

Paste Properties of Modified Starches from Partial Waxy Wheats¹

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ABSTRACT

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Properties of modified starches from partial waxy wheats have not been examined. Protease digestion of cracked kernels of three hard winter wheats varying in amylose content led to 82–85% recovery of starch, whereas kneading of the flour-water doughs gave 75–83% recovery. All starches had a protein content of <0.3% and ash content of <0.01%. Granule size distributions showed that starch from Ike kernels contained 86% A-type granules with a peak size of $\approx 18\mu\text{m}$, and Karl-92 starch contained 77% A-type granules with a peak size of $\approx 16\mu\text{m}$. The A-type granules (82%) from Rio Blanco starch were intermediate in size. The amylose content of Karl-92 starch, determined by concanavalin-A precipitation of amylopectin, was 28%, which was 17% higher than that of Ike starch (23%). The amylose content of Rio Blanco starch was 26%. The lipid content of Karl-92 starch, determined as fatty acid methyl esters, also was 18% higher than that of Ike starch (601 vs. 488 mg/100 g of starch, respec-

tively). Wheat starches were modified with hydroxypropyl (HP) groups to low (1.5–2.5%) and medium ($\approx 4.0\%$) levels, and the HP starches were cross-linked with phosphoryl chloride at levels of 0.003–0.075%. Pasting curves (amylograms) showed that Ike starch substituted with a low level of HP and optimally cross-linked with 0.025% phosphoryl chloride (starch basis) had a greater paste consistency than low HP cross-linked Karl-92, and Rio Blanco starches. At 4% HP and optimum cross-linking (0.003% phosphoryl chloride), the paste consistencies of the modified starches were nearly the same. The clarity of unmodified Ike starch paste was higher than that of Karl-92 or Rio Blanco starch pastes, and the clarity of all three pastes decreased as cross-linking was increased. Unmodified Ike starch formed a stronger gel than unmodified Karl-92 and Rio Blanco starches, but gel properties largely converged as the starches were modified.

The swelling of wheat starch granules and any leaching of amylose during cooking are important in establishing the eating texture of wheat foods. Wheat starch swells and solubilizes to different extents in various foods. Upon cooling, packaging, and storing of food products like crackers, cookies, breads, cakes, and soups, hydrated starch molecules interact with themselves and other components to generate a variety of textures including friability, chewiness, softness, and consistency (Hoseney et al 1977). Cereal scientists have attributed the variable swelling of wheat starch in foods to differences in ingredients and their concentrations including water, monoacyl lipids, sugars, and salts. Now, however, we are beginning to understand that inherent differences exist in the swelling of wheat starches and these differences are governed by genetic mutations (Nakamura et al 1993).

In the early to mid-60's, amylose synthesis in corn and rice starches was associated with granule-bound starch synthase (GBSS) also known as waxy protein (Nelson and Rines 1962, Murata et al 1965, Shannon and Garwood 1984). Wheat breeders in Japan examined 31 Japanese wheat starches for levels of GBSS and amylose (Nakamura et al 1993). They discovered that starches from 86% of the wheats had 27–30% amylose, starches from 7% of the wheats contained $\approx 22\%$ amylose, and starches from the remaining 7% of the wheats had intermediate levels. In wild-type hexaploid wheats, three waxy protein bands were resolved by SDS-PAGE: Wx-A1, Wx-B1, and Wx-D1 proteins. The Japanese cultivars Kanto 107 and Kanto 79 were null for two of the three waxy proteins; they contained only Wx-D1 protein and were called partial waxy mutant wheats. The hard red winter wheat Ike also was found null for Wx-A1 and Wx-B1 proteins, whereas Karl 92 showed all three waxy proteins, and the hard white winter wheat Rio Blanco lacked Wx-B1 protein (Demeke et al 1997, Graybosch et al 1998). Two of 63 Canadian wheats examined were null for Wx-B1 protein (Chibbar et al 1997).

Wheat starch is among commonly available starches of commerce, along with corn, potato, cassava, and waxy corn. It is isolated by wet-milling of flour by several processes (Fellers 1973, Seib 1994, Witt 1997, Cornell and Hoveling 1998). Its production has grown, particularly in Europe, during the past 10 years. Wheat starch has a

bimodal granule-size distribution, but most commercial wheat starch has predominantly large granules because of the low sedimentation rate of small granules. However, small-granule wheat starch is available in Germany and Holland.

Commercially modified wheat starches include hydroxypropylated, hydroxypropylated and cross-linked, acetylated and cross-linked, and octenylsuccinylated starches. Cross-linking of starch reduces the swelling and solubility of the granules and increases the rigidity, whereas hydroxypropylation increases swelling but inhibits gelling and improves cold-temperature storage stability. A combination of cross-linking and hydroxypropylating produces starches that generate a high paste consistency with stability to shear, acidic pH, and low temperature. Such starches are used widely in salad dressings, canned foods, frozen foods, and puddings. Hydroxypropylated starches that are not cross-linked are not used as thickeners because the high degree of swelling gives an undesirable stringy paste. Hydroxypropylating a starch with propylene oxide and cross-linking with phosphoryl chloride both are done in an aqueous slurry at 35–45% starch solids and alkaline pH in the presence of a salt, most often sodium sulfate (Rutenberg and Solarek 1984, Tuschoff 1986). Because the swelling power of partial waxy wheat starches is elevated (Wang and Seib 1996, Morris et al 1997), they might have more thickening power than the wild-type wheat starches when they are doubly modified by hydroxypropylation and cross-linking.

The objectives of this investigation were: 1) to isolate starches from three hard winter wheats with different amylose levels using protease digestion of kernels and dough-washing of flours; 2) to determine amylose levels, lipid levels, and granule size distributions of the starches; and 3) to hydroxypropylate the starches with 8% propylene oxide in the presence of sodium sulfate (≈ 2 or 10%) and then determine the level of cross-linking with phosphoryl chloride that produced doubly modified wheat starches giving a high paste consistency with low stringiness.

MATERIALS AND METHODS

Materials

Two hard red winter wheats, Ike and Karl-92, were obtained from T. J. Martin and Vernon Schaffer, Kansas State University, Manhattan, KS. Hard white winter wheat, Rio Blanco, was obtained from the American White Wheat Producers Association (Atchison, KS). Straight-grade flour was milled from tempered wheat ($\approx 16\%$ moisture) using a Buhler pilot mill. Protease (type XIV), propylene

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oxide, and thiomersal were from Sigma Chemical Co. (St. Louis, MO). A standard mixture of fatty acid methyl esters (FAME) was from Sigma, as was the heptadecanoic acid methyl ester (>99% purity) used as internal standard. All other chemicals were reagent-grade unless otherwise specified.

