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Genetic Variation for Starch Thermal and Functional Properties Among Nonmutant Maize Inbreds'

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Abstract

Differential scanning calorimetry (DSC) has been used in previous < 0.01) inbred by year interaction was present for all DSC parameters studies to detect differences in thermal properties among starches of non- with the exception of AH. Differences were observed in starch viscosities mutant maize (Zea mays L.) genotypes. This study was conducted to and gel strengths for six inbreds selected for highest and lowest Tp, AH, determine the magnitude of genetic and genotype by environmental effects and range (R,). Several significant ($P < 0.05$) correlations occurred on starch properties among a set of exotic and domestic inbred lines. between DSC parameters and starch paste viscosities and gel strengths. Functional properties of starches from selected lines exhibiting extreme These data suggest that evaluation of starches from nonmutant genotypes DSC values also were investigated. Highly significant ($P< 0.01$) differences by DSC can be used to predict some functional properties. A practical for DSC starch thermal properties were seen among the lines. Starches application of DSC in breeding programs may include screening maize from exotic lines generally had lower gelatinization onset temperature germplasm for extreme DSC values or population improvement through (TO), peak temperature (Tp), and enthalpy (AH). A highly significant (P recurrent selection. The maize wet-milling industry produces a number of starch- based products important in the food industry. Genetic variability in starch structure and functional properties has led to the use of specialty starches from waxy and high-amylose genotypes (Shannon and Garwood 1984). More recently, the introduction of starches containing double mutant combinations with proper- ties similar to chemically modified starches has resulted in several patents (Katz 1991). The application of differential scanning calorimetry (DSC) to the study of starch was first described by Stevens and Elton (1971). This technique offers a thermodynamic approach to the study of starch gelatinization by monitoring changes in the physical and chemical properties of starches (Donovon et al 1983). Use of DSC in investigating the thermal behavior of starches has become increasingly more popular because it requires only a small sample size and is easy to operate (Sanders et al 1990). Additionally, DSC is relatively rapid compared with more traditional methods of studying starch gelatinization, making it suitable for breeding programs. Extensive variations in DSC parameters have been observed among starches of single- and double-mutant genotypes of maize indicating differences in starch structure and function (Brockett et al 1988, Sanders et al 1990, Wang et al 1992). More recent studies have revealed variations in DSC parameters among non- mutant sources of maize starch. Krueger et al (1987), for example, found differences in DSC parameters among two maize inbred lines and suggested that AH and peak height index (PHI) could be used as a means of identifying maize genotypes. White et al (1990) reported variability in thermal properties by DSC in genetically variable maize populations. The largest differences were observed for gelatinization onset (T.), range (Rn), and total enthalpy (AH). In addition, Li et al (1994) found large variations in DSC values among several exotic populations of maize, sug- gesting that selection among these on the basis of DSC values would identify genotypes having desired starch properties.

Disciplines

Food Biotechnology | Food Processing | Food Science | Human and Clinical Nutrition

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Genetic Variation for Starch Thermal and Functional Properties Among Nonmutant Maize Inbreds[1]

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ABSTRACT

Differential scanning calorimetry (DSC) has been used in previous studies to detect differences in thermal properties among starches of nonmutant maize (*Zea mays* L.) genotypes. This study was conducted to determine the magnitude of genetic and genotype by environmental effects on starch properties among a set of exotic and domestic inbred lines. Functional properties of starches from selected lines exhibiting extreme DSC values also were investigated. Highly significant ($P \leq 0.01$) differences for DSC starch thermal properties were seen among the lines. Starches from exotic lines generally had lower gelatinization onset temperature (T_o), peak temperature (T_p), and enthalpy (ΔH). A highly significant ($P \leq 0.01$) inbred by year interaction was present for all DSC parameters with the exception of ΔH. Differences were observed in starch viscosities and gel strengths for six inbreds selected for highest and lowest T_p, ΔH, and range (R_n). Several significant ($P \leq 0.05$) correlations occurred between DSC parameters and starch paste viscosities and gel strengths. These data suggest that evaluation of starches from nonmutant genotypes by DSC can be used to predict some functional properties. A practical application of DSC in breeding programs may include screening maize germplasm for extreme DSC values or population improvement through recurrent selection.

