

Factors Governing Pasting Properties of Waxy Wheat Flours

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ABSTRACT

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Waxy wheat (*Triticum aestivum* L.) contains endosperm starch lacking in amylose. To realize the full potential of waxy wheat, the pasting properties of hard waxy wheat flours as well as factors governing the pasting properties were investigated and compared with normal and partial waxy wheat flours. Starches isolated from six hard waxy wheat flours had similar pasting properties, yet their corresponding flours had very different pasting properties. The differences in pasting properties were narrowed after endogenous α -amylase activity in waxy wheat flours was inhibited by silver nitrate. Upon treatment with protease, the extent

of protein digestibility influenced the viscosity profile in waxy wheat flours. Waxy wheat starch granules swelled extensively when heated in water and exhibited a high peak viscosity, but they fragmented at high temperatures, resulting in more rapid breakdown in viscosity. The extensively swelled and fragmented waxy wheat starch granules were more susceptible to α -amylase degradation than normal wheat starch. A combination of endogenous α -amylase activity and protein matrix contributed to a large variation in pasting properties of waxy wheat flours.

Starch in endosperm of normal wheat (*Triticum aestivum* L.) consists of ~25% amylose (mostly linear) and ~75% amylopectin (highly branched), whereas starch in endosperm of waxy wheat essentially contains only amylopectin (Graybosch 2005). Waxy wheat is a potentially valuable specialty wheat. The development, unique properties, and uses of waxy wheats in noodles, bread, and tortillas have been reviewed (Graybosch 1998; Seib 2000; Chibbar and Chakraborty 2005; Hung et al. 2006; Maningat et al. 2009). Because of its increased swelling and moisture-holding capacity, the addition of waxy wheat flour may provide increased softness and extended shelf life, as well as the potential to reduce fat in bakery foods. In dry cereal foods, such as instant breakfast cereals, crackers, and salty snacks, adding waxy wheat flour has been suggested as a means to soften a product's bite and increase its friability (Maningat et al. 2009). In addition, waxy wheat flour has the potential to be wet-milled to yield two valuable coproducts, waxy wheat starch and vital wheat gluten (Guan et al. 2009). Waxy wheat starch can be converted to maltodextrins or modified to produce thickeners with a low pasting temperature and a low rate of retrogradation (Maningat et al. 2009).

When waxy wheat flours are heated in excess water, they exhibit different properties compared with normal wheat flours (Graybosch et al. 2000; Ishida et al. 2003; Kim et al. 2003; Graybosch 2005). Waxy wheat flours exhibit peak viscosity at a lower temperature than normal wheat flour (Kim et al. 2003); this property can be used as an indicator to identify waxy wheat flours. In addition, waxy wheat flours have lower final viscosity during pasting than normal wheat flours (Yasui et al. 1999; Geera et al. 2006). However, reports are conflicted on the effect of the waxy character on peak viscosity when analyzed in distilled water. Some reports suggest that waxy wheat flours have higher peak viscosity than normal wheat flours (Abdel-Aal et al. 2002; Morita et al. 2002), whereas other studies have found that normal flours have higher peak viscosity than waxy wheat flours (Morris et al. 1998; Yasui et al. 1999; Ishida et al. 2003). The discrepancies have not been well investigated. In addition, Graybosch et al. (2003) found significant genetic variation in flour peak viscosities among 19 spring waxy wheats. Thus, factors other than waxy starch per se must be influencing this trait.

When examining hard waxy wheats from advanced breeding lines, we noted that different hard waxy wheat flours have different dough-mixing properties and gluten indexes (Guan et al. 2009). Our continued investigations have revealed large variations in pasting properties among hard waxy wheat flours. In addition to bakery applications (Garimella Purna et al. 2011), waxy wheat flour has the potential to be used as a thickener in foods; however, the inconsistency in pasting properties would be a concern and would affect the use of waxy wheat flours as a thickener. In light of the conflicting reports in the literature and our own observations, the objective of this study was to investigate and determine the factors governing the pasting properties of waxy wheat flours and compare their pasting properties with those of normal and partial waxy wheat flours.

MATERIALS AND METHODS

Materials. Six hard waxy wheat samples (NX03Y2114, NX03Y2115, NX03Y2205, NX03Y2315, NWX03Y2459, and NX03Y2489), one hard partial waxy wheat (Trego), and one hard normal red winter wheat (Karl 92) were used. Trego is a hard white wheat with null at the *Wx-B* locus. The single kernel characteristics of all the wheats including hardness index and the pedigrees of the waxy wheat lines were reported by Guan et al. (2009). The last four digits in each waxy sample identification are used in this paper. Wheat kernels were tempered to 16% moisture for 18 h and were roller-milled into straight-grade flour on a Bühler MLU 202 experimental mill (Bühler, Uzwil, Switzerland). Flour yield for all wheat samples was previously reported (Guan 2008). Starches were isolated by a dough-washing method, and protein content of the isolated starches as well as yield and recovery of the starches were previously reported (Guan et al. 2009).

