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Effect of heating temperature on the particle size distribution in waxy wheat flour

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ABSTRACT

The particle size of waxy (amylose-reduced) wheat (*Triticum aestivum* L.) starch was determined at isothermal temperatures by laser diffraction analysis. Flour samples were suspended in deionized water at temperatures ranging from 30 to 90 °C for 20–60 min. At 30 °C, all of the flour particles exhibited trimodal size distributions, i.e., the particles in the first, second, and third modes were <10 μm, 10–50 μm, and 51–300 μm, respectively. Control experiments with isolated starch indicated that the first and second modes were associated mainly with starch granules, whereas the third mode may have been related to gluten and gluten adhesion. The particle size distributions of waxy segregant wheat flours were temperature dependent. At 60 °C, there were significant changes in the particle size and distribution of waxy flours, which indicated the swelling of starch granules in response to elevated temperature. As the temperature increased, the peak particle size of waxy segregant wheat flours increased in different ways. The results suggest that variations in the swelling properties of selected waxy genotype flours may be due to the strength of starch–protein interaction and the capacity for starch granule gelatinization.

1. Introduction

Waxy (amylose-reduced) wheat (*Triticum aestivum* L.) is used extensively in the food industry throughout the world and is produced according to conventional breeding techniques and genetics (Nakamura et al., 1995; Sahlstrom et al., 2003). Waxy wheat carries null (non-functional) alleles in three loci that encode granule-bound starch synthase (GBSS), which is also known as the "waxy protein" (Nakamura et al., 1995). GBSS activates the synthesis of starch amylose, and the amylose levels of wheat genotypes are partly dependent on the number of active alleles that produce GBSS isoforms (Kim et al., 2003). The characteristics of waxy genotype wheat have been investigated, and flours with three sets of chromosome were found to be deficient in one or two of three possible waxy proteins, i.e., *wx-A1*, *wx-B1*, and *wx-D1* (Yamamori and Quynh, 2000). Waxy wheat flours have been reported as sources of improved shelf-life stability, suitable for producing quality baked wheat products, and are substitutes for waxy maize during the production of modified starches (Reddy and Seib, 2000; Lee et al., 2001).

Starch is the primary component of wheat endosperm and is synthesized in the amyloplasts. The amylose content plays an important role in the quality of wheat starch because it can affect the physical properties of processed wheat flour products (Lee et al., 2001; Kim et al., 2003). Wheat with the three GBSS null alleles produces fully waxy starch (WS) with no or little amylose content, whereas the presence of one or two GBSS null alleles results in the production of partial WS with a low amylose content (Graybosch et al., 1998). WS has been isolated and was found to contain 1.2–2.0% apparent amylose compared with 22–25% amylose in wild-type wheat flours (Morrison et al., 1984; Hayakawa et al., 1997). The physicochemical properties of WS have been investigated in the past, including its lower pasting temperature, higher peak viscosity values, and rapid swelling (Hayakawa et al., 1997). The crystalline ratio, thermal transition temperatures, onset of gelatinization, and viscosity of WS differ significantly in waxy genotype and wild-type flours (Kim et al., 2003). Starch containing large (A-type) and small (B-type) granules has a bimodal size distribution pattern (Peng et al., 1999). In addition to the morphology, size, and origin differences, the large and small

Abbreviations: GBSS, granule-bound starch synthase; LDPSA, laser diffraction particle size analyzer; WS, waxy starch.

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granules have been reported to possess distinct properties in terms of starch gelatinization (Geera et al., 2006; Ao and Jane, 2007) and granule swelling (Van Hung and Morita, 2005). The swelling properties of wheat flour during heating have also contributed to the assessment of genetic properties (Rasper and deMan, 1980; Ziegler et al., 1993; Kim et al., 2004).

Many efforts have been made to characterize waxy wheat, but little information has been reported on the swelling properties of waxy wheat flour and WS after heating. Thus, the objective of the present study was to investigate changes in the particle size distributions of waxy genotype wheat flour and WS suspensions after isothermal heating at different temperatures and variable time periods, by using a laser diffraction particle size analyzer (LDPSA) to understand and characterize the effects of GBSS isoforms on the swelling properties of waxy segregants.

