

Structure and function of starch from advanced generations of new corn lines

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Abstract

Objectives of this research were to evaluate functions and structures of starches from exotic × adapted inbred lines and exotic lines, to confirm that the functional traits continue into the next generation of inbreeding, and to establish relationships between the fine structure and functional properties of the starches. Several lines were characterized from the successive generations of exotic crosses and exotic inbreds containing kernels with unusual, and potentially useful, thermal properties as measured by differential scanning calorimetry (DSC, gelatinization onset temperature <60 °C or range of gelatinization temperature >14 °C). The frequency of these traits increased with succeeding generations, when selection of the plants was based on the desired trait. Strong correlations were found between DSC and Rapid ViscoAnalyser properties and the granular structure (granular size distribution and branch-chain-length distribution of amylopectin).

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1. Introduction

Corn is extremely important to the US economy because of the amount produced, and its value for domestic and export use. Less than one percent of the US germplasm base, however, consists of exotic germplasm (Goodman, 1985) leading to concerns about the genetic vulnerability of corn. The Latin American Maize Project (LAMP) was the first coordinated international effort to deal with the evaluation of the genetic resources of a major world crop (Salhuana, Pollak, Ferrer, Paratori, & Vivo, 1998). The project, involving the cooperative efforts of 12 countries in evaluating their native germplasm accessions, identified accessions (native and foreign) with good yield potential that could be incorporated into breeding programs. The next step in this overall program, entitled the Germplasm Enhancement of Maize (GEM) project, was to provide the corn industry with materials developed from LAMP by germplasm enhancement. The ultimate objective was to

improve and broaden the germplasm base of corn hybrids grown by American farmers. Traits targeted for improvement are agronomic productivity, disease and insect resistance, and value-added characteristics (Pollak & Salhuana, 1999).

Starch represents nearly 70% of the dry weight of the mature corn kernels and is the most economically important component. Therefore, it was essential to understand the starch characteristics of the GEM materials for their potential to increase the range of characteristics of native corn starch. From previous research, we targeted several GEM and exotic corn lines containing starches with thermal properties significantly different from those of normal corn starches and having potential use to the food industry. In addition to providing new corn lines with value-added properties, the starches from the corn provide an excellent vehicle to study structure–function relationships. The starches from the targeted corn lines showed unusually low gelatinization onset temperature (T_{OG}) and wide range of gelatinization (R_G , Ji et al., 2002). In some starches, two partly overlapping gelatinization transitions were found;

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one had a melting peak at $\sim 66^\circ\text{C}$, and the other at $\sim 69^\circ\text{C}$. The two peaks represented two different populations of starch granules, which gelatinized at different temperatures. All starches exhibited a typical A-type X-ray diffraction pattern. The low T_{OG} was consistent with the branch chain-length pattern of the amylopectin. For example, starches with a lower T_{OG} had a lower normalized concentration of chains with a degree of polymerization (dp) of 15–24 and/or a greater normalized concentration of chains with a dp of 6–12.

Previous work was accomplished by using starches from an early developmental stage, including S_3 and S_4 generations of exotic breeding crosses (exotic populations \times adapted lines), and the S_2 generation for exotic \times adapted inbred lines. The S_n designation defines the number of times the line has been self pollinated, starting with the breeding cross or Plant Introduction (the S_0 population), in the development of the line. The frequency of kernels within some lines containing starch with low T_{OG} was relatively low (1/10), which indicated that the genetic background controlling low T_{OG} was still heterogeneous. Therefore, all thermal and structural analyses were performed by using starch extracted from single kernels. By advancing inbreeding by self-pollination and selecting for a specific trait (low T_{OG}), this specific trait was fixed in the progeny generation. Furthermore, seed supply was increased to allow us to do more complete structural and functional analyses.

The objectives of this research were to evaluate functions and structures of starches from advanced generations of developmental lines developed from exotic inbred lines and exotic breeding crosses (exotic populations \times adapted lines), and to establish relationships between the fine structure and functional properties of the starches.

A secondary objective was to confirm that selected functional traits advanced into the next generation of corn upon inbreeding.

2. Materials and methods

2.1. Corn populations

Lines and their derivatives from three exotic by adapted breeding crosses from the GEM project and four exotic inbreds, plus public inbred lines B73 (Stiff Stalk heterotic pattern) and Mo17 (non-Stiff Stalk heterotic pattern) as controls, were studied (Table 1). The original exotic lines and populations used in this study are maintained at the North Central Regional Plant Introduction Station in Ames, IA. The breeding crosses (S_0) were developed by crossing the exotic populations (DK212T is a tropical three-way commercial hybrid developed by DeKalb Genetics in Thailand) with inbreds of the Stiff-Stalk heterotic pattern. The Stiff Stalk inbreds belong to companies that cooperate in GEM. All parent lines designated for advancing by self-pollination were selected for the desirable trait, low T_{OG} . All S_4 progeny derived from the exotic \times adapted lines, CHIS-37, CUBA-38, and for the Mo-17 control, were grown and self-pollinated in the same environment near Ames, IA in 1998. All S_5 progeny derived from the exotic \times adapted lines, DK-8-1, DK-8-4, DK-8-5, DK-10-1, and DK-10-25, S_2 progeny for the exotic inbreds, PI82-3, PI82-6, PI82-8 and PI82-18, and the B73 control, were grown and self-pollinated in the same environment near Ames, IA in 1999. Ears were harvested at full physiological maturity; dried at

Table 1
Exotic breeding crosses and exotic inbred corn lines and their origins

Exotic parent ^a	Pedigree for S_n lines ^b	Source identification ^c	Number of S_{n+1} lines analyzed ^d	Number of unusual S_{n+1} lines ^e	Origin of exotic parent
<i>Exotic bending crosses</i>					
PI 576258	CHIS 775:S1911b-37-1-2	CHIS-37	16	9	Mexico
PI 489361	CUBA164:S1511b-38-1-3	CUBA-38	8	2	Cuba
Ames 23670	DK212T:S0610-8-1-3-1	DK-8-1	1	1	Thailand
Ames 23670	DK212T:S0610-8-1-3-4	DK-8-4	6	6	Thailand
Ames 23670	DK212T:S0610-8-1-3-5	DK-8-5	3	3	Thailand
Ames 23670	DK212T:S0610-10-1-3-1	DK-10-1	4	4	Thailand
Ames 23670	DK212T:S0610-10-1-3-25	DK-10-25	4	2	Thailand
<i>Exotic inbreds</i>					
PI 186182	PI 186182-3	PI82-3	4	4	Uruguay
PI 186182	PI 186182-6	PI82-6	3	3	Uruguay
PI 186182	PI 186182-8	PI82-8	4	4	Uruguay
PI 186182	PI 186182-18	PI82-18	2	2	Uruguay

^a Original corn populations as maintained at the North Central Region Plant Introduction Center, Ames, IA.

^b Corn lines, selected for a specific starch characteristic. $n = 4$ for exotic breeding crosses CHIS-37 and CUBA-38. $n = 5$ for exotic breeding crosses DK-8-1, DK-8-4, DK-8-5, DK-10-1 and DK-10-25. $n = 2$ for exotic inbreds.

^c Abbreviated source identification for use in this paper.

^d S_{n+1} line means first generation of corn after self-pollination of S_n .

^e Number of S_{n+1} lines having at least one kernel containing starch with unusual thermal properties according to criteria given by Seetharaman et al. (2001).

38 °C for five days, shelled and stored at 4 °C and 45% relative humidity until the kernels were needed for analysis.

2.2. Single kernel starch extraction

Starch was extracted from single kernels using the method described by White, Abbas, Pollak, and Johnson (1990), with modifications (Krieger, Duvick, Pollak, & White, 1997). Each kernel of whole corn was steeped in 5 ml 1% sodium metabisulfite solution at 45 °C for 48 h, followed by manual removal of the pericarp and germ with forceps. The separated endosperm was placed in a 50 ml centrifuge tube with 10 ml of distilled water and homogenized by using a Tekmar tissue homogenizer (Ultra-Turrax T25, 600W, Cincinnati, OH) at 20,500 rpm for 30 s. The homogenized slurry was filtered by using a 30 µm nylon filter under vacuum with several washes, for a total wash-water volume of 300 ml. The starch slurry was allowed to settle in a refrigerator for 2 h and the supernatant drained. The starch was rinsed with 250 ml water, drained twice, and the resulting sediment air-dried.