Proximate Analysis and Analytical Tests on Flours. Moisture and protein contents of the eight flour samples were measured by AACC International Approved Methods 44-15.02 and 46-30.01, respectively, and were previously reported by Guan et al. (2009). Total and damaged starch contents were determined by AACCI Approved Methods 76-13.01 and 76-30.02, respectively, with assay kits from Megazyme International Ireland (Bray, Ireland). α -Amylase activity of flours was determined by AACCI Approved Method 22-02.01 with a Megazyme assay kit. Enzyme activity was reported in Ceralpha units (CU), in which one unit of activity is defined as the amount of enzyme in the presence of excess thermostable amyloglucosidase required to release 1 μ mol of *p*-nitrophenol from end-blocked *p*-nitrophenyl maltoheptaoside in 1 min under the defined assay conditions. Arabinoxylan content expressed as D-xylose was determined with phloroglucinol colorimetric reagent (Douglas 1981).

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Particle Size Analysis. Particle size distribution for each flour was determined with a single-wavelength Beckman Coulter LS 13 320 particle size analyzer (Beckman Coulter, Miami, FL, U.S.A.). Each sample was analyzed in duplicate, and the mean particle size (in micrometers), assuming spherical particles, was reported.

Pasting Properties. Flour pasting properties were determined with a Rapid Visco Analyzer (RVA) (Perten Instruments North America, Springfield, IL, U.S.A.). A mixture of flour (10% solids) in one of three solvents was prepared in an RVA canister. Final total weight in the RVA canister was 28 g. An RVA paddle was inserted into the canister, and the mixture was gently agitated to disperse flour lumps. The RVA canister was then subjected to a 13 min RVA test to determine flour pasting properties (Deffenbaugh and Walker 1989). The RVA pasting curve profile included holding the sample at 50°C for 1 min followed by heating the sample from 50 to 95°C in 3 min, holding the sample at 95°C for 3 min, cooling the sample back to 50°C in 4 min, and holding the sample at 50°C for 2 min. The RVA curves were analyzed for pasting properties (i.e., pasting temperature, peak viscosity, viscosity at trough, final viscosity, and setback viscosity) with Thermocline for Windows 3 software provided with the RVA. Pasting temperature was obtained by using the function TempAtViscRate (1,6,0.1,50). Setback viscosity was calculated as difference between final viscosity and viscosity at trough.

Three solvents were used: distilled water, 1mM silver nitrate (AgNO₃) solution, and 1mM AgNO₃ solution plus protease from *Streptomyces griseus* (P-5147, 4.5 units/mg of protein, Sigma, St. Louis, MO, U.S.A.). The effect of protease on pasting properties of flours was measured as described by Zhu et al. (2010). Flours (2.8 g, db) were suspended in water (12.5 g) containing 18 mg of protease and incubated at 37°C for 30 min. Subsequently, 2mM AgNO₃ was added to the protease-hydrolyzed flour for a total weight of 28 g. The objective of adding AgNO₃ was to inhibit the action of α -amylase on flours during the RVA test, thereby enabling us to study the specific effects of protease on flour.

Pasting properties of isolated starches were determined at 7% solids in water or a solution containing 0.01% α -amylase from porcine pancreas (A-3176, Sigma).

In a separate experiment, flour samples (10% solids) were treated with protease in AgNO₃ solution (containing 18 mg of protease) at 30°C for 30 min. The final weight of the flour sample and protease-containing AgNO₃ solution was 28 g. Subsequently, an aliquot (1 mL) of the flour suspension was centrifuged at 10,000 \times g and the supernatant was discarded. The residual protein in the pellet was analyzed with a LECO FP-428 nitrogen determinator (LECO, St. Joseph, MI, U.S.A.) and expressed as a percentage of the original protein content of the samples. Each sample was analyzed in duplicate.

Gel Permeation Chromatography (GPC) of Debranched Starches. Starch (20 mg) was added to 10 mL of 0.01M acetate buffer at pH 4.0 in a 12 mL glass vial with a microstirring bar. The

vial was placed in a boiling water bath on a stir plate for 1 h. Isoamylase (50 μ L) (EC 3.2.1.68, Hayashibara Biochemical Laboratories, Okayama, Japan) was added to the solution after cooling to room temperature. The vial was placed at 50°C in a water bath overnight, frozen in a dry ice/acetone bath, and freeze-dried. The freeze-dried debranched starch (4 mg, db) was dissolved in 4 mL of dimethyl sulfoxide by heating in a boiling water bath for 24 h with constant stirring. GPC analysis was performed with a Polymer Laboratories PL-GPC 220 integrated GPC/size-exclusion chromatography fully automated system (Polymer Laboratories, Amherst, MA, U.S.A.) as previously described (Cai et al. 2010).

Light Microscopy. The changes in morphology of starch granules during heating in water were examined by light microscopy. Starch (0.1 g) was added to distilled water (10 mL) and dispersed by using a vortex mixer. A small amount of high-vacuum grease (Dow Corning, Midland, MI, U.S.A.) was spread evenly as an extremely thin layer around the edge of a square-shaped coverslip. One drop of the starch suspension was transferred onto a slide and covered by the coverslip. The starch was viewed with an Olympus BX51 microscope (Olympus America, Melville, NY, U.S.A.) equipped with an STC200 hot stage. Images and photographs were captured under $\times 40$ objective with a SPOT Insight camera and SPOT 4.6 for Windows software (Diagnostic Instruments, Sterling Heights, MI, U.S.A.). The heating rate was set at 5°C/min.