General Methods

All analyses were done in duplicate unless otherwise stated. Moisture was assayed by oven-drying a sample for 1 hr at 130°C (AACC 1995). Protein was assayed by the Dumas combustion method (LECO Corp., St. Joseph, MI). Total starch and amylose contents were determined by enzyme digestion and concanavalin A methods, respectively (Megazyme Pty, Sydney, Australia). Falling number was determined by Approved Method 56-81B (AACC 1995). Lipid in starch was determined as FAME by the method of Morrison et al (1975), where lipid was estimated by the formula: Starch lipid = total FAME \times 1.7.

Gas-liquid chromatography was done on a Varian gas chromatograph (model 3600, Walnut Creek, CA) equipped with a flame-ionization detector and a capillary column SP-2340 (60 m \times 0.25 mm) with a stationary phase of cyanosilicone at a film thickness of 0.20 μ m (Supelco, Bellefonte, PA). The column temperature was programmed from 60 to 220°C at a rate of 15°C/min, injection temperature was 220°C, and detection temperature was 240°C. The flow rate was \approx 2.4 mL/min, and injection volume was 1 μ L. Response factors of the standard methyl esters to the internal standard (C17:0) were determined. In unknown samples, responses of the methyl esters were summed in a chromatogram to calculate the total fatty ester level. Corrections for losses of unsaturated fatty acids were determined by adding known concentrations of standard methyl esters to potato starch and then determining their recovery in the assay procedure. Losses were not observed for saturated FAME, but amounted to 15% for methyl oleate and linoleate and 30% for methyl linolenate.

Swelling power of starch was determined at 92°C by a slight modification of the method of Crosbie (1991). Starch concentration was 1.67%, and the hot paste was centrifuged soon after heating, instead of cooling the paste and then centrifuging. Granule size distributions were determined on a Coulter Counter with 16 channels

(model PCA I, Coulter Corp., Hialeah, FL). Starch samples (100 mg) were soaked for 16 hr at 25°C in distilled water (10 mL) before particle-size analysis. Hydrogen ion concentration was measured with a high pH electrode (55510-02 JJ6, Cole-Palmer) and a pH meter. Standard pH 7 and 10 buffers (SB116-500, Fisher Scientific) were used to calibrate the electrode.

Hydroxypropyl content was determined by high resolution $^1\text{H-NMR}$ spectroscopy of the starch's α -limit dextrin (Xu and Seib 1997). The integrated intensities were measured for the HP methyl signals (I_{HP}) and for the total methine and methylene signals on oxygenated carbons (I_{HCO}), except for the anomeric proton. The level of hydroxypropylation was calculated by this equation, which assumes the absence of polyhydroxypropylated chains on starch: HP (wt %) = $(59)(100)/[81(I_{\text{HCO}}/I_{\text{HP}}) - 23]$.

All data were analyzed using statistical software (SAS Institute, Cary, NC). Means were compared using Fisher's protected least significant differences method.

Differential Scanning Calorimetry

All samples were prepared by sealing starch in aluminum pans (2.5 mg, db) with water at a starch-to-water ratio of 1:3 (w/w). Samples were heated at 10°C/min to 130°C at a sensitivity of 0.5 mcal/sec in a Perkin-Elmer DSC Pyris-1 instrument equipped with an FTS Systems flexicooler and temperature controller. Indium was used to calibrate the instrument. Onset (T_o) and completion (T_c) temperatures of the gelatinization endotherm were determined by intersection of the extrapolated baseline and peak edges.

Starch Isolation

Starch was isolated by a modification (X. Liang, *personal communication*) of the protease method of Morrison et al (1984). Wheat grains were cracked by roller-milling through a 0.9-mm gap, and the cracked wheat (240 g) was added to 0.02M HCl (2.4 L) at 4°C. After 8–10 min, sodium metabisulphite (0.5%, based on wheat, 1,200 mg) and thiomersal (0.01%, 24 mg) were added to the slurry, and the pH was buffered to 7.6 (0.09M) by the addition of tris(hydroxymethyl)aminomethane (29 g) followed by adjustment with 1M HCl. Protease (type XIV, Sigma, 0.5%, based on wheat, 1,200 mg) was dissolved in 0.02M HCl (100 mL), and the solution

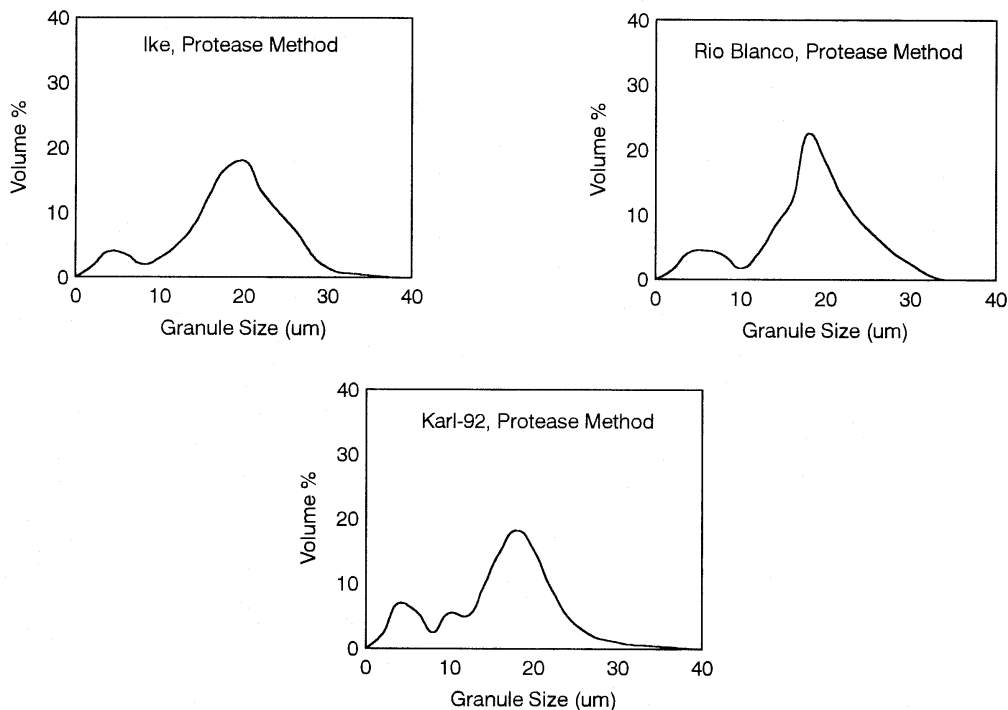


Fig. 1. Size distributions of starch granules isolated from Ike, Rio Blanco, and Karl-92 wheats by protease digestion (Group I).