For the initial screening, at least five randomly selected kernels from each of the 55 S_{n+1} lines were individually evaluated for starch characteristics after extraction. Thermal analysis by using Differential Scanning Calorimetry (DSC, described below) was conducted on these starch samples. Based on the results of the initial screening, 11 progeny lines from 11 exotic families, plus one line each from Mo17 and B73 as controls, were selected for further characterization.

2.3. Bulk starch extraction

Five to ten (depending on the availability of seeds) separate starch extractions, of ten kernels each, were made by using the ten-kernel extraction procedure as for screening, except that the homogenized slurry was filtered by using a nylon filter for a total wash-water volume of 500 ml. Starches were combined and mixed well to produce the large quantities of starch needed for the functional and structural studies described in this paper. After extraction, starch was stored at 4 °C until evaluated.

2.4. Moisture content of starch

Moisture content of starch was measured by heating starch at 110 °C for 3 h. The weight differences were used to calculate moisture content. Triplicate analyses were done for each sample.

2.5. DSC

Thermal properties of the isolated starch were analyzed with a DSC7 analyzer (Perkin–Elmer Corp.,

Norwalk, CT) equipped with a thermal analysis data station. A starch sample (about 4 mg starch, dry-weight basis) was carefully weighed in an aluminum DSC pan and 8 µl water was added. After sealing the pan, the starch was gelatinized as described elsewhere (White et al., 1990). All experiments were run at a scanning rate of 10 °C/min from 30 to 110 °C (Ng, Duvick, & White, 1997). DSC parameters recorded for this study included change in enthalpy (ΔH), peak onset temperature (T_o), peak temperature (T_p), and range of gelatinization temperature (R_G). A subscript G after the parameter denotes a gelatinization property. The parameters, T_o , T_p , T_c (peak conclusion temperature), and ΔH were given directly by the DSC software. The R_G was calculated as $T_c - T_o$, and peak height index (PHI) was calculated from the change in enthalpy of gelatinization divided by half the range. The same scanning method was used for measuring retrogradation of the gelatinized samples kept at 4 °C for 7 days. A subscript R after the DSC parameter denotes a retrogradation property. The T_o , T_p , T_c and ΔH of retrogradation were calculated automatically. Range of retrogradation (R_r) was calculated as $(T_c - T_o)$. All enthalpy calculations were based on the dry-starch weight, and all analyses were conducted in duplicate and the values averaged. Percentage of retrogradation (%R) was calculated from the ratio of ΔH of retrogradation to ΔH of gelatinization.

2.6. Pasting properties

The pasting properties of starches were analyzed by using a Rapid Visco Analyzer (RVA, model 4, Newport Scientific Pty. Ltd, Warriewood, NSW, Australia). At least two RVA profiles were obtained for each sample and the results for each sample were averaged. An 8% (dwb) starch in water slurry at a final weight of 28 g was used for each RVA analysis (Jane et al., 1999). Pasting temperature (P_{temp}), peak time (P_{temp}), peak viscosity (PV), trough or hot paste viscosity (HPV), final or cool paste viscosity (CPV), breakdown (PV-HPV), and set-back (CPV-HPV) were recorded.

2.7. Gel strength

The texture properties of starch pastes from the RVA analysis were analyzed with a texture analyzer (Stable Micro Systems TA.XT2, Texture Technologies Corp., Scarsdale, NY) by the procedure of Takahashi and Seib (1988). Gel strength was tested after two storage conditions: one day at 25 °C and 7 days at 4 °C. Samples stored at 4 °C for 7 days were equilibrated at room temperature for 1 h before analysis. From each of two replicates, gel strength was measured at five different locations on each gel sample and the results per replicate were averaged.

2.8. Microscopy analyses

Granule-size distributions of native starches were obtained by following the procedure described by Jane and Chen (1992). Three slides from each sample were analyzed separately, with 400 particles measured from each slide, to give a total of 1200 starch granules analyzed per starch type. The starch granules selected for the measurement were randomly chosen from each slide. For each granule, cross sectional area (area), perimeter, radial S.D., and major axis were determined. The radial S.D. is a measure of the 'roundness' of a particle. A perfect circle would have a radial S.D. measurement of 0. The less round the particle, the bigger its radial S.D. number. The equivalent diameter was assessed by: equivalent diameter = $\sqrt{4\text{area}/\pi}$.

2.9. Apparent amylose content

Starch samples were defatted by the method of Lim, Kasemsuwan and Jane (1994). Iodine affinities of defatted whole starch were determined by using a potentiometric autotitrator (702 SM Titrino, Brinkman Instrument, Westbury, NY) following the procedure of Kasemsuwan, Jane, Schnable, Stinard and Robertson (1995). Apparent amylose contents were calculated by dividing the iodine affinity of starch by 19.0% (Takeda and Hizukuri, 1987). Iodine affinity measurements of the samples were replicated four times and the results averaged.

2.10. Branch-chain-length distribution of whole starch

Branch-chain-length distribution of starches was determined following the procedure described by Jane and Chen (1992). Starch was debranched by using isoamylase, and the branch-chain-length distributions were analyzed by using a high-performance anion-exchange chromatography system equipped with an enzyme column reactor and a pulsed amperometric detector (Dionex, Sunnyvale, CA) (HPAEC-ENZ-PAD) by using the method reported by Wong and Jane (1997). A CarboPac PA1 anionexchange column ($250 \times 4 \text{ mm}^2$) and a CarboPac PA1 guard column ($25 \times 3 \text{ mm}^2$) were used for sample separation. The results reported are an average of at least two replicates for each sample.

2.11. Molecular weight distribution of amylopectin by HPSEC-MALLS-RI

The weight-average molecular weight (M_w) and z-average radius (R_z) of gyration of amylopectin were measured by using HPSEC (HP1050 series isocratic pump) equipped with multiangle laser lightscattering (MALLS, model Dawn-F, Wyatt Tech. Co., Santa Barbara, CA) and refractive index (RI) detectors (HP1047A) following the procedure of Yoo and

Jane (2002). In the procedure, a Shodex OHpak KB-G guard column, and KB-806 and KB-804 analytical columns (Shodex Denko, Tokyo, Japan) were used for the separation of amylopectin from amylose.

2.12. Statistical analysis

Between-sample variations of granule size and shape parameters, which included mean area, equivalent diameter, perimeter, radial S.D., and major axis, were assessed by using the analysis of variance (ANOVA) for mixed effects model with nested design (i.e. three plates were nested within a starch sample). The Tukey multiple comparison test was used to calculate differences in means of these parameters among starch samples. The apparent amylose content, molecular weight of amylopectin, starch paste properties, and starch texture properties were analyzed by using the general linear model (GLM) procedure. Multiple comparison procedures of the Tukey test were used to calculate the differences among starch samples. The family-wise confidence level used for calculating the differences among starch samples was 95% (i.e. $\alpha = 5\%$). Relationships between starch functional properties and structural properties were analyzed by using the Pearson correlation test. Calculations were performed by using SAS version 8.2 (SAS Institute, Cary, NC) for the Unix Operating system.

3. Results and discussion

3.1. Selection and verification of trait by Using DSC

Starches from 55 S_{n+1} lines (Table 1) were screened for unusual thermal properties by using DSC. Among these 55 S_{n+1} lines, 24 exotic breeding cross-lines were from the S_4 generation (CHIS-37 and CUBA-38), 18 exotic crosses were from the S_5 generation (DK-8-1, DK-8-4, DK-8-5, DK10-1 and DK10-25), and 13 exotic inbreds were from the S_3 generation (PI82-3, PI82-6, PI82-8, PI82-18). Five to 13 randomly selected single kernels per line were individually extracted, and the starch was then analyzed by using DSC. Forty out of 55 lines were identified to have at least one kernel out of 10 whose starch exhibited thermal properties that are significantly different from that of normal Corn Belt lines (Mo17 and B73), such as a low T_{oG} and wide R_G . For example, 9 out of 16 progeny lines of CHIS-37 contained at least one kernel having starches with unusual gelatinization properties.