Thermal Properties of Flours. Thermal properties of flours were determined by differential scanning calorimetry (DSC) (Q100 DSC, TA Instruments, New Castle, DE, U.S.A.). For gelatinization properties, each flour sample and distilled water were added to the DSC pan in a 1:2 ratio (w/w). The pan was hermetically sealed and held at 25°C for 24 h. The samples were then heated from 10 to 140°C at 10°C/min. An empty DSC pan was used as a reference. Onset, peak, and end temperature along with enthalpy were determined. Each sample was analyzed in duplicate, and mean values were reported.

Statistical Analysis. MacAnova 4.12 software (School of Statistics, University of Minnesota, Minneapolis, MN, U.S.A.) was used to perform ANOVA and Tukey's honest significance difference analysis. The level of significance was $P < 0.05$ throughout the paper. Each sample was analyzed at least in duplicate.

RESULTS AND DISCUSSION

Composition and Particle Size of Flours. Protein content of waxy wheat flours ranged from 12.0 to 15.1% (db), whereas the normal wheat flour (Karl 92) and the partial waxy wheat flour (Trego) had protein contents of 15.47 and 14.04% (db), respectively (Guan et al. 2009; Table I). The protein content of Karl 92 was significantly higher than most of the waxy wheat flours except for sample 2205. Total starch content of the six waxy wheat flours was between 75.0 and 81.7% (db), whereas Karl 92 and Trego had

TABLE I
Damaged Starch, Arabinoxylan Content, and α -Amylase Activity of Wheat Flours^y

Sample	Protein Content ^z (%)	Total Starch Content ^z (%)	% Damaged Starch	% Arabinoxylan Content (as D-Xylose)	α -Amylase Activity (CU/g)	Mean Flour Particle Size (μ m)
2114	13.88 \pm 0.1b	75.0	6.02 \pm 0.10e	1.73 \pm 0.15a	0.12 \pm 0.03bc	34 \pm 0.6f
2115	13.78 \pm 0.1b	81.7	8.98 \pm 0.07bc	1.78 \pm 0.29a	0.20 \pm 0.02a	37 \pm 0.2e
2205	15.10 \pm 0.0a	78.3	10.08 \pm 0.03b	1.36 \pm 0.09a	0.09 \pm 0.01cd	54 \pm 0.1a
2315	12.00 \pm 0.0e	81.7	8.52 \pm 0.25c	2.05 \pm 0.42a	0.14 \pm 0.02b	47 \pm 0.0b
2459	13.33 \pm 0.2c	78.3	11.38 \pm 0.69a	1.65 \pm 0.09a	0.13 \pm 0.00b	42 \pm 0.0d
2489	12.82 \pm 0.1d	80.0	9.33 \pm 0.04bc	1.59 \pm 0.09a	0.08 \pm 0.01de	41 \pm 0.2d
Karl 92	15.47 \pm 0.0a	76.7	7.19 \pm 0.35d	1.69 \pm 0.05a	0.03 \pm 0.00f	45 \pm 0.1c
Trego	14.04 \pm 0.2b	76.7	6.77 \pm 0.06de	1.26 \pm 0.08a	0.04 \pm 0.00ef	46 \pm 0.5c

^y Mean \pm SD values are reported. Means not sharing the same letter within each column are significantly different ($P < 0.05$). Full sample identification numbers: NX03Y2114, NX03Y2115, NX03Y2205, NX03Y2315, NWX03Y2459, and NX03Y2489. CU = Ceralpha unit.

^z Values reported by Guan et al. (2009).

76.7% (db), as reported by Guan et al. (2009). Although starch content of wheat flours varied, no correlation emerged between starch content and pasting properties, as discussed in the next section. Other factors besides starch content affected the pasting properties of wheat flours.

Damaged starch values for flours ranged from 6.02 to 11.38% (Table I). Most waxy wheat flours had significantly higher damaged starch content than the normal wheat flour and partial waxy wheat flour. This could be owing to greater susceptibility of waxy wheat to mechanical damage during milling (Bettge et al. 2000). No significant differences were present in arabinoxylan content between Karl 92 and waxy wheat flours (Table I). Our results differ from those of Sayaslan et al. (2006), who reported 20–30% higher arabinoxylan (pentosan) content in waxy wheat samples than in normal wheat flour (Karl 92). The underlying reasons for the different results are unclear but could be owing to the different cultivars used in the pedigree of waxy wheat lines in this study or owing to differences in cultural environments.

Except for sample 2489, waxy wheat flours exhibited significantly higher α -amylase activity compared with normal wheat flour (Karl 92) and partial waxy wheat flour (Trego). Among waxy wheat flours, sample 2115 had the highest α -amylase activity (0.20 CU/g), and sample 2489 had the lowest α -amylase activity (0.08 CU/g), whereas normal wheat flour (Karl 92) had an α -amylase activity of 0.033 CU/g.

Mean particle size of all eight flours varied within a narrow range of 34–54 μ m (Table I), which is within the range of medium fraction for flours (Wang and Flores 2000). Flour particle size could influence both the extent of starch damage (Wang and Flores 2000) and subsequent enzyme activity in our proposed RVA studies.