The maize wet-milling industry produces a number of starch-based products important in the food industry. Genetic variability in starch structure and functional properties has led to the use of specialty starches from waxy and high-amylose genotypes (Shannon and Garwood 1984). More recently, the introduction of starches containing double mutant combinations with properties similar to chemically modified starches has resulted in several patents (Katz 1991).

The application of differential scanning calorimetry (DSC) to the study of starch was first described by Stevens and Elton (1971). This technique offers a thermodynamic approach to the study of starch gelatinization by monitoring changes in the physical and chemical properties of starches (Donovon et al 1983). Use of DSC in investigating the thermal behavior of starches has become increasingly more popular because it requires only a small sample size and is easy to operate (Sanders et al 1990). Additionally, DSC is relatively rapid compared with more traditional methods of studying starch gelatinization, making it suitable for breeding programs.

Extensive variations in DSC parameters have been observed among starches of single- and double-mutant genotypes of maize indicating differences in starch structure and function (Brockett et al 1988, Sanders et al 1990, Wang et al 1992). More recent studies have revealed variations in DSC parameters among nonmutant sources of maize starch. Krueger et al (1987), for example, found differences in DSC parameters among two maize inbred lines and suggested that ΔH and peak height index (PHI) could be used as a means of identifying maize genotypes. White et al (1990) reported variability in thermal properties by DSC in genetically variable maize populations. The largest differences were observed for gelatinization onset (T_o), range (R_n), and total enthalpy (ΔH). In addition, Li et al (1994) found large variations in DSC values among several exotic populations of maize, suggesting that selection among these on the basis of DSC values would identify genotypes having desired starch properties.

Little information exists regarding the nature of genetic variability among nonmutant sources of maize starch for thermal properties. More importantly, how this variation relates to the structural and functional characteristics of the starches must be established. This study, therefore, was conducted to describe the genetic variability in DSC parameters among a set of domestic and exotic maize inbred lines and to relate these values to differences in plant morphological traits and in starch structural and functional properties.

MATERIALS AND METHODS

Experiment 1

Twenty-six maize inbreds were chosen on the basis of differences in maturity, kernel characteristics, and pedigree (Table I). Inbreds were grown in 1992 and 1993 at the Agronomy and Agricultural Engineering Research Center, Ames, IA. The experiments were planted in a randomized complete-block design with two replicates. A plot consisted of one 12-ft row thinned to ~25 plants. Plants were self-pollinated to ensure genetic purity of the grain. Flowering date (FD) was recorded as the number of days after planting to self-pollination, and ears were harvested at physiological maturity as determined by presence of the black layer (Daynard and Duncan 1969). Ears were dried at 38°C for 48 hr to a moisture content of ~13%. One-hundred kernel weight (HKW) was determined from the dried grain. Poor growing conditions in 1993 did not allow the inbred ND246 to reach physiological maturity and it was eliminated from the combined analysis of variance.

A small-scale starch isolation procedure was conducted on a bulk of five kernels obtained from the center portion of each ear (White et al 1990). Separate starch extractions were made from two ears per replicate for the determination of DSC thermal properties and amylose content (AM).

DSC

For DSC analysis, a Perkin-Elmer DSC 7 analyzer equipped with a thermal-analysis data station (Perkin-Elmer Corp., Norwalk, CT) was used. Analysis of starch gelatinization was conducted as described by White et al (1990). Starch (~4.0 mg, dwb) was weighed into aluminum sample pans with 8 mg of distilled water. Samples were heated from 30 to 102°C at a rate of 10°C/min. DSC parameters recorded for this study included ΔH, T_o, T_p, and R_n. The parameters T_o, T_p, and ΔH were calculated directly by the DSC software. The R_n was calculated as $2(T_p - T_o)$ (Krueger et al 1987). Samples were stored for seven days at 4°C and rerun

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to determine starch gel retrogradation (%R) as described by White et al (1989). A standard of o-terphenyl, which produces a peak at ~60°C, was run each day to detect day-to-day variation.

Amylose Determination

AM was determined colorimetrically by dissolving ~5.0 mg of starch in 10 ml of 90% DMSO containing $6 \times 10^{-3}M$ iodine (Knutson 1986). One milliliter of the dissolved sample was diluted to 9 ml with H_2O, and the absorbance measured at 600 nm on a spectrophotometer (Hitachi U-2000, Tokyo, Japan). Purified AM was prepared from maize starch as described by Schoch (1942) and used to construct a standard curve.