Based on the screening results (represented in Table 1), 16 S_{n+1} lines containing kernels with unusual thermal properties ($T_{oG} < 60^\circ\text{C}$ or $R_G > 14^\circ\text{C}$) were selected for further characterization (Table 2). The frequency of kernels within each S_{n+1} line with starch exhibiting the specific DSC properties varied from 1/10 to 10/10. Starch gelatinization properties of individual kernels within an S_{n+1} line exhibited

Table 2

Differential scanning calorimetry (DSC) data of starches from single kernels of selected corn S_{n+1} lines^a

Corn source ^b	Frequency ^c	T_{oG} ^a (°C) ^d			T_{pG} (°C)			T_{cG} (°C)			ΔH_G (J/g)		
		Mean	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
Mo17	0/18	66.2	65.2	67.3	70.5	69.7	71.2	75.8	74.5	76.3	12.5	12.1	12.9
B73	0/10	64.9	64.5	65.3	68.7	68.2	69.3	72.8	71.9	73.8	10.9	10.4	11.2
CUBA-38													
CUBA-38-5	8/10	59.7	56.9	62.1	66.4	64.4	67.8	72.7	71.7	74.2	9.4	9.0	9.7
CHIS-37													
CHIS-37-5	5/13	62.2	59.2	63.9	67.9	66.3	69.4	74.9	74.1	75.5	10.8	10.3	11.5
DK-8-1													
DK-8-1	4/5	60.3	59.0	63.2	68.1	67.5	69.7	73.6	72.6	74.7	11.4	11.1	11.7
DK-8-4													
DK-8-4-3	10/10	57.1	52.8	59.1	67.6	66.9	68.5	73.0	72.3	74.2	11.7	9.5	12.9
DK-8-4-5	10/10	59.2	55.7	60.6	68.2	67.3	68.6	73.1	73.7	72.3	12.1	11.5	12.8
DK-8-4-6	10/10	57.5	55.1	59.9	67.3	66.8	68.2	72.5	71.9	73.5	11.2	9.6	12.7
DK-8-4-8	10/10	58.5	55.7	60.6	67.7	67.0	68.3	73.2	72.2	74.4	11.0	9.7	12.2
DK-8-5													
DK-8-5-1	9/10	58.4	54.8	62.8	68.6	67.6	69.7	73.6	72.7	74.9	10.8	10.5	11.8
DK-8-5-2	9/10	57.0	51.7	65.1	68.3	67.8	68.8	73.4	72.3	74.3	10.9	9.6	12.1
DK-10-1													
DK-10-1-3	9/10	60.1	58.3	61.7	69.1	68.0	69.8	74.0	73.4	74.8	11.2	10.7	11.9
DK-10-1-4	7/10	60.2	58.5	61.7	69.1	68.2	69.7	73.8	72.7	74.4	10.8	9.9	11.7
DK-10-25													
DK-10-25-1	9/10	59.4	57.2	61.5	67.4	67.0	67.7	71.9	70.8	72.7	10.6	9.2	11.4
PI82-3													
PI82-3-1	8/10	60.0	57.4	65.1	68.4	67.3	69.0	73.0	71.6	73.5	10.8	10.1	11.2
PI82-6													
PI82-6-1	5/10	60.4	58.6	62.4	67.1	66.0	67.9	73.2	72.5	74.1	9.4	8.6	10.6
PI82-8													
PI82-8-5	7/10	60.6	57.9	64.4	67.8	66.4	69.0	73.2	72.3	73.8	11.0	10.1	11.9
PI82-18													
PI82-18-1	1/10	62.0	59.5	64.3	68.8	67.3	70.0	74.1	73.0	75.4	11.1	10.8	11.3

^a See Table 1 for the definition of S_{n+1} lines.^b See Table 1 for an explanation of source identification. The number after the dash signifies the ear number for the pedigree of the S_n line.^c Frequency of kernels whose starch exhibited the unusual DSC properties, $T_{oG} \leq 61$ °C or $R_G \geq 14$ °C.^d T_{oG} , Gelatinization onset temperature; T_{pG} , Gelatinization peak temperature; T_{cG} , Gelatinization end temperature ΔH_G , Enthalpy of gelatinization. The results reported are an average of at least two replicates for each sample, and standard deviations are less than 0.38.

considerable variability. The mean, minimum and maximum values for T_{oG} , T_{pG} , T_{cG} and ΔH_G of all kernels analyzed within a progeny line are noted (Table 2). These selected lines have the potential for developing inbred lines with fixed unusual thermal properties. None of the starches exhibited unusual retrogradation properties as measured by DSC.

To obtain large quantities of starch for further functional and structural analyses, all corn lines listed in Table 2 underwent starch extraction by using a bulk extraction procedure for 50–100 kernels depending on the availability of the seeds. The DSC properties of each starch type, bulked for each sample, are listed in Table 3. The values are the average of at least two DSC analyses of the bulked starch.

Some starches from exotic lines also exhibited gelatinization thermogram shapes with shoulders (Fig. 1), suggesting two independent cooperative transitions located in different populations of starch granules starting to gelatinize at different temperatures (Ji et al., 2002). This finding verifies the presence of the two populations first

noted in the previous paper, and demonstrates that their presence is genetic.

3.2. Pasting and textural properties of starch from selected corn lines

Significant differences were observed in the pasting properties of starch from different lines (Table 4). The B73 starch had the greatest P_{temp} (79.2 °C), whereas DK-8-4-5 had the lowest P_{temp} (71.1 °C). Among all exotic lines, DK-8-4-6 starch had the greatest PV (222.7 RVU), DK-10-1-4 starch had the greatest breakdown (114.9 RVU) and PI82-6-1 starch had the greatest setback value (108.5 RVU). The values for starch from the two Corn Belt inbred lines, B73 and Mo17, were quite different from each other in their pasting properties. B73 starch exhibited relatively high PV values (195.2 RVU), high breakdown (77.5 RVU) and high setback values (97.2 RVU), compared with Mo17 starch.

Data for the firmness and stickiness of the gels measured with a TA after the two storage treatments are summarized

Table 3
3 DSC data of starches from 50 to 100-kernel extractions of selected corn lines

Source of starch (S_{n+1} line ^a)	Native starch				Retrograded starch					
	T_{oG} (°C)	T_{pG} (°C)	T_{eG} (°C)	ΔH_G (J/g)	R_G (°C)	PHI	T_{or} (°C)	R_r (°C)	ΔH_r (J/g)	$r\%$
Mo17	67.7	72.0	76.5	13.0	8.6	3.0	43.3	19.3	6.9	52.8
B73	65.0	69.8	75.0	11.6	10.1	2.3	43.6	19.0	6.4	54.9
CUBA-38										
CUBA-38-5	61.2	68.1	74.3	11.2	13.0	1.7	42.1	20.8	6.4	57.1
CHIS-37										
CHIS-37-5	61.4	68.6	76.2	11.5	14.9	1.5	42.5	19.6	5.6	48.8
DK-8-1										
DK-8-1	58.7	68.7	74.6	12.4	15.9	1.6	42.3	19.7	6.2	50.2
DK-8-4										
DK-8-4-3	56.2	68.2	73.8	11.1	17.6	1.3	42.2	19.5	4.6	42.0
DK-8-4-5	60.0	67.7	72.6	12.2	12.6	1.9	40.4	21.1	6.2	51.3
DK-8-4-6	56.7	67.7	73.5	12.1	16.8	1.4	41.0	20.6	5.6	46.6
DK-8-4-8	57.2	68.0	73.8	12.1	16.6	1.3	42.5	18.7	5.4	49.0
DK-8-5										
DK-8-5-1	55.3	68.8	74.9	11.3	19.7	1.1	41.6	20.7	5.8	51.6
DK-8-5-2	53.5	69.3	75.5	12.1	22.1	1.1	41.2	21.3	6.2	51.0
DK-10-1										
DK-10-1-3	60.3	69.5	74.8	12.3	14.5	1.7	42.7	18.7	6.7	54.4
DK-10-1-4	62.2	69.2	74.2	11.6	12.0	1.9	41.1	21.6	6.4	55.5
DK-10-25										
DK-10-25-1	56.7	67.3	72.0	11.6	15.3	1.5	39.7	22.0	6.5	56.6
PI82-3										
PI82-3-1	61.4	68.8	74.1	10.6	12.7	1.7	43.5	18.6	5.2	49.4
PI82-6										
PI82-6-1	59.7	67.8	74.2	10.8	14.5	1.5	42.4	19.8	6.5	59.8
PI82-8										
PI82-8-5	59.2	67.3	73.7	11.4	14.4	1.6	40.9	20.5	6.2	54.6
PI82-18	62.0	69.8	75.9	12.2	13.8	1.8	42.9	19.4	5.5	45.0
PI82-18-1	62.0	69.8	75.9	12.2	13.8	1.8	42.9	19.4	5.5	45.0