Debranched waxy wheat starch showed a bimodal distribution with similar proportions of the two fractions as debranched normal wheat starch (curves almost overlaying on top of each other, data not shown), indicating that the molecular structure of the waxy wheat starch was close to that of amylopectin in the normal wheat starch. Reports of the amylopectin structure of waxy wheats and

their parent lines have conflicted. Hayakawa et al. (1997) reported that amylopectin in waxy wheat starch had significantly higher degree of polymerization values than amylopectin in starch isolated from their parent lines (nonwaxy wheat). In a more recent study, Yoo and Jane (2002) showed that waxy wheat amylopectin lacked the extralong branched chains that are present in normal wheat, whereas Yasui et al. (1996) reported similar amylopectin chain length distribution profiles for both waxy and parent lines. In our study, we observed little difference in amylopectin chain length distribution among the advanced waxy wheat lines.

Pasting Properties of Flours. Pasting properties of flours in different solutions are given in Table II and Figure 1. In all solvents, the waxy wheat flours exhibited lower pasting temperatures ($\sim 70^\circ\text{C}$) than normal wheat flour ($\sim 90^\circ\text{C}$). To compare the whole pasting profile, one waxy wheat flour (sample 2489) that had a low viscosity and another waxy wheat flour (sample 2115) that had a high viscosity in water were selected and compared with normal wheat flour (Fig. 1).

Distilled Water. Waxy wheat flours had a significantly lower peak temperature ($\sim 67^\circ\text{C}$) in water than normal (93°C) and partial waxy wheat flours (91°C). No significant differences were observed in the pasting temperatures among waxy wheat flours. The low pasting and peak temperatures for these waxy wheat flours agreed with previous findings on waxy wheat flours (Yasui et al. 1999; Ishida et al. 2003) and isolated waxy wheat starches (Hayakawa et al. 1997; Sasaki et al. 2000; Grant et al. 2001; Kim et al. 2003).

We observed remarkable variations in peak viscosity for waxy wheat flours in water, ranging from 472 cP (sample 2489) to 2,011 cP (sample 2205) (Table II). Samples 2114 and 2205 had peak viscosities similar to Karl 92, but the rest of the waxy wheat flours had significantly lower peak viscosities than Karl 92. Among waxy wheat flours, sample 2489 exhibited the lowest peak viscosity despite low α -amylase activity.

Waxy wheat flours had significantly lower trough (hot paste) viscosity and exhibited greater breakdown compared with normal and partial waxy wheat flours. The great reduction in the viscosity

TABLE II
Pasting Properties of Wheat Flours in Different Solvents ($N = 2$)^y

Sample	Peak Viscosity (cP)	Trough Viscosity (cP)	Final Viscosity (cP)	Setback Viscosity (cP)
Distilled water				
2114	1,884 \pm 137b	734 \pm 32b	1,038 \pm 28b	305 \pm 4b
2115	1,592 \pm 6c	616 \pm 6c	852 \pm 8c	236 \pm 14c
2205	2,011 \pm 15b	731 \pm 15b	1,019 \pm 25b	289 \pm 11bc
2315	1,461 \pm 6c	626 \pm 6c	920 \pm 11c	294 \pm 5bc
2459	1,450 \pm 1c	392 \pm 1d	550 \pm 6d	159 \pm 6d
2489	472 \pm 13d	38 \pm 13e	73 \pm 4e	35 \pm 9e
Karl 92	1,971 \pm 15b	1,125 \pm 15a	2,118 \pm 25a	994 \pm 11a
Trego	2,389 \pm 11a	1,151 \pm 11a	2,168 \pm 47a	1,018 \pm 36a
Silver nitrate (1mM)				
2114	3,488 \pm 12bc	1,180 \pm 3bc	1,668 \pm 19b	488 \pm 16c
2115	3,319 \pm 38c	1,148 \pm 20cd	1,548 \pm 20bc	400 \pm 0de
2205	3,623 \pm 51ab	1,208 \pm 23bc	1,633 \pm 45b	426 \pm 22cde
2315	2,924 \pm 31d	1,084 \pm 6d	1,549 \pm 21bc	465 \pm 14cd
2459	3,713 \pm 63a	1,098 \pm 22d	1,462 \pm 41cd	365 \pm 19e
2489	2,810 \pm 103d	1,014 \pm 25e	1,380 \pm 45d	367 \pm 21e
Karl 92	2,222 \pm 19e	1,242 \pm 8b	2,871 \pm 28a	1,629 \pm 20a
Trego	2,764 \pm 30d	1,365 \pm 2a	2,775 \pm 17a	1,411 \pm 19b
Protease and silver nitrate ^z				
2114	1,929 \pm 26b	835 \pm 23c	1,230 \pm 27d	396 \pm 4cde
2115	2,270 \pm 12a	510 \pm 8e	782 \pm 17e	273 \pm 9e
2205	2,238 \pm 24a	599 \pm 6d	895 \pm 2e	296 \pm 4de
2315	1,778 \pm 6b	739 \pm 3c	1,138 \pm 5d	399 \pm 8c
2459	2,262 \pm 41a	872 \pm 7b	1,192 \pm 24cd	320 \pm 17cde
2489	2,276 \pm 21a	901 \pm 6b	1,268 \pm 11c	367 \pm 5cd
Karl 92	1,652 \pm 19b	886 \pm 7b	1,987 \pm 18b	1,101 \pm 11b
Trego	2,333 \pm 12a	1,123 \pm 11a	2,455 \pm 4a	1,332 \pm 14a

^y Mean \pm SD values are reported. Means not sharing the same letter within each column are significantly different ($P < 0.05$). Full sample identification numbers: NX03Y2114, NX03Y2115, NX03Y2205, NX03Y2315, NWX03Y2459, and NX03Y2489.

