme Susceptibility. The enzyme susceptibility of potato starch is substantially increased by exhaustive drying. For example, the original potato starch gave 29.7% solubles when digested for 24 hours at 50°C. with 3% bacterial alpha-amylase. When the potato starch was dried for 16 hours *in vacuo* at 120°C., a similar enzyme treatment gave 49.7% solubles. This might perhaps be attributed to dextrinization or heat-damage during drying. However, when untreated potato starch was slowly dried to a moisture level of 0.27% by 10 weeks *in vacuo* over phosphorus pentoxide at room temperature, a similar enzyme treatment gave 54.5% solubles. Exhaustively dried starches develop considerable heat when contacted with water, which might conceivably cause damage or cracks within the granule. To avoid this, both of the above dried starches were exposed to laboratory air for several days to regain their equilibrium moisture contents, before treating with enzyme. No hydrolytic modification would be expected during drying, since the slurry pH of the original potato starch and the two dried samples was in the range of 6.7 to 6.8.

Heat-Moisture Treatment. Some concern was felt lest the enzyme resistance of potato starch might be due to the substantial heat-moisture modification incurred during prolonged digestion at 50°C. While four 24-hour digestions of potato starch at 50° C. cause only a minor change in the Brabender viscosity (Table III), nevertheless this amount of heat-moisture treatment does decrease the swelling and solubility of the starch. For example, swelling and solubility patterns were determined on the following three starches: A, unmodified potato starch; B, the residue after 56.6% solubilization of potato starch by four successive treatments with 1% bacterial alpha-amylase at 50°C.; C, potato starch after four similar digestions in water at 50°C. without enzyme. The two latter samples show considerable reduction in both swelling power and solubility (Table IV). However, from a practical standpoint, it was necessary to use a digestion temperature of 50°C. in order to solubilize potato starch to a significant extent, since even 20% of bacterial alpha-amylase at 30°C. solubilized only 9.2% of the starch in 24 hours. While heat-moisture modification undoubtedly does occur at 50°C., this cannot possibly account for the enzyme resistance of potato starch, for two reasons. First, as previously mentioned, corn and potato starches still show major differences in solubilization when treated for 24 hours with bacterial alpha-amylase at 30°C. and at 40°C. Second, even though tapioca starch is very prone to undergo heat-moisture modification, it is still highly susceptible to enzyme attack.

Discussion and Conclusions

Certain general conclusions seem to be valid:

1. Enzyme susceptibility is not related to external surface area of the starch granules, in agreement with conclusions by Schwimmer (11). Thus the small-granule rice starch is in an intermediate position in Table II, and the other starches show no correlation between granule size and extent of solubilization.

2. The X-ray diffraction pattern of the starch has no relation to enzyme susceptibility. The starches tested are reputed to have the following spectra: wheat, rice, corn, sorghum, waxy maize, waxy sorghum, and heat-moisture-treated potato give an A-pattern; potato and high-linear corn starches give a B-pattern; arrowroot, tapioca, and sago give a C-pattern (2).

3. The concept of a peripheral membrane or hull enclosing the granule is untenable for corn and sorghum starches and their waxy counterparts. Even after extensive erosion and fragmentation, the residues still give Brabender viscosity curves typical of thick-boiling starches. As additional evidence for the absence of any membrane, swelling and solubility data were determined on the residue after 50% solubilization of corn starch by bacterial alpha-amylase. Despite the fissured and fragmented nature of the granules, this product showed even lower swelling and solubilization than the parent starch (Table V). However, the possibility of an enclosing membrane is not excluded with starches such as potato which are digested by selective-granule attack.

4. At least in the cases of corn and sorghum starches, the linear and branched fractions must be distributed uniformly throughout the granule. Even after half the total starch substance was solubilized,

TABLE V

EFFECT OF ENZYMATIC DIGESTION ON THE SWELLING POWER AND SOLUBILITY OF CORN STARCH

(Comparison of untreated corn starch and residue after 50% solubilization by 0.5% bacterial alpha-amylase for 24 hours at 50°C.)