T_{oG} , gelatinization onset temperature; T_{pG} , gelatinization peak temperature; T_{eG} , Gelatinization end temperature; ΔH_G , enthalpy of gelatinization; R_G , range of gelatinization temperature; PHI, peak height index (enthalpy of gelatinization divided by half the range); T_{or} , retrogradation onset temperature; R_r , range of retrogradation temperature; ΔH_r , enthalpy of retrogradation; $r\%$, percentage of retrogradation. The results reported are an average of at least two replicates for each sample, and standard deviations are less than 0.41.

^a See Table 1 for an explanation of S_{n+1} line.

in Table 5. For each starch type, 7 days of storage at 4 °C gave significantly firmer gels than did 1 day of storage at 25 °C (significance data for this comparison is not listed in Table 5). The increase in gel firmness over time is mainly caused by retrogradation of starch gels, which is associated with syneresis of water and crystallization of amylopectin, leading to harder gels (Miles, Morris, Orford, & Ring, 1985).

Among the different lines, significant differences were observed in the textural properties. Starch from B73, CHIS-37-5 and PI82-18-1 required the least amount of force to break the gel among all starch types after both 1 and 7 days storage, and starch from DK-8-4-6 required the greatest amount of force to break the gel among all starch types after both one and seven days storage. Gel of DK-8-4-5 starch had the greatest stickiness value (21.99 g s) after 1 day of storage at 25 °C, whereas gel of PI82-6-1 starch had the greatest stickiness value (27.57 g s) after seven days of storage at 4 °C. Gel of DK-10-1-4 starch had the least stickiness value

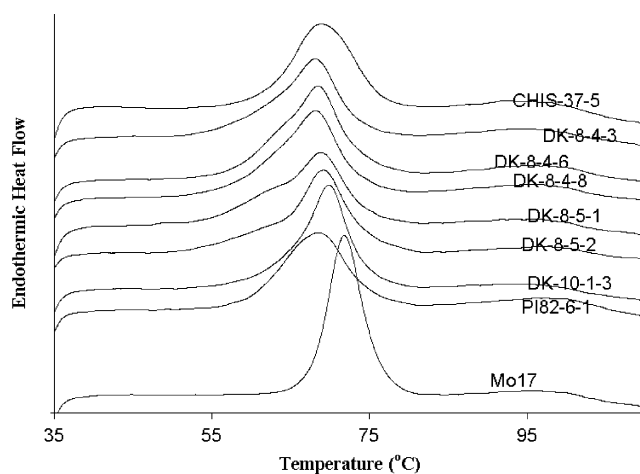


Fig. 1. Differential scanning calorimetry thermographs of gelatinization of the starches in Table 3.

Table 4
Pasting properties of starches from selected corn lines measured with a rapid viscoanalyser (RVA)

Source of starch (S_{n+1} line ^a)	P_{temp}	P_{time}	Viscosity (RVU)				
			PV	HPV	CPV	Breakdown	Setback
Mo17	75.5	8.2	152.4	104.0	181.1	48.4	77.1
B73	79.2	8.3	195.2	117.7	214.9	77.5	97.2
CUBA-38							
CUBA-38-5	71.6	8.1	198.3	105.7	202.2	92.6	96.5
CHIS-37							
CHIS-37-5	72.3	8.3	210.0	136	230.6	74.0	94.6
DK-8-1							
DK-8-1	72.9	8.1	204.9	131.3	220.1	73.6	88.8
DK-8-4							
DK-8-4-3	72.1	8.3	184.0	111.6	209.9	72.4	98.3
DK-8-4-5	71.1	8.0	198.4	117	212.9	81.4	95.9
DK-8-4-6	71.2	8.2	222.7	140.1	232.4	82.6	92.3
DK-8-4-8	75.3	8.2	195.9	123.8	215.2	72.1	91.4
DK-8-5							
DK-8-5-1	73.4	8.3	200.3	123.4	221.3	76.9	97.9
DK-8-5-2	77.9	8.5	186.1	128.8	216.8	57.3	88.0
DK-10-1							
DK-10-1-3	72.7	8.1	205.7	119.3	208.8	86.4	89.5
DK-10-1-4	73.9	7.7	222.5	107.6	211.3	114.9	103.7
DK-10-25							
DK-10-25-1	77.1	8.4	204.2	122.4	229.2	81.8	106.8
PI82-3							
PI82-3-1	77.7	8.1	191.2	106.1	202.4	85.1	96.3
PI82-6							
PI82-6-1	71.9	8.0	207.2	105.4	213.9	101.8	108.5
PI82-8							
PI82-8-5	77.7	8.3	208.4	131.8	233.9	76.6	102.1
PI82-18							
PI82-18-1	78.1	8.3	192.8	127.7	213.9	65.1	86.2
S.D.	1.78	0.13	7.66	7.15	12.00	8.82	9.01

P_{temp} , pasting temperature (°C), P_{time} , peak time (min), PV, peak viscosity, HPV, hot paste viscosity, CPV, cool paste viscosity, breakdown, PV – HPV, setback, CPV – HPV. S.D., standard deviations. The results reported are an average of at least two replicates for each sample.

^a See Table 1 for an explanation of S_{n+1} line.

(14.12 g s) after 1 day of storage at 25 °C, whereas gel of DK-8-4-3 starch had the least stickiness value (18.8 g s) after 7 days of storage at 4 °C. Textural properties of starch gels are very important criteria used to evaluate the performance of starch in a food system. For example, a firm and short (low stickiness) gel is desirable in a pudding or salve-like product, whereas long (great stickiness) gel is desirable in syrup-like products (Thomas & Atwell, 1999b). The variability of the gel properties of starches obtained from these developmental lines makes them suitable to be used in a wide variety of foods.

3.3. Apparent amylose content

No significant differences were observed in the apparent amylose content among the starches analyzed. The average apparent amylose content for all starches ranged from 29.0 to 32.4% (data not shown). Because of the limited quantity

of starch available from each line, we did not measure the absolute amylose content of starch. It is difficult to make any conclusion about absolute amylose content from apparent amylose content, because long branch chains of amylopectin, like amylose, could bind iodine to form a single helical complex during potentiometric titration, and consequently inflate the iodine affinity and the apparent amylose content of starch (Jane et al., 1999; Kasemsuwan et al., 1995).