^z Flours digested with protease (80 U) pasted in silver nitrate (1mM).

of waxy wheat flours could be owing to the lack of granular rigidity of waxy wheat starches when heated in excess water (Guan 2008). Final viscosity of waxy wheat flours was significantly lower than Trego and Karl 92 flours (Table II), which could be attributed to the lack of amylose in waxy wheat flours and thus the inability to form a gel matrix (Leloup et al. 1991). Because of the lack of amylose, the setback viscosities of the waxy wheat flours were much lower than those of the normal and partial waxy wheat flours (Table II).

Silver Nitrate Solution. To examine the influence of α -amylase activity, pasting properties of flours were analyzed in the presence

of AgNO_3 (Fig. 1B). Previous researchers have utilized silver nitrate solution to inhibit α -amylase activity in sprouted and normal wheat flours (Crosbie and Lambe 1993; Bhattacharya and Corke 1996; Batey et al. 1997; Bhattacharya et al. 1997; Yasui et al. 1999; Abdel-Aal et al. 2002; Mariotti et al. 2005). We observed an increase in peak viscosity for all wheat flours when α -amylase activity was inhibited, but the overall increase in pasting properties was greater in waxy wheat flours compared with normal and partial waxy wheat flours (Table II, Fig. 1A and B). Peak viscosity of the waxy wheat flours was significantly higher than normal wheat flour when tested in the presence of AgNO_3 . The differences in peak viscosities among waxy wheat flours were narrowed when α -amylase activity was inhibited. Our results suggest that waxy flours were more susceptible to amylolytic degradation than normal wheat flour, but the extent of degradation differed among the waxy wheat flours. To understand the full scope of amylolytic degradation in waxy wheat flour, separate studies on isolated waxy wheat starches were conducted and are reported in the following section.

The hot paste viscosity and final viscosity of the waxy wheat flours were still lower than that of the normal and partial waxy wheat flours in silver nitrate solution (Table II), reflecting the larger breakdown of the waxy wheat flours. The final viscosity and setback viscosity of the waxy wheat flours were also lower (Table II) owing to the lack of amylose.

Pasting Properties of Isolated Starches and Their Susceptibility to α -Amylase. Starch is the major flour component and is largely responsible for pasting properties of flour. Hence, starches were isolated from flours, and their pasting properties were determined. Interestingly, starches isolated from the six waxy wheat flours had similar pasting properties (Fig. 2). Waxy wheat starch had a greater increase in viscosity at a lower temperature than normal wheat starch, indicating that waxy wheat starch developed viscosity more rapidly. In fact, the peak viscosity of the waxy wheat starches was more than 2.5 times higher than that of the normal wheat starch. Baik and Lee (2003) reported that the peak viscosity was greater than 1,780 BU for two waxy wheat starches but only 210 BU for a normal wheat starch at 8.9% starch solids. Amylopectin has been suggested to be responsible for swelling of starch granules (Tester and Morrison 1990), whereas amylose and amylose-lipid complex inhibit granule swelling (Tester and Morrison 1990, 1992; Sasaki and Matsuki 1998). The high peak viscosity of waxy wheat starch suggests that it has great potential to be used as a base starch to make modified starch products as thickeners in food applications. The large breakdown in viscosity can be eliminated or reduced by chemical cross-linking of waxy wheat starch (Reddy and Seib 2000).

To further understand the susceptibility of waxy wheat starches to enzyme activity, low levels of exogenous α -amylase (0.01% v/w) were added to starches isolated from waxy wheat and normal wheat (Karl 92). We observed a significant decrease in paste viscosity of waxy wheat starch but only a small change in pasting properties of normal wheat starch (Fig. 3). The final viscosity of normal wheat starch decreased from about 1,000 to 750 cP, whereas the final viscosity of waxy wheat starch dropped from about 1,250 cP to less than 100 cP. The greater loss in viscosity further validated the point that waxy wheat starches were more susceptible to α -amylase activity than normal wheat starch. The added α -amylase was not a thermostable enzyme but active at 60–80°C. Waxy wheat starch swelled more at lower temperatures (60–80°C) and thus was more prone to α -amylase hydrolysis.