was held for 3–5 min at 25°C to denature α -amylase and then added to the slurry of cracked grain. The mixture was digested at 4°C for 30 hr with continuous stirring. The digest was placed atop a 75- μ m wire mesh sieve, and the softened grain was rubbed on the sieve and washed with water (2×200 mL). The throughs were collected and centrifuged at $2,500 \times g$ for 10 min. The overs were placed in a Waring blender with water (200 mL) and ground at low speed for 30 sec. The resulting slurry was placed atop the 75- μ m mesh sieve, and the throughs and overs were collected. The grinding procedure was repeated twice more and the overs were discarded. All throughs were combined (≈ 600 mL) and centrifuged at $2,500 \times g$ for 10 min. The supernatant was discarded, and the sedimented starch was washed with water (3×200 mL) and centrifuged, and the dark tailings layer was removed with a spatula. The tailings fractions were combined, slurried in water, and centrifuged to recover additional starch, which was combined with the other starch. The combined starch then was washed with 1% sodium chloride solution (200 mL) and water (3×100 mL) and air-dried for 48 hr. The isolated starches from the three wheats contained $\approx 12\%$ moisture and $<0.3\%$ protein, and the average yield was 130 g (54%).

Starch was also isolated as described by Shogren et al (1969), wherein a wheat flour-water slurry was prepared at 4°C. The slurry was centrifuged, and the sediment was worked into a dough mass that was washed repeatedly with water to remove the starch-rich aqueous phase. The starch was purified as described above. Starches isolated by protease digestion of wheat kernels and dough washing were assigned to Group I and Group II, respectively.

Hydroxypropylating and Cross-Linking of Wheat Starch

Hydroxypropylating and cross-linking of wheat starch were done by the procedure of Tuschoff et al (1969). Group I (isolated from wheat kernels) starches were modified in the presence of 2% sodium sulfate. Wheat starch (79.5 g, db) was slurried in water (122.0 mL), and a mixture (11–12 mL) of 1.5M sodium hydroxide and 0.75M sodium sulfate (total of $\approx 2\%$, starch basis) was added to bring to pH 11.5 at 25°C. The alkaline slurry was placed in a three-necked, round-bottom flask (500 mL) equipped with a ground-glass stirring rod and adapter. Propylene oxide (7.5 mL, 6.4 g) was added,

the side arm was sealed with a stopper, and the reaction mixture placed in a constant temperature bath at 45°C with continuous agitation. Glycerol was used to lubricate the stir rod fitting, which helped to retain volatiles in the flask. After 24 hr, the reaction mixture was cooled to room temperature, and phosphoryl chloride (0.025 or 0.075% based on starch, equal to 12 or 36 μ L) was added with stirring at 25°C. After 60 min, the slurry was adjusted to pH 5.5 with 1M hydrochloric acid, and the hydroxypropylated and cross-linked starch was isolated by centrifuging ($15,000 \times g$, 10 min), washing with water (4×100 mL), and air-drying overnight. Pieces of dried starch gels, which indicated that some starch granules were gelatinized during modification but which amounted to $<5\%$, were removed by hand. The remaining dried material (≈ 77.5 g) was ground gently to a fine powder with a mortar and pestle. The starch then was ground for 10 sec in a high-speed impact mill (Tekmar, IKA-LaborTechnik, Germany).

Modification of Group II starches was done with a higher level ($\approx 8\%$ starch basis, 5.96 g) of sodium sulfate in the starch slurry before raising to pH 11.5 with the mixture of 1.5M sodium hydroxide and 0.75M sodium sulfate, which gave a total level of sodium sulfate of $\approx 10\%$. Hydroxypropylated (8% propylene oxide) starches in this group were cross-linked at five different levels of phosphoryl chloride (0.003, 0.006, 0.013, 0.025, and 0.075%).

Starch Paste Properties

The pasting of a starch sample was examined in a Brabender Viscograph-E (C. W. Brabender Instruments, Hackensack, NJ) equipped with a 700 cm-g torque sensor. Starch (33.75 or 28.2 g, db) was slurried in water (416.25 or 421.8 mL), which equals a starch concentration of 7.5 or 6.25% (w/w). The slurry was heated from 25 to 95°C at the rate of 1.5°C/min, and the paste was held at 95°C for 30 min, then cooled to 50°C at 1.5°C/min, and held at 50°C for 30 min. Consistency was recorded in BU by a chart recorder.

Stringiness of a starch paste was determined subjectively by comparing an unknown to a 5% potato starch paste and a 7.5% wheat starch paste. Stringiness was rated on a scale of 1–5, where 5 was the highest stringiness (equal to that of the potato starch paste), and 1 was the lowest stringiness (equal to that of the wheat starch paste).

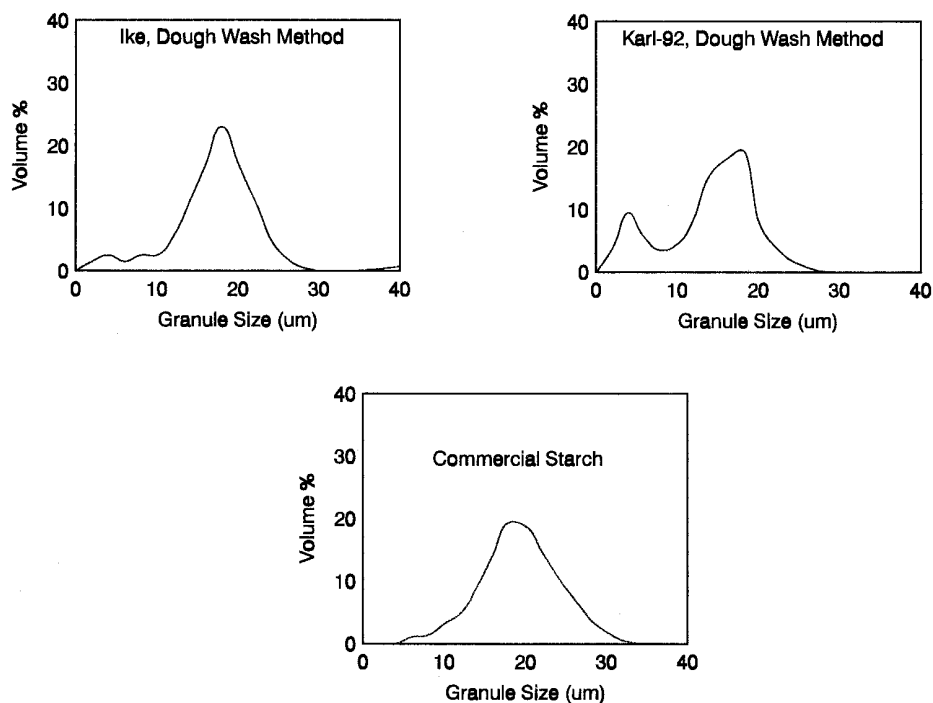


Fig. 2. Size distributions of starch granules isolated from straight-grade flours from Ike and Karl-92 by dough washing (Group II) compared to commercial starch.