PASTING TEMPERATURE	SWELLING POWER[a]		SOLUBILITY	
	Untreated	Residue	Untreated	Residue
°C	*times*	*times*	%	%
65	4.4	4.0	2.6	1.1
70	7.8	7.1	6.8	3.7
75	9.5	8.7	8.7	5.4
80	10.5	9.0	9.9	6.5
85	11.8	9.9	10.2	8.5
90	16.4	12.8	16.7	12.6
95	23.6	25.8	24.6	23.5

[a]Corrected for solubles.

the eroded and fragmented residues have substantially the same iodine affinities as the parent starches. This same evidence shows that the enzyme has no preference for either fraction. Since tapioca, arrowroot, sago, and potato starches are solubilized by attack on individual granules, no similar conclusion can be made regarding distribution of their fractions.

5. The waxy starches are somewhat more susceptible to enzyme attack than the corresponding normal starches. Also, high-linear corn starch is least affected by enzyme. Similarly, Schwimmer (11) found that normal pea starch is attacked more rapidly than the high-linear starch from wrinkled peas. Hence the presence of linear fraction probably retards solubilization by enzyme.

6. In general, intrinsic viscosities of the residues after enzyme attack are only slightly lower than the values for the parent starches. This is in contrast with the action of mineral acids on granular starches, which causes extensive hydrolytic breakdown throughout the granule. Hence it is presumed that the enzyme does not penetrate freely into the molecular lattice of the granule, but is limited to certain accessible surfaces or regions.

7. The absolute densities of the residues are very little different than the densities of the parent starches (Table II). This suggests that the enzyme does not preferentially digest the more amorphous or disorganized regions of the granule, since such action should cause a significant increase in density.

8. Enzyme susceptibility has no relationship to the patterns of swelling and solubilization of the starches when heated in water. Thus, weakly-bonded potato and tapioca starches swell freely to give stringy cohesive pastes, yet they show opposite susceptibilities toward

enzyme. Strongly bonded corn and heat-moisture-treated potato starches both show restricted swelling and give "short" heavy-bodied pastes, yet they differ widely in enzyme susceptibility.

Hence it is concluded that the enzyme susceptibility of granular starches is not influenced by physicochemical considerations of micellar structure, internal molecular association, or type of crystallinity. As Badenhuizen (2) has plausibly suggested, the more susceptible granules may possess pores or a coarse spongelike structure, with openings of sufficient size to admit the enzyme molecules. These pores must be characteristic of the particular species of starch, and not merely fissures produced during drying or manufacture. However, extreme drying (even without heat) increases the enzyme susceptibility of potato starch, perhaps by formation of a spongy internal structure.

Acknowledgment

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Literature Cited

1. Alsberg, C. L., and Griffing, E. P. Effect of fine grinding on flour. Cereal Chem. **2:** 325–344 (1925).
2. Badenhuizen, N. P. Chemistry and biology of the starch granule. Protoplasmatologia. Edited by L. V. Heilbrunn and F. Weber. Band II, B, 2, b. Springer-Verlag: Vienna, Austria (1959).
3. Balls, A. K., and Schwimmer, S. Digestion of raw starch. J. Biol. Chem. **156:** 203–210 (1944).
4. Blish, M. J., Sandstedt, R. M., and Kneen, E. The cereal amylases with reference to flour and malt behavior. Cereal Chem. **15:** 629–657 (1938).
5. Gates, R. L., and Sandstedt, R. M. A method of determining enzymatic digestion of raw starch. Cereal Chem. **30:** 413–419 (1953).
6. Lansky, S., Kooi, M., and Schoch, T. J. Properties of the fractions and linear subfractions from various starches. J. Am. Chem. Soc. **71:** 4066–4075 (1949).
7. Leach, H. W., McCowen, L. D., and Schoch, T. J. Structure of the starch granule. I. Swelling and solubility patterns of various starches. Cereal Chem. **36:** 534–544 (1959).
8. Mazurs, E. G., Schoch, T. J., and Kite, F. E. Graphical analysis of the Brabender viscosity curves of various starches. Cereal Chem. **34:** 141–152 (1957).
9. Sandstedt, R. M. Photomicrographic studies of wheat starch III. Enzymatic digestion and granule structure. Supplement to Cereal Chem. **32:** 17–47 (May 1955).
10. Schoch, T. J., and Maywald, E. C. Microscopic examination of modified starches. Anal. Chem. **28:** 382–387 (1956).
11. Schwimmer, S. The role of maltase in the enzymolysis of raw starch. J. Biol. Chem. **161:** 219–234 (1945).
12. Tsuchiya, H. M., Corman, J., and Koepsell, H. J. Production of mold amylases in submerged culture. II. Factors affecting the production of alpha-amylase and maltase by certain Aspergilli. Cereal Chem. **27:** 322–330 (1950).