Our hot-stage microscopic results (Fig. 4) demonstrate the fate of starch granules of waxy and normal wheat upon heating in excess water and help to understand the pasting properties of the starches. At 73°C, normal wheat starch granules maintained granular integrity, whereas waxy starch granules swelled excessively and lost their granular integrity. These observations help to explain the large

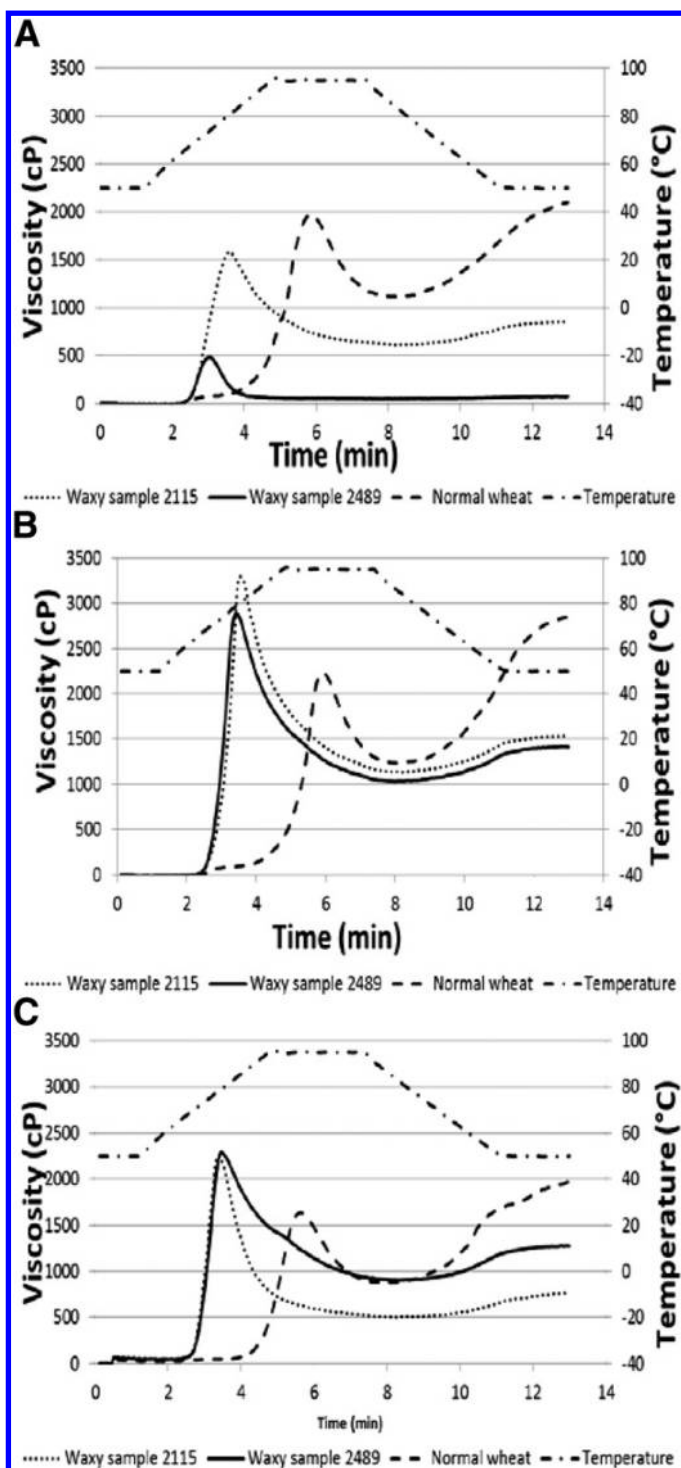


Fig. 1. Rapid Visco Analyzer pasting properties of waxy and normal wheat flours: **A**, in distilled water; **B**, in 1mM silver nitrate solution; and **C**, flours digested with protease and pasted in 1mM silver nitrate.

breakdown in the pasting profiles of the waxy wheat flours (Fig. 1) and starches (Fig. 2) and the aberrant falling numbers of waxy wheat flours observed by Graybosch et al. (2000). Significant swelling of granules contributed to the high peak viscosity of waxy wheat starch (Fig. 2). Excessive swelling and loss of granular integrity probably contribute to the sticky and soft texture of cooked white salted noodles prepared from waxy wheat flours (Baik and Lee 2003).

Silver Nitrate and Protease Solution. To assess the role of protein in flour pasting properties, flour samples were treated with protease and then pasted in AgNO_3 solution (Fig. 1C). Peak viscosities for all flours digested with protease and pasted in AgNO_3 solution were lower than that in AgNO_3 solution without protease treatment. In addition, peak viscosities of waxy wheat flours in protease plus AgNO_3 solution were significantly higher than normal wheat flour. Debet and Gidley (2007) have suggested that starch swelling properties are affected owing to surface protein (likely storage protein) adsorbed onto the surface of starch granules. In cereal flour, the protein matrix could protect the starch granules from mechanical damage during pasting (Hamaker and Griffin 1993). Our results suggest that in flours, protease action could destroy the protein matrix and facilitate mechanical destruction of waxy starch granules during pasting.

When waxy wheat flours were treated with protease and pasted in AgNO_3 solution, their peak viscosities became similar (Table II; Fig. 1C); however, differences in viscosity profiles of waxy wheat flours persisted. To understand the effects of protein on pasting properties, we determined residual protein in the pastes. For instance, ~60% of flour protein in sample 2115 was solubilized by protease in the 30 min digestion period, whereas only 52% of the flour protein was solubilized in sample 2489. The extensive proteolysis could have weakened the interaction between starch granules and protein, making the starch more susceptible to shear thinning. After the protein in flour was essentially removed, the isolated starches from all the waxy wheat starches demonstrated similar pasting properties (Fig. 2).