To determine paste clarity, starch (0.05 g, db) was pasted in water (5.0 mL) in a glass-stoppered tube at 95°C for 30 min with shaking every 5 min. After cooling, clarity was measured on a spectrophotometer (Spectronic 601) at 650 nm against water as a blank (Craig et al 1989). The highly cross-linked starches with low swelling power tended to sediment during measurement of absorbance, so readings had to be taken quickly after agitation. Freeze-thaw stability of starch pastes was tested according to Wu and Seib (1990). Pastes were prepared in the amylograph and transferred to ≈20 polypropylene centrifuge tubes (20 g each), which were tightly capped and stored at 4°C for 24 hr. The tubes then were put in the freezer at -20°C and subjected to ≈7–8 freeze-thaw cycles. Each cycle consisted of 22 hr in the freezer and 2 hr at 30°C in a waterbath. After

each cycle, duplicate tubes were centrifuged for 15 min at 1,500 × g, and the amount of liquid separated was determined by weight.

Dynamic Rheological Properties of Starch Pastes and Gels

The viscoelastic properties of starch pastes and gels were determined by the method of X. Liang (*personal communication*). Starch (7.5%, 0.375 g, db) was weighed into a 50-mL polycarbonate centrifuge tube, and water was added to bring the weight of the contents to 5 g. The slurry was stirred and placed in a water bath maintained at 70°C. The contents were stirred manually for 7 min, then ≈3 g of slurry was transferred into the concentric cylinder cell (C14), (gap 3 mm) of a rheometer (VOR, Bohlin Rheologi, Edison, NJ), which had been preheated in the instrument to 70°C. A thin layer of mineral oil was used to cover the exposed surface of the warm paste. The rheological measurements were made at an applied strain of 2% and a frequency of 1 Hz as the paste was heated at 2°C/min from 70 to 95°C, cooled at 1°C/min from 95 to 25°C, and then aged at 25°C for 12 hr in the cell.

RESULTS AND DISCUSSION

Isolation of Wheat Starches

The protease digestions of Karl-92 and Ike wheats, both of which contained 64% starch, gave 85% recovery of their starches. Recovery was 82% for Rio Blanco, which contained 65% starch. Isolation losses in Group I starches may have been due to incomplete release of starch from cells in the digested cracked kernels and to occlusion of starch in the thick tailings fraction. Starch recoveries were 75, 78, and 83% from water-washing of doughs (Group II) made from straight-grade flours milled from Ike, Rio Blanco, and Karl-92 wheats, respectively, which amounted to 66, 69, and 74% recoveries of the starch in the grain. The Group I starches with 82–85% recoveries better represent the starch in a wheat, and they were isolated without exposure to α -amylase. The Group II starches better represent commercial wheat starch produced by the Martin process and, in this work, the three flours contained little α -amylase, as indicated by falling numbers of 348, 384, and >400 sec, respectively, for Ike, Rio Blanco, and Karl-92.

Properties of Starches

The granule size distributions of Group I (protease digestion) wheat starches (Fig. 1) showed that Karl-92 starch contained more small granules ($\leq 10 \mu\text{m}$ diameter) than Ike starch. Rio Blanco starch was intermediate. Most of the small granules in Ike wheat flour were lost when starch was isolated by dough washing (Group II) (Fig 2), and its granule size distribution resembled that of a commercial wheat starch. On the other hand, small granules were retained in Karl-92 starch isolated by dough washing, probably because the 83% recovery was almost the same as the recovery by protease digestion (85%). Generally, wheat contains a bimodal distribution of starch granules with the A-type granules ($\approx 70\%$ of weight of starch) ranging in size from 10–50 μm and the B-type granules from 1–10 μm in size (Seib 1994).

The diameters of A-type granules for Ike from dough-washing ranged from 10–33 μm ; $\approx 86 \text{ wt}\%$ were $\geq 10 \mu\text{m}$ in diameter and

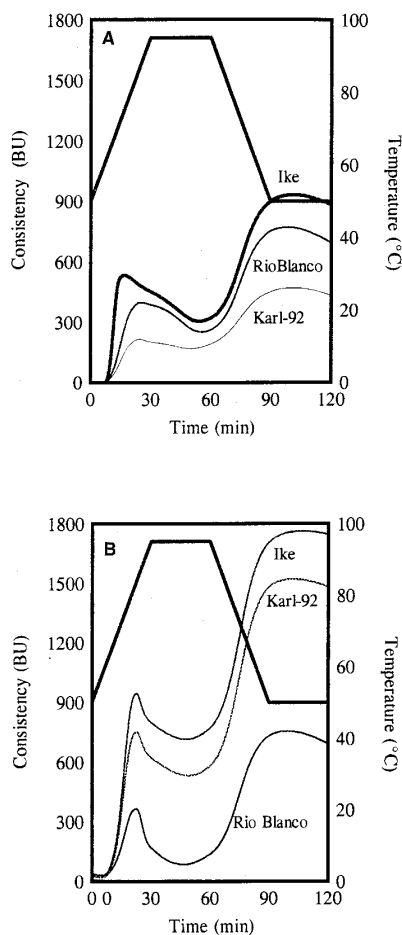


Fig. 3. A, Amylograms (7.5% starch solids) of unmodified starches from Karl-92, Rio Blanco, and Ike isolated by protease method (Group I). B, Amylograms (7.5% starch solids) of low ($\approx 2\%$) hydroxypropylated cross-linked starches from Karl, Rio Blanco, and Ike prepared by reaction of starch with 8.0% (starch basis) propylene oxide at pH 11.5 and 25°C for 24 hr in the presence of 2.0% sodium sulfate (starch basis) followed by reaction with phosphoryl chloride (0.025%, starch basis).

TABLE I
Properties and Components of Wheat Starches Isolated by Protease Digestion (Group I)^a

Sample	Amylose (%)	FAME ^b (mg/100 g)	Blue Value ^c	Protein (%)	SP ₉₂ ^d (g/g)	Differential Scanning Calorimetry ^e			
						T _o (°C)	T _p (°C)	T _c (°C)	ΔH (J/g)
Karl-92	28.0	601a	0.690a	<0.3	14.2a	57.8	63.0a	67.5	11.0a
Ike	23.0	488b	0.627b	<0.3	20.1b	58.3	64.0b	68.8	12.2b
Rio Blanco	26.0	562c	...	<0.3	16.0c	58.5	63.6b	68.3	11.7c

^a Each value is a mean of duplicate measurements. Mean values followed by different letters in the same column are significantly different ($\alpha = 0.05$).