3.4. Granule-size distribution

Significant differences were observed in the mean granule-size between the selected starches (Table 6). Granules of starch from DK-8-5-2 had the smallest mean area, equivalent diameter, and major axis, whereas granules of starch from B73 had the greatest values for those parameters. The mean granule-shape parameters also showed significant differences. Granules from B73 starch tended to deviate least from a spherical shape

Table 5

Gel properties of starches from selected corn lines measured with a texture analyzer

Source of Starch (S_{n+1} line ^a)	Firmness (g)		Stickiness (g s)	
	25 °C, 1 Day	4 °C, 7 Days	25 °C, 1 Day	4 °C, 7 Days
Mo17	14.86ab	19.20ed	18.53bd	24.43a–e
B73	10.46ij	16.44fg	14.40g	22.52a–f
CUBA-38				
CUBA-38-5	12.54d–g	20.46b–d	20.53ab	21.25c–f
CHIS-37				
CHIS-37-5	10.26j	16.40fg	20.33ab	20.44
DK-8-1				
DK-8-1	12.12e–h	20.31b–d	21.87a	26.68ab
DK-8-4				
DK-8-4-3	10.77h–j	14.28h	18.26be	18.8f
DK-8-4-5	13.51b–e	21.67ab	21.99a	18.94f
DK-8-4-6	16.24a	23.42a	18.90bc	21.87b–f
DK-8-4-8	13.39b–e	17.68fe	20.62ab	20.58d–f
DK-8-5				
DK-8-5-1	12.42d–g	16.39fg	20.09ab	19.68ef
DK-8-5-2	11.85f–i	17.36fg	17.42cf	25.46a–d
DK-10-1				
DK-10-1-3	15.3a	21.18bc	15.74eg	22.41a–f
DK-10-1-4	13.63b–d	21.21bc	14.12g	27.14a–c
DK-10-25				
DK-10-25-1	14.10a–c	NA ^b	15.44fg	NA
PI82-3				
PI82-3-1	11.17g–j	15.53hg	18.94bc	20.50d–f
PI82-6				
PI82-6-1	12.27d–h	19.65cd	20.17ab	27.57a
PI82-8				
PI82-8-5	13.09c–f	18.65ed	20.22ab	24.47a–e
PI82-18				
PI82-18-1	10.03j	17.36fg	16.08dg	20.06

The results reported are an average of at least five replicates for each sample. Standard deviations are not listed, because of the unbalanced data. Values followed by the same letter in the same column are not significantly different ($P < 0.05$).

^a See Table 1 for an explanation of S_{n+1} line.

^b Not available. Data is missing because of the limited quantities.

(radial S.D. = 7.7), whereas granules from CUBA-38-5 starch tended to deviate most from a spherical shape (radial S.D. = 9.4).

Starch granules were assigned to five groups according to their equivalent diameters: <5 , ≥ 5 and <9 , ≥ 9 and <13 , ≥ 13 and <17 , and ≥ 17 μm , and reported as a percentage of the total number of granules measured (Table 7). Significant differences were observed in the percentage distribution profiles of some of these selected starches. In general, B73 had the smallest proportion of granules smaller than 5 μm in diameter (0.92%), whereas Mo17 had the greatest proportion of granules smaller than 5 μm in diameter (12.0%). Also, DK-10-1-3 tended to have the smallest proportion of large granules (0.25% of granules ≥ 17 μm), and B73 had the greatest proportion of large granules (8.17% of granules ≥ 7 μm).

Table 6

Mean granule size and shape parameters of starches from selected corn lines used in structural analyses

Source of starch (S_{n+1} line ^a)	Mean area (μm^2)	Mean diameter (μm)	Mean perimeter (μm)	Mean major axis (μm)	Mean radial S.D. (μm)
Mo17	88.9D	9.8D	41.1C	10.8D	8.8A–C
B73	123.6A	12.0A	49.9A	13.1A	7.7F
CUBA-38					
CUBA-38-5	111.9B	11.4A	48.3A	12.6A	9.4A
CHIS-37					
CHIS-37-5	91.3CD	10.4C	43.4C	11.3C	7.9EF
DK-8-1					
DK-8-1	85.1D	9.9D	41.2C	10.9D	8.1D–F
DK-8-4					
DK-8-4-3	69.1E	8.9F	36.8F	9.6E	8.2D–F
DK-8-4-5	83.7D	9.9D	41.6C	10.8D	8.2DE
DK-8-4-6	86.7D	10.0CD	41.9C	10.9CD	7.8EF
DK-8-4-8	74.2E	9.3EF	38.7DE	10.1E	7.9EF
DK-8-5					
DK-8-5-1	85.7D	10.0D	41.6C	10.9CD	8.5CD
DK-8-5-2	70.7E	9.0F	37.5EF	9.8E	8.3DE
DK-10-1					
DK-10-1-3	71.3E	9.1F	37.9EF	9.9E	8.5CD
DK-10-1-4	94.4C	10.5C	43.9B	11.4C	8.3DE
DK-10-25					
DK-10-25-1	81.9D	9.7DE	40.4CD	10.7D	8.9AC
PI82-3					
PI82-3-1	83.2D	9.9D	41.8C	10.9CD	9.3A
PI82-6					
PI82-6-1	105.7B	10.9B	45.7B	11.9B	8.6BD
PI82-8					
PI82-8-5	83.5D	9.8D	41.2C	10.7D	8.9A–C
PI82-18					
PI82-18-1	85.6D	9.8D	41.2C	10.7D	9.1AB

The results reported are an average of 1200 granules from three slides for each sample. Values followed by the same letter in the same column are not significantly different ($P < 0.05$).

^a See Table 1 for an explanation of S_{n+1} lines.

Similar to results previously noted (Ji et al., 2002), Mo17 and B73 were significantly different from each other in granular size and shape distributions, granules from B73 were larger and more spherical than granules from Mo17. In addition, B73 starch had a lower percentage distribution of small granules (<9 μm) and greater percentage distribution of large granules (≥ 9 μm) than did Mo17.

3.5. Branch-chain-length distribution of amylopectin

Normalized branch-chain-length distributions of the starches were measured by using HPAEC-ENZ-PAD. The comparison of normalized chain length distributions of three selected exotic starches with starch from control line, B73, are shown in Fig. 2a–c. The results are expressed as means \pm standard deviation of a minimum of duplicate analyses, and the height of each bar at each degree of polymerization (dp) represents its relative concentration to peak I, located at dp 13–14. All

Table 7
Distribution profiles of starch from selected corn lines

Source of starch (S_{n+1} line ^a)	Distribution profiles (%) ^b				
	< 5 (μm)	5–9 (μm)	9–13 (μm)	13–17 (μm)	≥ 17 (μm)
Mo17	12a	35.2c–e	29e	18.3c–e	5.5a–c
B73	0.92f	21.33f	37.92b–e	31.67a	8.17a
CUBA-38					
CUBA-38-5	4.23b–f	22.25f	38.14b–e	31.06ab	4.32b–d
CHIS-37					
CHIS-37-5	2.3ef	30.8d–f	48.1a	17.8c–g	1.0e
DK-8-1					
DK-8-1	2.92d–f	39.5bd	40.67a–c	14.42d–h	2.5c–e
DK-8-4					
DK-8-4-3	7.67a–d	49.58a	31.25d–e	10.67f–h	0.83e
DK-8-4-5	3.08ef	40.25a–d	41.25a–c	14.33d–h	1.08e
DK-8-4-6	2.08ef	40a–d	38.75a–c	17.67c–g	1.5de
DK-8-4-8	4b–f	47ab	37.33b–e	10.92e–h	0.75e
DK-8-5					
DK-8-5-1	4.42b–f	36.25c–e	42a–c	15.08d–h	2.25de
DK-8-5-2	7a–e	46.33ab	35.75b–e	10.42gh	0.5e
DK-10-1					
DK-10-1-3	8.42a–e	41.83a–c	39.92a–d	9.58h	0.25e
DK-10-1-4	3d–f	33.08c–e	41.75a–c	19.17cd	3c–e
DK-10-25					
DK-10-25-1	3.42c–f	42.67a–c	38.42b–e	14.08d–h	1.42d–e
PI82-3					
PI82-3-1	2.33ef	38.33b–d	44.5ab	13.92d–h	0.92e
PI82-6					
PI82-6-1	6.33b–e	28.33e–f	34.58c–e	23.83bc	6.92ab
PI82-8					
PI82-8-5	4.5b–f	37.83b–e	41.83a–c	14.5d–h	1.33de
PI82-18					
PI82-18-1	9.17ab	35.92c–e	34.33c–e	18c–f	2.58c–e

Values followed by the same letter in the same column are not significantly different ($P < 0.05$).