2. Materials and methods

2.1. Materials

This study used a set of waxy segregant populations. The cross Kanto107/Bai-Huo/3/Bai-Huo/Kanto107//A92-3327/Kanto107 was used to develop segregant populations, as reported in our previous study (Kim et al., 2003). Forty-four homozygous F5 lines were allocated to eight genotypes based on the absence of one or more isoforms of GBSS. The eight genotype groups were fully waxy (*wx-A1*/*wx-B1*/*wx-D1* null), single null (*wx-A1* null, *wx-B1* null, and *wx-D1* null), double null (*wx-A1/B1* null, *wx-A1/D1* null, and *wx-B1/D1* null), and the wild type. Wheat grains were harvested in Plains, GA (USA), using standard cultural practices. Shriveled seeds were removed from each sample, and the samples were milled using a 30-mesh sieve. Approved method 26–32 was used to evaluate the adjusted flour yield and softness-equivalent (Approved Method Committee, 2000) at the USDA-ARS Soft Wheat Quality Laboratory, Wooster, OH, as reported in a previous study (Kim et al., 2003) (Supplemental Table SI).

2.2. Starch preparation

Starch was extracted from the flour samples using deionized water according to a published method (Knutson and Grove, 1994), which was based on Approved AACC Method 46–30 (Approved Method Committee, 2000). First, 250 g of flour was mixed with 160 ml of isopropanol in a cake mixer at low speed for 2 min. Additional water (340 ml) was added to the slurry, followed by mixing for 3 min. The slurry was transferred to a Waring Blender and blended at high speed for 1 min. The liquid portion was decanted and centrifuged at 1000× *g* for 15 min. The tailing layer and gluten pad were removed from the bottom of the starch layer, and the starch was transferred to a shallow tray and air-dried beneath a hood.

2.3. Protein and amylose content

Approved AACC Method 46–30 was used to determine the flour protein concentration (Approved Method Committee, 2000). The amylose content of the extracted starch was determined according to the method of Knutson and Grove (1994).

2.4. Particle size distribution

The particle sizes were determined using a Malvern LDPSA (Malvern Instruments Ltd, Malvern, England), as reported previously (Kim et al., 2003). The particle size distributions were analyzed using the Mastersizer-s (V 2.18) program (Dengate and Meredith, 1984). The particle sizes were measured for the hydrated and suspended particles after mixing with water for 30 min. Dry samples (30 mg) were weighed in glass tubes with screw-on caps, suspended in 30 ml of deionized water at the desired temperature (30, 40, 50, 60, 70, 80, and 90 °C), covered immediately, and shaken vigorously. The tubes containing the suspensions were transferred to a controlled-temperature water bath and maintained at the specified temperature for 30 min before the experiment. The tubes were vortexed every 10 min throughout that period. The suspension was transferred to the instrument's dispersion circulator tank, which contained deionized water, and fed into the diffraction cells. Sufficient sample was added to yield an obscuration factor of >15%. All of the particle size distributions were measured in triplicate, as reported previously (Kim et al., 2004).

To breakdown aggregated particle components and hydrated flour samples, the starch was placed in a sonication bath (Model FS20, Fisher Scientific, Pittsburgh, PA, USA) at 30 °C for 10–30 min. The WS proteins were different in the waxy segregant flours (Sayaslan et al., 2006), so the WS proteins were measured separately as a control.

2.5. Statistical analysis

The results were analyzed using a general linear model in SAS 9.0 (SAS Institute, Inc., Cary, NC, USA, 2009). $P < 0.05$ was considered statistically significant.

3. Results and discussion

3.1. Protein and amylose content

The mean flour protein and starch amylose contents of the wild-type flour and waxy segregant wheat flours are shown in Table 1. The protein content varied among the waxy segregants, and the wild type had lower protein content than the waxy flour. In general, there were small but significant differences in the protein contents of the waxy segregants and wild type, i.e., the *wx-A1/D1* null, *wx-B1* null, and full waxy flours had significantly higher protein contents than the wild-type, and the full waxy flour had the highest protein content. The average amylose content of full WS was 1.2% and the starch amylose contents of double nulls were significantly lower than those of single nulls and the wild type. *wx-A1/D1* null had a higher amylose content (21.5%) than the other double nulls, whereas *wx-B1/D1* null had the lowest content (17.8%). However, there were no significant differences in the amylose contents of the single null and wild type, which agreed with our previous report (Kim et al., 2003). Similar results were also reported by Yamamori and Quynh (2000), who demonstrated that *wx-B1* null had lower

Table 1
Mean compositions of wheat flour protein and starch amylose contents of wild-type and segregant wheat flours.

Genotype	Flour protein	Starch amylose
	%	
Waxy	11.5a[a]	1.2d
wx-B1/D1 null	11.0b	17.8c
wx-A1/D1 null	11.4a	21.5b
wx-A1/B1 null	11.1b	19.2c
wx-D1 null	11.0b	24.2a
wx-B1 null	11.4a	23.9a
wx-A1 null	11.2b	24.1a
Wild type	11.2b	25.8a
LSD	0.2	2.0

[a] Values with the same letter in the same column do not differ significantly at $P < 0.05$.