^b Fatty acid methyl ester produced from lipids in the starch.

^c Blue value determined on 1 mg of starch/mL.

^d Swelling power of starch corrected for solubles lost during heating at 92°C.

^e Starch-to-water ratio of 1:3 (w/w). T_o, T_p, and T_c = gelatinization temperatures at onset, peak, and completion.

≈22% were ≥21 μm. By contrast, A-type granules for Karl-92 had diameters ranging from 10–32 μm; ≈78 wt% were ≥10 μm and ≈10% were ≥21 μm. Mean granule diameters for the Ike and Karl-92 wheat starches were 15.9 and 13.1 μm when isolated by dough washing or 2–7% higher than the mean diameters (14.8 and 12.8 μm) for the starches isolated by protease digestion where increased recovery of small granules occurred. Wet processors prefer wheat flours with large starch granules because they sediment rapidly from water and are recovered readily.

Table I gives a comparison of some properties of Group I starches. The amylose content of Karl-92 starch was 28%, 18% higher than that of Ike starch (23%). The lipid in Karl-92 starch determined as FAME was 18% higher than that in Ike starch. These results are in agreement with Morrison et al (1984), who showed a positive correlation between amylose and lipid contents in starches from maize, barley, or rice. The swelling power of Ike starch at a concentration of 1.66% in water at 92°C was 20.1 g/g, which was 40% greater than that for Karl-92 starch (14.2 g/g). The swelling power of Rio Blanco starch under the same conditions was 16.0 g/g. The order of the swelling power is the one expected from its inverse relation to amylose and lipid levels (Morrison et al 1993). The temperatures and enthalpies of gelatinization of the three starches showed that Ike had a somewhat elevated peak temperature and enthalpy of melting because of its higher amylopectin content (Table I).

Modified Starches

Although there was a 10–20% difference in the recoveries of starches from Groups I and II, their swelling powers and gelatinization properties were not different (data not shown). The hydroxypropylation reactions on Group I starches with 8% propylene oxide gave low levels (1.6–2.4%) of hydroxypropyl groups because of the use of a low level of sodium sulfate (2% starch basis). In addition, the cross-linking of Group I starches also was done in 2% salt. On the other hand, starches in Group II were modified to a medium level (≈4%) of hydroxypropyl groups with 8% propylene oxide because of the presence of 10% sodium sulfate. Cross-linking of Group II also was done in 10% salt. Modification of Group I and II starches to the same extent probably would result in the same paste properties.

Modified starches are abbreviated by a shorthand. For example, Ike starch hydroxypropylated to a low level of HP (1.7%) and cross-linked with 0.025% phosphorus oxychloride, is designated Ike-LHP250. Low HP wheat starches contained levels of 1.5–2.4% HP and were cross-linked with two levels (0.025 and 0.075%) of phosphoryl chloride (Table II), whereas the medium HP starches contained levels of 3.6–4.5% HP and were cross-linked at five levels of phosphoryl chloride (0.003–0.075%).

TABLE II
Properties of Unmodified and Modified Wheat Starches with Low Hydroxypropyl (HP) Content

Samples	HP (%)	ΔH (J/g) ^a	SP ₉₂ (g/g) ^b	Paste Clarity (%T ₆₅₀)
Unmodified				
Karl-92	...	11.0	14.2	16.4
Ike	...	12.2	20.1	25.3
Rio Blanco	...	11.7	16.0	22.2
Low HP cross-linked ^c				
Karl-92	1.5	8.9	20.7	12.8
Ike	1.7	8.8	21.8	11.9
Rio Blanco	2.3	8.0	14.4	8.7
Low HP cross-linked ^d				
Karl-92	1.5	8.9	12.6	10.4
Ike	1.8	5.7	13.4	11.8
Rio Blanco	2.4	6.5	10.4	7.0
LSD (α = 0.05)	...	0.6	1.0	2.1

^a Gelatinization enthalpy.

^b Swelling power.

^c Cross-linked at 25°C with 0.025% phosphoryl chloride (starch basis) in aqueous slurry at pH 11.5 with 2% sodium sulfate (starch basis).

^d Cross-linked with 0.075% phosphoryl chloride.

Enthalpies of gelatinization of the modified starches with low and medium HP levels were 5.7–8.9 J/g and 4.8–7.8 J/g, respectively, compared to 11.0–12.2 J/g for the unmodified starches (Tables II and III), which calculated to a maximum enthalpy loss of 49–59%. Gelatinization enthalpies increased with the degree of cross-linking (Table III) but decreased with increasing levels of hydroxypropylation (Table IV). Temperatures of gelatinization also decreased with increasing levels of hydroxypropylation (Table IV). Decreases in melting enthalpy and temperature are consistent with fewer crystals being present after modification (Rutenberg and Solarek 1984) and with a cooperative melting process (gelatinization) enhanced by added swelling (Jenkins and Donald 1998).

In both low- and medium-HP wheat starches, increasing the level of cross-linking decreased clarity (Rutenberg and Solarek 1984, Woo and Seib 1997) (Tables II and III). In addition, because the stringiness of a starch paste increases with the swelling power of a starch, increasing levels of cross-linking generated ever shorter-textured pastes.

Pasting Curves of Starches with a Low Level of HP

Amylograms of the unmodified and modified (low HP) wheat starches at 7.5% starch solids are shown in Figs. 3 and 4. Ike starch gave a paste with a higher consistency when compared to Karl-92 starch, but its paste showed lower shear stability as evidenced by the reduction in consistency during stirring at 95°C for 30 min (Fig. 3a). The difference in paste consistencies was maintained when the two starches were hydroxypropylated to 1.5–1.7% and cross-linked to a low degree (Fig. 3b). On the other hand, Rio Blanco-LHP250, for some unknown reason, gave a low paste consistency. The final paste consistency at 50°C of the doubly modified Ike-LHP250 was >300 BU higher than that of Karl-LHP250 (Fig. 3b). Stringiness of the low HP wheat starches was rated at 1–2 (low).