Statistical Analysis

All kernel measurements were analyzed by using an analysis of variance (ANOVA) procedure for a randomized complete block design. The ANOVA model was:

$$X_{ijk} = u + Y_i + R_{(i)y} + I_k + YI_{ik} + \epsilon_{(ijk)}$$

where X_{ijk} represents mean DSC characteristics, HKW, FD, and AM for inbred k in replicate j within year i; u is the overall mean value; Y_i is the random effect for year; $R_{(i)y}$ is the random effect for replicates; I_k is the fixed effect for inbreds; YI_{ik} is the interaction effect of inbred with year; and $\epsilon_{(ijk)}$ is the pooled experimental error. Differences among treatment means for traits were determined by using Fisher's least significant difference test ($a = 0.05$) (Steel and Torrie 1980). Repeatability (R) was calculated in a manner similar to heritability as described by Hallauer and Miranda (1981) from the combined ANOVA, with the exception that the effect of inbred (k^2_I) was considered to be fixed rather than random:

$$R = k^2_I / [\sigma^2/\mathrm{yr} + r\sigma^2_{Iy}/y + k^2_I]$$

Experiment 2

Starches of six inbreds evaluated for thermal properties by DSC from the 1992 growing season (Experiment 1) were selected for having the highest and lowest values for T_p, ΔH, and R_n (Fig. 1). These inbreds were analyzed for starch functional properties.

TABLE I
Thermal Properties of Exotic and U.S. Corn Belt Maize Inbreds Grown in 1992 and 1993 at Ames, IA

Inbred	Source	Race	Seed Color	Seed Type
PI 186190[a]	Uruguay	Cateto	Orange	Flint
PI 186216	Argentina	Cateto	Orange	Flint
PI 186227	Uruguay	Cateto	Orange	Flint
PI 186228	Uruguay	Cateto	Yellow	Flint
PI 198904	Argentina	Cateto	Orange	Flint
PI 221805	South Africa	Cateto	Yellow	Flint
PI 303943	Taiwan	Intermediate[b]	Yellow	Flint
A619	USA-MN	CBD[c]	Yellow	Dent
A632	USA-MN	CBD	Yellow	Dent
A641	USA-MN	CBD	Yellow	Dent
B73	USA-IA	CBD	Yellow	Dent
CM105	USA-Canada	CBD	Yellow	Dent
CM145	USA-Canada	CBD	Yellow	Dent
H99	USA-IL	CBD	Yellow	Dent
MO17	USA-MO	CBD	Yellow	Dent
NC250	USA-NC	CBD	Yellow	Dent
ND246	USA-ND	CBD	Yellow	Dent
OH43	USA-OH	CBD	Yellow	Dent
OS420	USA-IA	CBD	Yellow	Dent
SDp310	USA-SD	CBD	Yellow	Dent
SDp312	USA-SD	CBD	Yellow	Dent
W117	USA-WI	CBD	Yellow	Dent
W64A	USA-WI	CBD	Yellow	Dent
W845	USA-WI	CBD	Yellow	Dent
MBS7W	USA-MBS[d]	CBD	White	Dent
MBS9W	USA-MBS	CBD	White	Dent

[a] Non-Corn Belt inbreds were obtained from North Central Regional Plant Introduction Station, Ames, IA.
[b] Intermediate in characteristics between the races Cateto and Corn Belt dent.
[c] Corn Belt dent.
[d] MBS, Inc., 225 W. 1st St., Story City, IA 50248.

Starch Extraction

Remaining starch from the six selected inbreds previously prepared for DSC analysis was used for determination of birefringence end-point temperature (BEPT). For all other analyses, starch was isolated from the bulk of remnant seed obtained from at least 10 ears per replicate as described by Steinke and Johnson (1991). After isolation of starch, samples were purified with five volumes of $0.2M$ sodium chloride-toluene (5:1, v/v) at least five times, and starch granules were allowed to sediment. The final sediment was washed three times with distilled water and dried at 45°C for 24 hr.