^a See Table 1 for an explanation of S_{n+1} lines.

^b The results reported are an average of three replicates for each sample.

starches showed a bimodal distribution with the first peak at dp 13–14 and the second peak at dp 42–46. Branch chains of dp ~ 13 –14 had the largest proportion. The chain-length distribution patterns of all exotic starches were different from that of B73 starch: all exotic starches had either larger relative concentration of short branch-chain at dp 6–12 (Fig. 2a) or less relative concentration of long branch-chain of dp 15–24 (Fig. 2b), or both these characteristics (Fig. 2c) than did B73. Starches from two controls, Mo17 and B73, had slightly different normalized distribution patterns (Fig. 3). The Mo17 starch contained a lower relative concentration of short branch-chain at dp 6–12, a higher relative concentration of long branch-chains at dp 15–18, and a lower relative concentration of long branch-chain at dp 19–32 than did B73 (Fig. 3).

To classify the starches, the chains were assigned into four groupings: dp 6 to 12, 13 to 24, 25 to 36, or > 37 , corresponding to A, B1, B2, and B3 or longer chains, respectively, based on Hizukuri's model (1986) (Table 8). All exotic starches, except DK-10-1-3 (dp 25.3), had relatively lower average chain lengths (dp 23.4–24.5)

than did Mo17 and B73 starches (dp 25.0 and dp 25.5, respectively). All exotic starches contained a greater proportion of A chains at dp 6–12 (16.55–18.77%) than did Mo17 (16.11%) and B73 (16.32%) starches, and a smaller proportion of long B-chains (B2 or B3) at dp ≥ 37 (17.79–21.68 %) than did Mo17 and B73 (22.27 and 22.50%, respectively). Among all exotic starches, CUBA-38-5 starch had the shortest average chain length (dp 23.4), the greatest proportion of A chains at dp 6–12 (18.58%), and the smallest proportion of B2 or B3 chains at dp ≥ 37 (17.79%).

3.6. Molecular weight and gyration radii of amylopectin

Weight-average molecular weight (M_w) and Z-average radius of gyration (R_z) of the amylopectin molecules, determined by using an HPSEC-MALLS-RI system, showed significant differences in (M_w) and (R_z) among all starches (Table 9). Starch from DK-10-1-3 had the largest (M_w) (12.4×10^8) and (R_z) (398.1 nm), whereas starch from DK-8-5-2 had the smallest (M_w) (7.9×10^8), and starch from DK-8-4-3 had the smallest (R_z) (351.1 nm). A Pearson

correlation analysis showed a strong correlation between (M_w) and (R_z) ($r = 0.72$ with $p < 0.0001$).

3.7. Relationship between starch function and granule size distribution

Relationships between starch functional behavior and granule size and shape distributions were analyzed by using the Pearson correlation test. Significant correlations at

$p < 0.10$ and $p < 0.05$ are listed (Table 10). The correlation coefficient (r) values were fairly low, because functional properties of starch depend on many variables, and the effects of each variable might interfere with each other. Thus, even a low r value, when accompanied by a statistically significant value may be important.

The DSC parameter of T_{oG} was positively, and RG was negatively correlated with mean area. Mean area refers to the mean cross sectional area, and is related to the granular

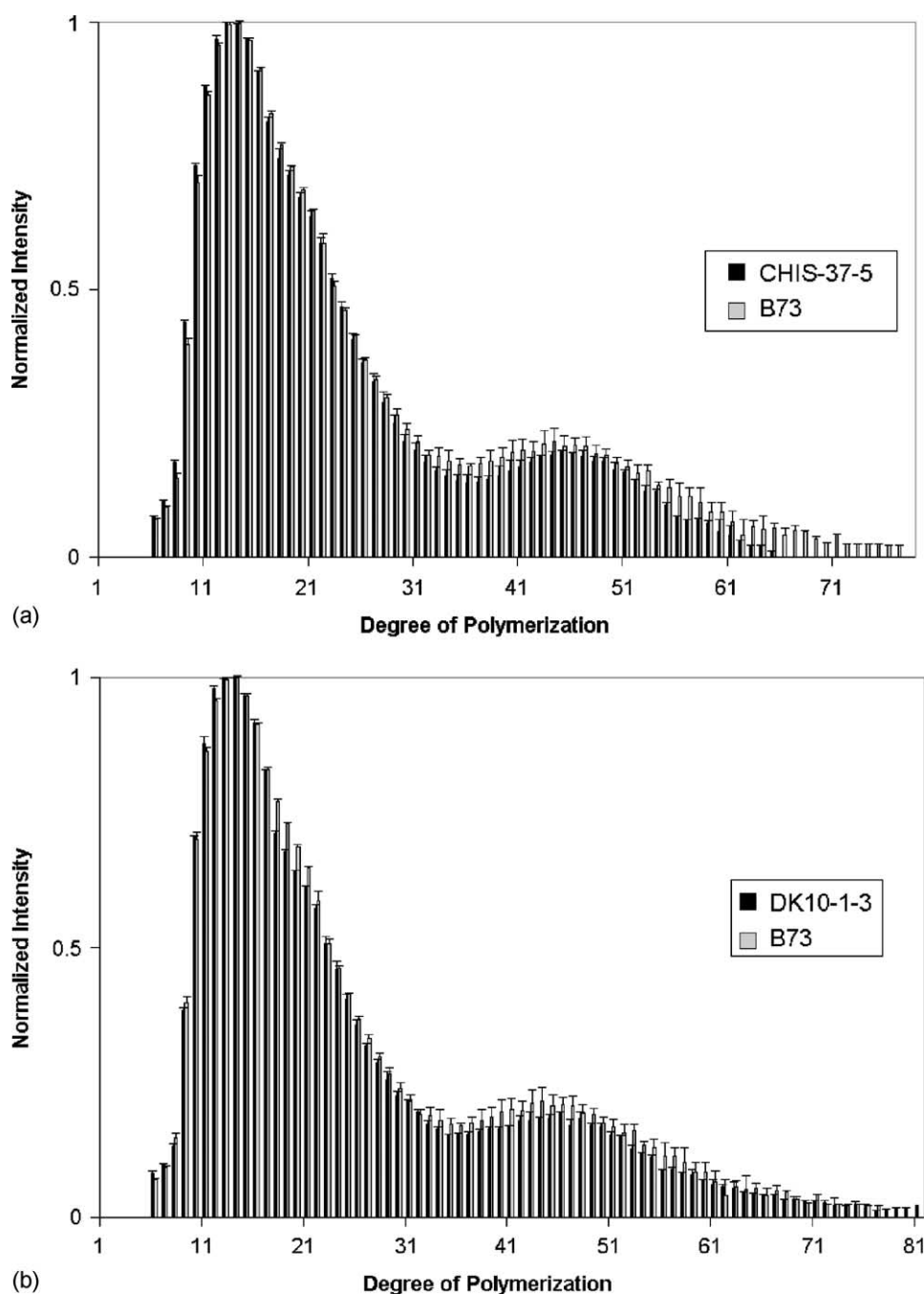


Fig. 2. Normalized branch-chain length distributions of exotic starches with B73 starch determined by using a high-performance anion-exchange chromatography system equipped with an enzyme column reactor and a pulsed amperometric detector (HPAEC-ENZ-PAD). A Carbpac PA100 column and an immobilized amyloglucosidase column were used for the analysis.