The low HP Ike, Rio Blanco, and Karl-92 starches also were cross-linked to a high extent to give Ike-LHP750, Karl-LHP750, and Rio-LHP750. All three starch pastes became stable against breakdown at 95°C and generally decreased in consistency (Fig. 4a) compared to those at the low level of cross-linking (Fig. 3b). Still, the doubly modified Ike-LHP750 had almost twice the consistency of the doubly modified Karl-LHP750, whereas doubly modified Rio Blanco-LHP750 showed the lowest consistency (Fig. 4a).

TABLE III
Properties of Modified Wheat Starches with Medium Hydroxypropyl (HP) Content

Samples	HP (%)	ΔH (J/g) ^a	SP ₉₂ (g/g) ^b	Paste Clarity (%T ₆₅₀)
Cross-linked at 0.0%				
Karl-92	4.0	4.8	49.4	48.1
Ike	4.2	7.2	48.9	48.1
Rio Blanco	4.0	4.8	50.8	54.9
Cross-linked at 0.003%				
Karl-92	4.3	5.8	32.0	17.4
Ike	4.2	7.8	29.6	18.3
Rio Blanco	4.2	5.5	32.8	19.5
Cross-linked at 0.006%				
Karl-92	4.0	6.1	22.2	12.1
Ike	4.1	6.7	20.8	13.8
Rio Blanco	4.2	6.2	26.4	15.2
Cross-linked at 0.013%				
Karl-92	4.5	4.9	19.5	6.9
Ike	4.2	6.2	20.2	8.4
Rio Blanco	4.1	6.5	18.0	8.8
Cross-linked at 0.025%				
Karl-92	4.1	5.9	12.9	5.0
Ike	4.3	6.3	14.1	5.8
Rio Blanco	4.3	6.2	13.3	6.2
Cross-linked at 0.075%				
Karl-92	4.1	6.8	10.0	...
Ike	3.9	7.4	9.3	3.9
Rio Blanco	3.6	7.1	9.5	3.6
LSD (α = 0.05)	...	0.5	0.6	1.6

^a Gelatinization enthalpy.

^b Swelling power.

The peak and final consistencies of unmodified Rio Blanco starch were higher than those of Karl-92 starch but lower than those of Ike starch (Fig. 3a). Yet the peak and final consistencies of doubly modified low HP Rio Blanco starches were lower than those of both Karl-92 and Ike (Figs. 3b and 4a). The pastes of Rio Blanco-LHP starches remained fluid at room temperature for several hours, whereas the LHP derivatives of the other two starches formed gels. The efficiency of reactions of Rio Blanco starch with propylene oxide and phosphoryl chloride may have been elevated compared to those of Karl-92 and Ike as indicated, respectively, by its increased level of HP substitution and its decreased clarity (Table II). The reason for the difference in reactivity of Rio Blanco starch is not known, but its amylopectin may have shorter chains with a more highly branched structure compared to that of the other two wheat starches.

Pasting temperature of Ike starch in the amylograms was 2–3°C below that of Karl (≈85°C, Fig. 3a). That difference was maintained in the doubly modified low HP starches that were cross-linked to a low extent (Fig. 3b) but not in those cross-linked to a high extent (Fig. 4a).

Pasting Curves of Starches with a Medium Level of HP

The pasting curves of starches substituted to ≈4.0% hydroxypropylation are shown in Figs. 4b and 5b. Surprisingly, peak and final consistencies of the medium HP starches cross-linked at 0.025 and 0.075% levels of phosphoryl chloride were dramatically lower than their low HP counterparts. This phenomenon was caused by the presence of increased levels (2–10%) of sodium sulfate in the reaction mixture, which increased the efficiency of the cross-linking reaction and dramatically decreased paste viscosity for the starches, as shown previously (Rutenberg and Solarek 1984, Wu and Seib 1990).

The differences in the paste viscosities of the HP 4.0% wheat starches depended on cross-linking because the pasting curves of the HP starches without cross-linking were identical (not shown). The HP 4.0% increased swelling of the wheat starches such that they all swelled to about the same extent, which at 95°C was 49–51 g/g (Table III). Substituting medium level HP in the wheat starches apparently modified the amylose sufficiently so that the difference in levels of amylose was discounted. Cross-linking of the HP 4.0% starches led to some differences in swelling power (Table III).

Ike-MHP30, -MHP60, and -MHP130 starches had paste consistencies higher than those of Midsol 4 (Fig. 4b). The latter is a medium HP wheat starch presumably made from a diverse mixture of hard winter wheats, most of which contain all three waxy proteins. The final consistency of 7.5% Ike-MHP30 was >2,000 BU, which was ≈800 BU greater than the final consistency of Midsol 4, but its paste was stringy with a rating of 4. The paste of Ike-MHP60 was rated slightly less stringy at 3, whereas the paste of Ike-MHP130 was rated at 2.

Surprisingly, Karl-MHP60 gave an amylogram at 6.25% starch solids that almost matched that of Ike-MHP60, as did that of Rio Blanco-MHP60 (data not given), even though the swelling powers of those doubly modified starches measured at low concentration (1.67%) showed some differences (Table III). The pasting curves of Karl-MHP130 and Rio Blanco-MHP130 matched that of Midsol 4 in Fig. 5a, whereas Ike-MHP130 gave a somewhat higher pasting curve. All three starches with medium HP and cross-linked with 0.025% phosphoryl chloride gave low pasting curves (Fig. 5b).

Rheology of Unmodified Starches

The storage modulus (G') and $\tan \delta$ values of unmodified Karl-92, Rio Blanco, and Ike starches were recorded at 7.5% starch solids during heating (≈12.5 min), cooling (≈70 min), and aging (≈12 hr), all inside the measurement cell of the Bohlin rheometer. Among the unmodified starches, Ike starch exhibited the highest G' at all three stages. Only the data on the gels during aging at 25°C are given in Fig. 6a; the $\tan \delta$ values were 0.1, 0.2, and 0.4 for Ike, Rio Blanco, and Karl-92, respectively. When $\tan \delta$ is >0.1–0.2, viscous forces add to elastic forces to give the composite force vector. Viscous forces occurred in the aged gels of unmodified Karl-92 starch at 7.5% solids. The high swelling of Ike wheat starch appeared to release more amylose into the continuous aqueous phase of its paste than swelling of Karl-92 starch, in spite of the 17% higher amylose content in Karl-92. The higher amylose in the continuous phase of the Ike wheat starch paste generated a stronger continuous gel phase.

Rheology of HP Starches

The rheologies of cooked and cooled pastes of the modified wheat starches also were followed in the Bohlin instrument at 7.5% starch solids. Ike-LHP250 gave a maximum G' during heating at 87.7°C, and then it decreased in magnitude upon heating to 95°C (data not shown). The value of G' continued to drop during the cooling cycle until a temperature of 88°C was reached, after which it increased and continued to rise further as cooling was continued to 25°C, at which $\tan \delta = 0.08$. Karl-LHP250 ($\tan \delta = 0.06$ at 25°C) and Rio Blanco-LHP250 ($\tan \delta = 0.09$) gave profiles similar to that of Ike with similar values of G' (data not shown).