To confirm that the changes in pasting properties after the protease treatment were because of its action on protein in the flours but not because of any amylase activity in the protease, we used the same level of protease to treat a waxy maize starch at 37°C for 30 min and found that the pasting viscosity of the waxy maize starch was not changed after the protease treatment.

Thermal Properties of Flour. Although gelatinization temperatures for normal and partial waxy wheat flours were slightly (1–2°C) lower than the waxy wheat flours (Table III), an apparent amylose-lipid complex peak was present at ~100°C but

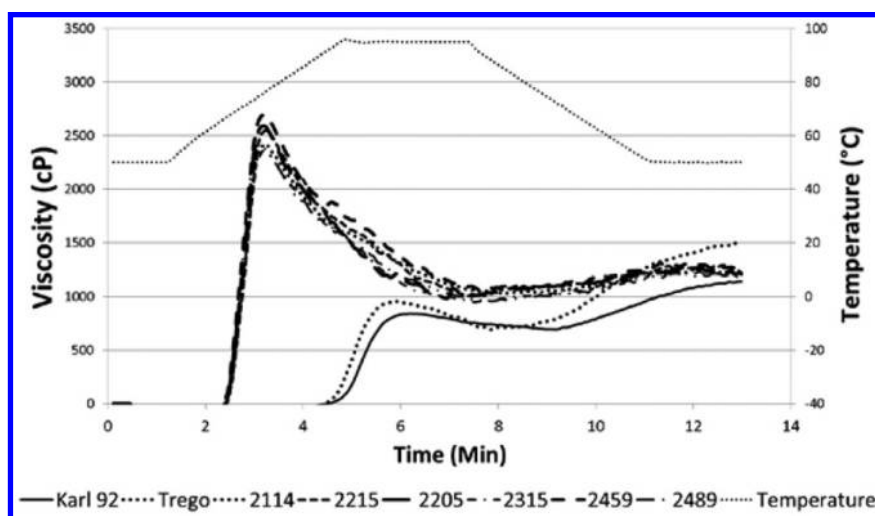


Fig. 2. Rapid Visco Analyzer pasting properties of waxy wheat starch (samples 2114, 2115, 2205, 2315, 2459, and 2489), normal wheat starch (Karl 92), and partial waxy wheat starch (Trego) determined at 7% solids.

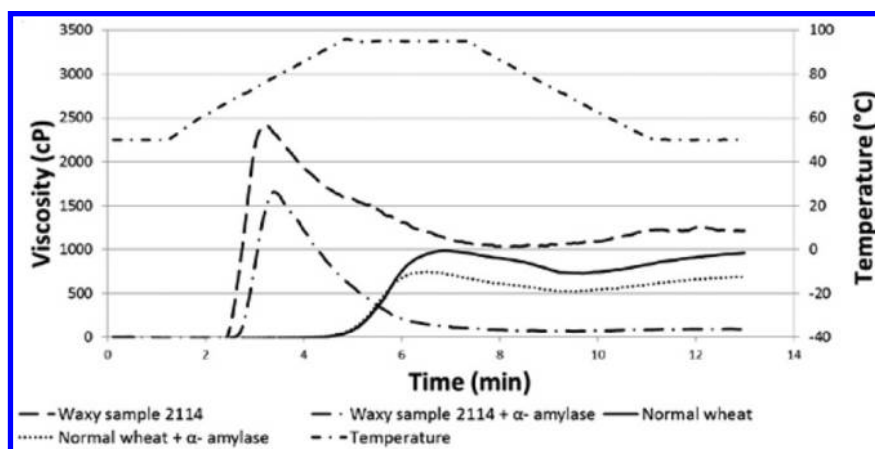


Fig. 3. Rapid Visco Analyzer pasting properties of isolated starches from waxy and normal wheat flours with and without addition of 0.01% α -amylase.

was absent in waxy wheat samples (data not shown). The results help explain the lower pasting peak temperatures of waxy wheat flours compared with normal (Karl 92) or partial waxy (Trego) wheat flour (Fig. 1). A lack of amylose enables waxy wheat flours to swell extensively at their gelatinization temperature and exhibit a peak viscosity at about the same temperature ($\sim 70^{\circ}\text{C}$). In contrast, amylose and the amylose-lipid complex inhibit the swelling of starch granules (Tester and Morrison 1990, 1992; Sasaki and Matsuki 1998) in normal and partial waxy wheat flours. As a result, normal wheat flour had a higher pasting temperature than waxy wheat flours (Fig. 1) even though the gelatinization temperature of the normal wheat flour as determined by DSC was slightly lower (Table III).

CONCLUSIONS

At least two factors (protein matrix and susceptibility to α -amylase activity) contribute to the wide range of pasting properties of waxy wheat flours. Waxy wheat flours are more prone to endogenous α -amylase degradation than normal and partial waxy wheat flours. Isolated waxy wheat starches that no longer contained α -amylase activity had similar pasting properties. The protein matrix may affect the swelling of starch granules and, in turn, cause variations in pasting properties of waxy wheat flours. Waxy wheat starch had lower pasting temperatures but more than 2.5 times higher peak viscosity than the normal wheat starch, and it can be used as a base starch to produce modified starch products with better thickening power.