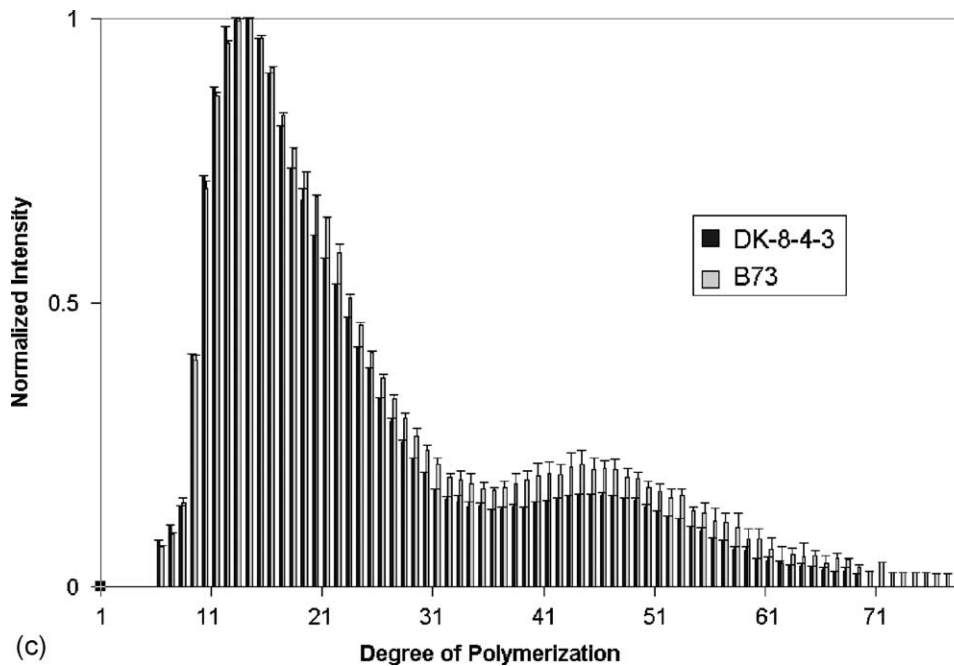


Fig. 2 (continued)

size. In addition, T_{oG} and T_{cG} were positively correlated with percentage of granules having an equivalent diameter of $\geq 17 \mu\text{m}$, whereas T_{oG} was negatively correlated with percentage of granules having an equivalent diameter of $5\text{--}9 \mu\text{m}$.

Other correlations indicated that R% was positively correlated with size of starch granules. The correlation coefficients between R% with starch mean area and major axis both were 0.54, at $p \leq 0.05$. The reason for this might be that larger granules typically contain a greater content of

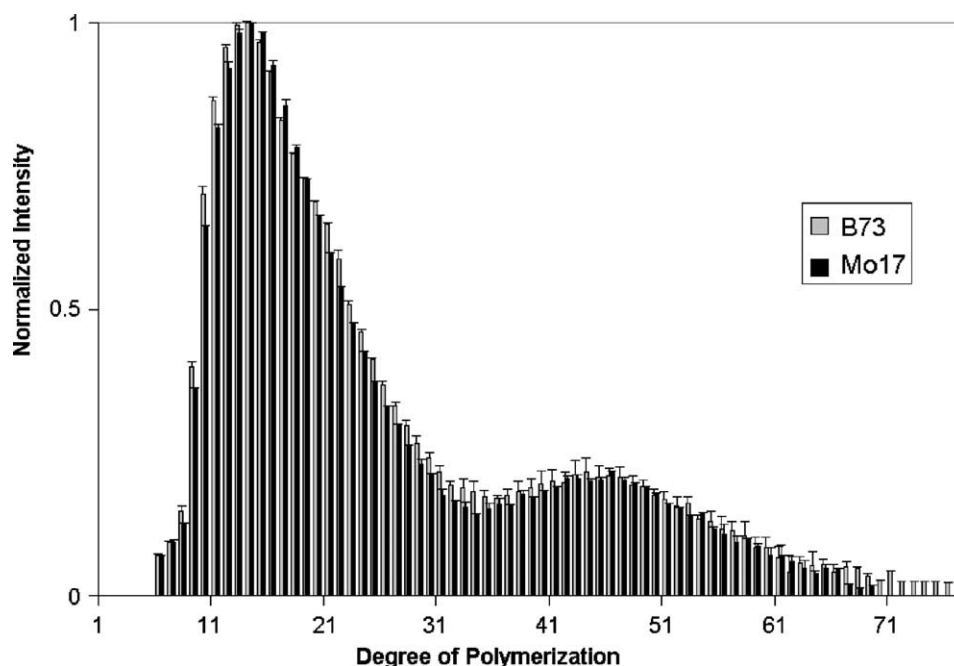


Fig. 3. Normalized branch-chain length distributions of starches from two control lines, B73 and Mo17, determined by using a high performance anion-exchange chromatography system equipped with an enzyme column reactor and a pulsed amperometric detector (HPAEC-ENZ-PAD). A Carbpac PA100 column an immobilized amyloglucosidase column were used for the analysis.

Table 8
Relative branch chain-length (CL) distributions of whole starch

Source of starch (S_{n+1} line ^a -Kernel)	Peak dp		Average CL	Distribution (%)			
	I	II		dp 6–12	dp 13–24	dp 25–36	dp ≥ 37
Mo17	14	46	25.0	16.11	47.54	14.07	22.27
B73	14	44	25.5	16.32	45.92	15.25	22.50
CUBA-38							
CUBA-38-5	13	46	23.4	18.58	48.57	15.05	17.79
CHIS-37							
CHIS-37-5	13	46	23.7	18.00	48.12	15.06	18.82
DK-8-1							
DK-8-1	14	46	24.5	17.08	47.01	15.54	20.37
DK-8-4							
DK-8-4-3	14	44	23.7	18.53	48.53	14.34	18.60
DK-8-4-5	14	45	24.3	17.84	47.91	14.40	19.85
DK-8-4-6	14	45	24.2	17.40	47.63	15.57	19.39
DK-8-4-8	14	45	23.9	18.77	48.06	14.08	19.09
DK-8-5							
DK-8-5-1	14	44	24.5	17.66	47.09	15.64	19.61
DK-8-5-2	14	44	24.4	17.98	47.63	14.47	19.92
DK-10-1							
DK-10-1-3	14	46	25.3	16.96	46.25	15.11	21.68
DK-10-1-4	14	44	24.1	17.69	48.13	15.07	19.11
DK-10-25							
DK-10-25-1	14	45	24.4	16.55	47.14	16.69	19.60
PI82-3							
PI82-3-1	14	47	24.0	17.34	47.97	15.97	18.71
PI82-6							
PI82-6-1	13	44	23.5	18.26	49.80	13.95	18.00
PI82-8							
PI82-8-5	13	46	24.0	17.44	48.02	15.86	18.69
PI82-18							
PI82-18-1	14	46	23.6	17.28	49.60	15.20	17.92

dp, Degree of polymerization. The results reported are an average of at least two replicates for each sample. Grouping of dp numbers followed that of Hanashiro, Abe, and Hizukuri (1996).

^a See Table 1 for an explanation of S_{n+1} lines.

amylose than do small granules (Duffus & Murdoch, 1979). During the cooling and storage of gelatinized starch, starch molecules begin to reassociate, with amylose retrograding faster than amylopectin. Thus the greater amylose content causes more junction zones and a higher $R\%$.

The PV by RVA was negatively correlated with the percentage of small granules having diameter $< 5 \mu\text{m}$. At the same degree of swelling, small granules occupy less volume than larger granules, resulting in the low PV (Medcalf & Gilles, 1968). The breakdown values were positively correlated with the mean major axis of starch granules, meaning that paste stability decreased as the granule size increased. Highly swollen large granules are more fragile and more easily broken by stirring because of their large volume, resulting in a greater decrease in viscosity than for small granules (Medcalf & Gilles, 1968).

3.8. Relationship between starch functional behavior and chain-length distribution

A-chains and B1 chains of amylopectin are primary participants in the crystalline regions. All exotic starches

studied contained a greater proportion of A-chains with dp 6–12 than did starch from Mo17 and B73 (Table 8). Also, the relative concentration of branch chains with dp 15–24 compared with that of peak I (dp 13–14) in the starches displaying lower T_{oG} were lower than those in Mo17 and B73 (Fig. 2a–c). This observation suggests that crystalline structure in the exotic starch was not as perfect as that in Mo17 or B73, because fewer chains in the exotic starches would be long enough to go through the crystalline region, thus resulting in defects in the crystallites (Jane et al., 1999). This observation is corroborated by the DSC data that show lower than normal gelatinization onset temperatures ($T_{oG} < 62^\circ\text{C}$) for all exotic starches of interest (Table 3).