The values of G' for the various gels of low HP wheat starches as they were stored for 24 hr at 25°C are shown in Fig. 6b. At the low level of cross-linking, Fig. 6b shows the G' values of modified Rio Blanco-LHP250 and Ike-LHP250 were separated by ≈100 Pa, and they remained constant from 2 to 24 hr. However, the lightly modified Karl-92 behaved differently, and its G' value increased steadily during storage at 25°C up to 12 hr. The amylopectin in Karl-LHP25 may have retrograded slowly and stiffened the granules, which translated into a strengthening of the solid phase (Eliasson and Bohlin 1982, Ring 1985).

TABLE IV
Gelatinization Temperatures^a and Enthalpies of Unmodified Wheat Starches and Low and Medium Hydroxypropylated (HP)/Cross-Linked Modified Forms

Starch Base	T_o (°C)	T_c (°C)	T_p (°C)	ΔH^b (J/g)
Karl-92				
Unmodified	57.8	67.5	63.0	11.0
Low HP cross-linked	56.1	64.5	60.2	8.9
Medium HP cross-linked	49.4	60.1	54.6	4.9
Ike				
Unmodified	58.3	68.8	64.0	12.2
Low HP cross-linked	56.1	65.9	60.9	8.8
Medium HP cross-linked	50.1	61.7	55.7	6.2
Rio Blanco				
Unmodified	58.5	68.3	63.6	11.7
Low HP cross-linked	56.1	65.3	60.6	8.0
Medium HP cross-linked	49.8	60.3	54.8	6.5
LSD ^c ($\alpha = 0.05$)	0.3	0.7	0.5	...

^a Determined at starch-to-water ratio of 1:3 (w/w) in differential scanning calorimeter. T_o , T_p , and T_c = gelatinization temperatures at onset, peak, and conclusion.

^b Gelatinization enthalpy.

^c Least significant difference.

At the high level of cross-linking, Ike-LHP750 showed maximum G' at 91.6°C, and then G' decreased during heating to 95°C and cooling to 82°C, after which it increased as cooling continued to 25°C. Karl-LHP750 and Rio-LHP750 displayed similar profiles, but the heating, cooling, and aging maxima for G' were lower than those of the corresponding Ike derivative (Ike-LHP750). Fig. 6b shows that Ike-LHP750 had the highest value of G' upon storage at 25°C when compared to that of Karl-LHP750 and Rio Blanco-LHP750.

Heating aqueous slurries (7.5% starch solids) of medium-level HP starches with a low level of cross-linking, Ike-MHP130, Karl-MHP130, and Rio-MHP130, in the rheometer showed maximum G' at ≈80°C, followed by a decrease in G' during heating to 95°C (data not shown). G' increased for all three starches as cooling commenced and continued to increase to 25°C. Midsol 4 had a similar profile, with $\tan \delta$ values of 0.09 during cooling and 0.06 during aging, both of which were nearly identical to those obtained for Karl-MHP130 and Rio-MHP130. Ike-MHP130 gave $\tan \delta$ values of 0.1 and 0.13, respectively. Figure 7a shows the G' value of the starch gels from this set as they were aged for 12 hr. G' remained constant at 2–12 hr for all medium HP wheat starches, but Karl-

MHP130 gave a slightly higher G' than the other medium HP wheat starches cross-linked with 0.013% phosphoryl chloride.

The complex modulus values of cooked starches with a medium level of HP but a high level of cross-linking, Ike-MHP250, Karl-MHP250, and Rio-MHP250, showed considerable viscosity (G'') after cooling to 25°C as evidenced by $\tan \delta$ values of 0.26, 0.28, and 0.59, respectively (data not shown). These results indicate that the high level of cross-linking inhibited leaching of amylose from the granules during pasting. The continuous phase of the 7.5% starch pastes contained too low a concentration of amylose to form a strong gel embedded with swollen granules.

Unmodified Ike starch swelled more than unmodified Karl-92 and Rio Blanco starches in hot water (<10% starch solids), and more amylose apparently leached out of its granules to form a strong gel network (Fig. 6a). As the starches were hydroxypropylated to a low degree, their swelling powers increased, and their behaviors began to be more similar. As HP content was increased to the medium level of ≈4%, the gel properties of the cross-linked starches largely converged, as shown in Fig. 7.

Cross-linking is important in determining gel strength, as illustrated by Rio Blanco LHP starches in Fig. 6b. Rio-LHP750 gave a much lower storage modulus (≈100 Pa) when compared to Rio-LHP250 (≈600 Pa), apparently because the granules of Rio-LHP750 did not swell fully and less amylose was leached from them. This

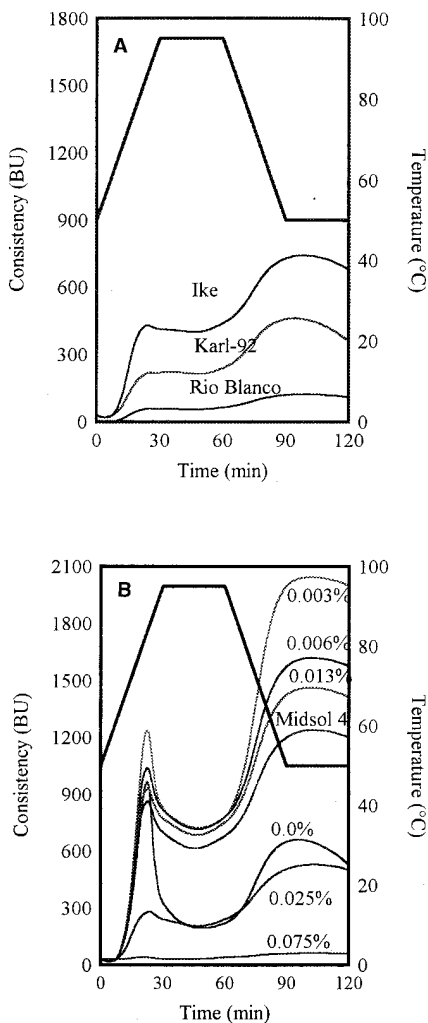


Fig. 4. A, Amylograms (7.5% starch solids) of low (≈2%) hydroxypropylated cross-linked starches (LHP), Karl-LHP750, Rio Blanco-LHP750, and Ike-LHP750. **B,** Amylograms (7.5% starch solids) of medium (≈4%) hydroxypropylated (MHP) Ike starch cross-linked with different levels of phosphoryl chloride prepared by reaction of starch with 8% (starch basis) propylene oxide at pH 11.5 and 25°C for 24 hr in the presence of ≈10% sodium sulfate (starch basis) followed by reaction with phosphoryl chloride (starch basis). Midsol 4 is a commercial hydroxypropylated cross-linked wheat starch.