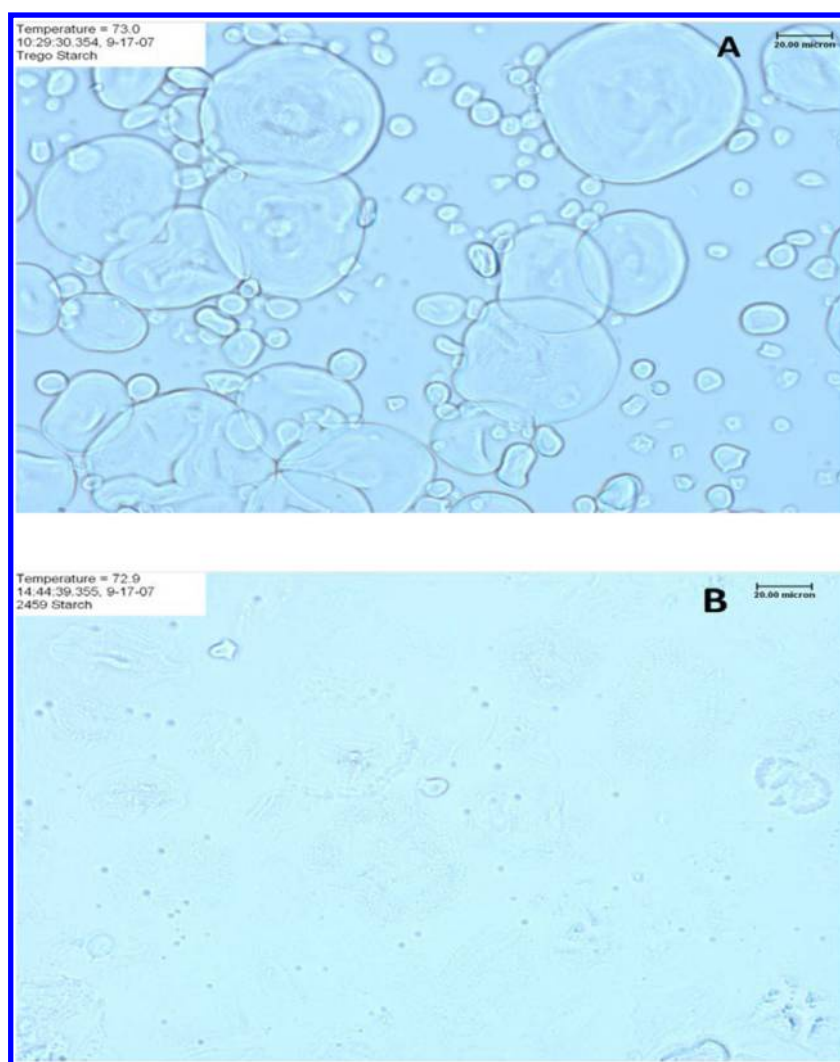


Fig. 4. Granule morphology at 73°C for normal (A) and waxy (B) wheat starches. The bar in the photographs is $20\ \mu\text{m}$.

TABLE III
Gelatinization Properties of Flours (33.3% Solids) Determined by Differential Scanning Calorimeter ($N = 2$)^z

Sample	Onset Temperature ($^{\circ}\text{C}$)	Peak Temperature ($^{\circ}\text{C}$)	End Temperature ($^{\circ}\text{C}$)	Enthalpy (ΔH , J/g)
2114	$61.3 \pm 0.37\text{a}$	$69.3 \pm 0.52\text{a}$	$81.1 \pm 0.78\text{ab}$	$8.0 \pm 0.19\text{a}$
2115	$61.2 \pm 0.49\text{a}$	$69.6 \pm 0.55\text{a}$	$80.8 \pm 0.06\text{bc}$	$8.2 \pm 0.55\text{a}$
2205	$62.0 \pm 0.18\text{ab}$	$70.0 \pm 0.46\text{a}$	$82.6 \pm 0.17\text{ab}$	$8.3 \pm 0.38\text{a}$
2315	$62.0 \pm 0.10\text{ab}$	$70.6 \pm 0.42\text{a}$	$81.7 \pm 0.47\text{ab}$	$6.0 \pm 0.24\text{b}$
2459	$61.8 \pm 0.60\text{a}$	$70.6 \pm 0.69\text{a}$	$84.7 \pm 0.68\text{a}$	$7.6 \pm 0.61\text{ab}$
2489	$61.9 \pm 0.01\text{a}$	$70.0 \pm 0.08\text{a}$	$82.9 \pm 0.88\text{ab}$	$7.1 \pm 0.15\text{ab}$
Karl 92	$60.4 \pm 0.22\text{b}$	$66.1 \pm 0.17\text{b}$	$78.2 \pm 0.00\text{cd}$	$6.2 \pm 0.52\text{b}$
Trego	$60.1 \pm 0.16\text{b}$	$66.1 \pm 0.01\text{b}$	$79.0 \pm 0.40\text{d}$	$6.8 \pm 0.35\text{ab}$

^z Mean \pm SD values are reported. Means not sharing the same letter within each column are significantly different ($P < 0.05$). Full sample identification numbers: NX03Y2114, NX03Y2115, NX03Y2205, NX03Y2315, NWX03Y2459, and NX03Y2489.

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