The values of T_{pG} and ΔH_G were positively correlated with average CL, and negatively correlated with the proportion of A-chains at dp 6–12 (Table 10). These results agree with that reported by Shi and Seib (1992, 1995). Tester and Morrison (1990) suggested that T_{pG} represents a measure of starch crystallite perfection, whereas ΔH_G represents the amount of crystalline amylopectin.

The ΔH_R values were positively correlated with the average CL, and negatively correlated with the proportion

Table 9
Amylopectin molecular weights and gyration radii of starches from selected corn lines

Source of starch (S_{n+1} line ^a)	$M_w (\times 10^8)^b$	R_z (nm) ^c
Mo17	11.5ab	386.8ab
B73	8.6d–f	365.7bc
CUBA-38		
CUBA-38-5	8.3ef	368.3a–c
CHIS-37		
CHIS-37-5	8.5d–f	359.1bc
DK-8-1		
DK-8-1	10.2b–d	366.6bc
DK-8-4		
DK-8-4-3	8.0f	351.1c
DK-8-4-5	9.0d–f	365.7bc
DK-8-4-6	9.4c–f	368.3a–c
DK-8-4-8	8.9d–f	374a–c
DK-8-5		
DK-8-5-1	8.5d–f	367.6bc
DK-8-5-2	7.9f	361.9bc
DK-10-1		
DK-10-1-3	12.4a	398.1a
DK-10-1-4	9.8c–e	376.6a–c
DK-10-25		
DK-10-25-1	9.3c–f	361.1bc
PI82-3		
PI82-3-1	9.1d–f	360.8bc
PI82-6		
PI82-6-1	8.9d–f	368.8a–c
PI82-8		
PI82-8-5	8.3ef	368.5a–c
PI82-18		
PI82-18-1	10.9a–c	385.2ab

^a See Table 1 for an explanation of S_{n+1} lines.

^b Weight—average molecular weight. The results reported are an average of three replicates for each sample.

^c z -Average radius of gyration. The results reported are an average of three replicates for each sample.

of A-chains at dp 6–12, because A-chains at dp 6–12 inhibit retrogradation (Shi & Seib, 1992). The proportions of A-chains at dp 6–12 also were negatively correlated with P_{temp} by RVA. Perhaps more short chains at dp 6–12, leading to less tightly packed crystalline structures, also promoted swelling.

3.9. Correlation among functional properties of starches

The PV and breakdown from RVA were negatively correlated with T_{pG} and T_{cG} (Table 11). High gelatinization temperatures reflected by T_{pG} and T_{cG} will slow the gelatinization process, thus leading to a low PV. The proportion of short A chains of amylopectin could be the reason for the negative correlation between breakdown values and T_{pG} and T_{cG} . A great proportion of A-chains at dp 6–12 will result in a less perfect crystalline structure in the granule and lead to a low gelatinization temperature. At the same time, a great proportion of A-chains at dp 6–12 will promote swelling and result in high breakdown values (Jane et al., 1999).

Firmness and stickiness of gels by the TA were strongly correlated with retrogradation of starch (Table 11). As the starch became more crystallized, water was squeezed out of the gel, causing a firmer, more concentrated gel. With less water, the gels also had greater stickiness (Thomas & Atwell, 1999a).

3.10. Comparison of functional and structural properties of two corn-belt lines Mo17 and B73

Starch from Corn Belt inbreds, Mo17 and B73, used as control lines in this study were significantly different from each other in function and structure. The Mo17 corn belongs

Table 10
Pearson correlation coefficients (r) of starch functional properties with structural properties

Functional properties	Granule size parameter		Granule size distribution profiles ^a					Branch chain length distribution		
	Mean area	Mean major axis	≥ 17 (μm)	13–17 (μm)	9–13 (μm)	5–9 (μm)	< 5 (μm)	CL	dp 6–12 (%)	dp ≥ 37 (%)
<i>DSC parameter^b</i>										
T_{oG}	0.56**	0.53**	0.48**	0.50**		–0.60**			–0.49**	
T_{pG}								0.50**	–0.52**	–0.59**
T_{cG}			0.50**		–0.48**					
H_G								0.49**	–0.42*	0.58**
R_G	–0.56**	–0.54**		–0.44*		0.52**		0.47*		
H_R								0.48**	–0.48**	0.49**
$r\%$	0.54**	0.54**								
<i>RVA parameter^c</i>										
Setback								–0.41*		–0.46*
Breakdown		0.40*c								
PV					0.56**		–0.55**			

*, **: p -values for test $H_0: \rho = 0$ vs. $H_a: \rho = 0$ are smaller than 0.10 and 0.05, respectively. The value of 0.10 was used, instead of 0.01, because many features contribute to starch structure and function, resulting in small correlation coefficients.

^a Starch granules are divided into different groups according to equivalent diameter.

^b See Table 2 for DSC parameter descriptions.

^c See Table 4 for RVA parameter descriptions.

Table 11
Pearson correlation coefficients (r) among functional properties of starch

	DSC parameter ^a									Gel parameter ^b			
	T_{oG}	T_{pG}	T_{cG}	R_G	H_G	T_{or}	R_r	H_r	$r\%$	P_{temp}	PV	Breakdown	F1
Gel parameter													
S7								0.63**	0.59**				
F7					0.42*	−0.47*		0.57**		−0.49**	0.43*		0.78**
S1										−0.49**			
F1					0.44*	−0.42*		0.45*					
Setback		−0.71**	−0.64**		−0.77**	−0.45*	0.42*		0.44*		0.59**	0.74**	
Breakdown		−0.45*	−0.46*	−0.54*					0.48**		0.73**		
PV		−0.65**	−0.42*			0.45*							
P_{temp}													
DSC parameter													
$r\%$								0.83**					
ΔH_r													
R_r				−0.43*		−0.88**							
T_{or}	0.57**	0.62**	0.67**			−0.88**							
R_G	−0.94**												
ΔH_G		0.50**											
T_{cG}	0.43*	0.79**											
T_{pG}	0.62**												

S7, Stickiness after 7 days of storage at 4 °C. F7, firmness after 7 days of storage at 4 °C. S1, stickiness after 1 day of storage at 25 °C. F1, firmness after 1 day of storage at 25 °C. *, **: p -values for test $H_0: \rho = 0$ vs. $H_a: \rho \neq 0$ are smaller than 0.10 and 0.05, respectively. The value of 0.10 was used, instead of 0.01, because many features contribute to starch structure and function, resulting in small correlation coefficients.

^a See Table 2 for DSC parameter descriptions.

^b See Table 4 for gel RVA parameter descriptions.

to the non-Stiff Stalk heterotic pattern and B73 belong to the Stiff Stalk heterotic pattern. The Mo17 starch contained a smaller relative concentration of A-chains at dp 6–12, a larger relative concentration of B1 chains at dp 15–18, and a smaller relative concentration of chains at dp 19–22 than did B73 (Fig. 3). This observation is corroborated by the DSC data in that Mo17 showed higher T_{oG} , T_{pG} and ΔH_G than B73 starch. In addition to the structural differences in the chain-length distributions, starch from Mo17 and B73 had significantly different granular sizes and shape distributions. Starch granules from B73 were larger in size and more spherical in shape than were starch granules from Mo17. Because of the larger size, more spherical shape and higher proportions of A-chains at dp 6–12, starches from B73 had greater PV, and larger breakdown values than did starch from Mo17.

4. Conclusion

The exotic lines were selected because of a low T_{oG} and a wide R_G . The different gelatinization and pasting properties could be explained by the branch chain-length pattern of the amylopectin. Starch granule-size distribution profiles of exotic starches were different than those of control starches. These results suggest that incorporation of exotic alleles into Corn Belt germplasm is an excellent means to obtain value-added traits to produce starch with desirable functions.

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