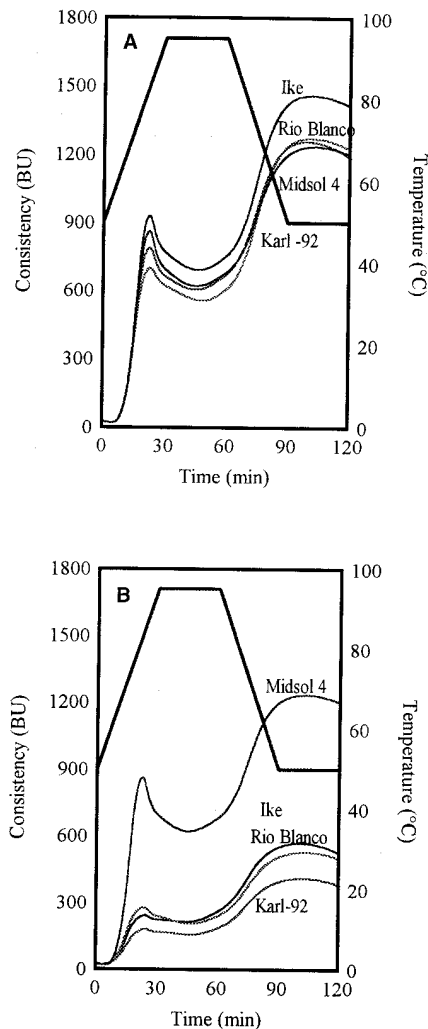


Fig. 5. A, Amylograms (7.5% starch solids) of medium (≈4%) hydroxypropylated (MHP) cross-linked (0.013% phosphoryl chloride) starches from Ike, Karl-92, and Rio Blanco. **B,** Amylograms (7.5% starch solids) of MHP cross-linked (0.025% phosphoryl chloride) starches. Midsol 4 is a commercial hydroxypropylated cross-linked wheat starch.

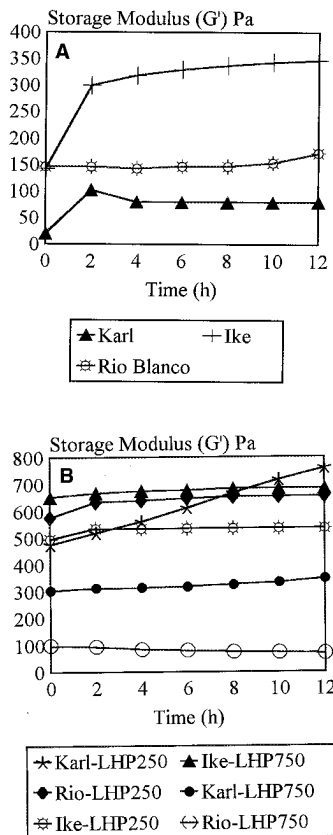


Fig. 6. Storage modulus of gels (7.5% solids) of unmodified (A) and low hydroxypropylated cross-linked starches (LHP250 and LHP750) (B) during aging for 12 hr at 25°C.

also can be seen in medium-HP starches cross-linked with 0.025% versus 0.013% phosphoryl chloride (Fig. 7a and b); the latter starches all showed a larger storage modulus and lower $\tan \delta$.

Each rheological experiment was repeated three times. The trend was always the same, but the exact G' values varied and were somewhat unstable during the heating stage, especially for the unmodified starches. At low concentrations of $\approx 7.5\%$ starch, the variability tended to be high. Sample preparation also may have introduced some errors. The curves reported here are those that are nearest to the mean.

Freeze-Thaw Stability

After Ike-LHP250 paste was subjected to the five freeze-thaw cycles, it formed a spongy mass that released water when squeezed between the fingers, but not when centrifuged at $1,500 \times g$. Unmodified Rio Blanco starch paste behaved in a similar manner. Ike-MHP250 paste gave 10% water of syneresis after the fourth freeze-thaw cycle and centrifuging, but Ike-MHP130, Ike-MHP60, and Ike-MHP30 did not synerese even after eight freeze-thaw cycles. The low level of cross-linking in those three hydroxypropylated Ike starches apparently allowed high swelling in the amylopectin molecules, which retarded retrogradation. Modified Ike starch pastes were generally more freeze-thaw stable than similarly modified Karl-92 starch pastes.

CONCLUSIONS

Substitution of $\approx 2\%$ HP in the high swelling wheat starch Ike followed by optimum cross-linking gives a doubly modified wheat starch that has a thickening power about the same as that of a low swelling wheat starch, such as Karl-92, substituted with 4% HP and cross-linked to optimum. Etherifying wheat starch to 4% HP or higher appears to cancel differences in thickening powers among starches from partial waxy wheats.

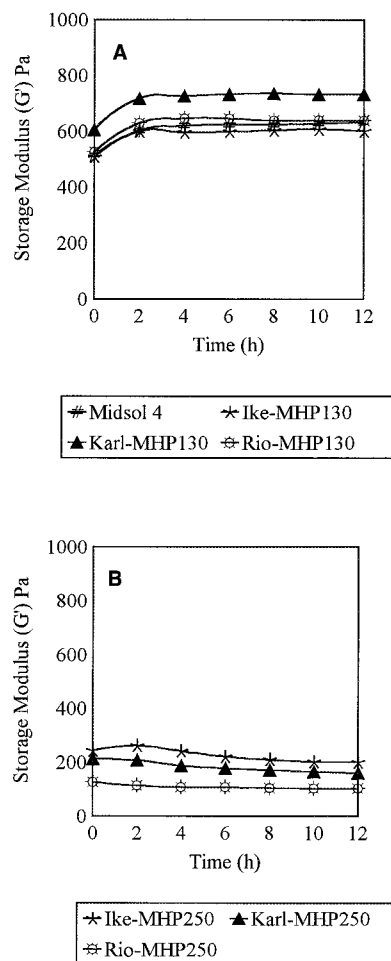


Fig. 7. Storage modulus of gels (7.5% solids) of medium hydroxypropylated starches cross-linked with 0.013% POCl_3 (MHP130) (A) and cross-linked with 0.025% POCl_3 (MHP250) (B) during aging for 12 hr at 25°C. Midsol 4 is a commercial hydroxypropylated cross-linked wheat starch.

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