BEPT

A single starch sample from each replicate was used to determine BEPT as described by Brown (1966). Granules dispersed in water were viewed on slides by using a light microscope equipped with a microscope heating stage 350 (Dialux, Ernst Leitz, Midland, ON, Canada) adjusted to a 3–4°C increase per minute. Ten granules in the viewing field were monitored for disappearance of birefringence upon heating of granules, beginning at 50°C. This procedure was conducted in triplicate, recording 30 granules per sample. BEPT was calculated as the temperature at which 50% of the granules lost birefringence. The BEPT range was recorded as the difference in temperature of initial loss to complete loss.

Amylography

A viscoamylograph (C. W. Brabender, S. Hackensack, NJ) equipped with a 700-cmg sensitivity cartridge operating at a bowl speed of 75 rpm was used to determine pasting characteristics of starch suspensions (8%, dwb), which were adjusted to pH 5.5 as described by Wang et al (1992). The temperature was raised from 30 to 95°C at a rate of 1.5°C/min, maintained at 95°C for 30 min, then lowered to 50°C at the same rate, and held for 30 min. Measurements for starches of each inbred were made in duplicate.

Gel Strength

Starch pastes from each genotype prepared by using the Brabender Viscoamylograph were used to measure gel strength after storage for one and seven days at 4°C. Starch pastes were poured into aluminum dishes (27 mm, i.d. × 27 mm), taped around the rims to increase the depth, and later cut back to produce a fresh surface before analysis. From each of two replicates, gel strength was measured at five different locations on each gel sample, and two gel samples per starch type were measured after one or seven days by using a Voland texture analyzer (Texture Technologies, Scarsdale, NY) as previously described by Takahashi et al (1989).

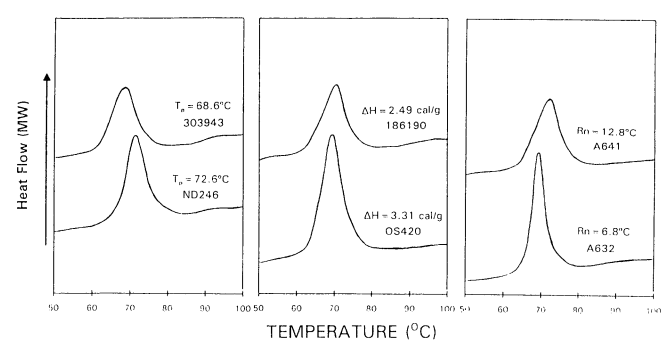

Fig. 1. Representative differential scanning calorimetry thermograms for selected inbreds grown in 1992 exhibiting the highest and lowest peak temperature (T_p), enthalpy (ΔH), and gelatinization range (R_n).

Statistical Analysis

All parameters were analyzed by using ANOVA for a completely randomized design. Differences among treatment means for traits were tested with Fisher's LSD test ($a = 0.05$) (Steel and Torrie 1980). Relationships among DSC parameters, BEPT, amylography, and gel strengths were determined with Pearson's simple correlation test (SAS 1990).

RESULTS AND DISCUSSION

Experiment 1

DSC parameter means of starches from the 25 inbreds combined across the 1992 and 1993 environments are shown in Table II. Levels of significance for year, inbred, and year by inbred effects from ANOVA are shown in Table III. Year of evaluation contributed to significant ($P \leq 0.05$) variations for ΔH, R_n, and $\%R$. Large variations in growing temperature and precipitation between 1992 and 1993 may have contributed to these differences. Previous studies also have revealed that environmental effects, such as date of planting and growing location, may influence these values (White et al 1991, Campbell et al 1992).

Differences among inbreds for all DSC parameters were highly significant ($P \leq 0.01$). Two inbreds of the Cateto race (PI 198904 and PI 303943) exhibited the lowest T_o and T_p values, respectively, whereas two Corn Belt inbreds (OH43 and A619) had the greatest values for T_o and T_p. Two Cateto inbreds (PI 186190 and PI 303943) also had the smallest ΔH and greatest R_n and $\%R$, whereas Corn Belt inbreds (OH43, MO17 and W845) had the greatest ΔH and smallest R_n and $\%R$. Collectively, all Cateto inbreds differed significantly from Corn Belt inbreds (Table III). Generally, means for T_o, T_p, and ΔH were significantly greater, and R_n and $\%R$ were significantly smaller for Corn Belt inbreds than for inbreds of the Cateto race. Previous investigations have shown that $\%R$ reflects stability of starch gels under refrigerated storage conditions for chemically modified and endosperm mutant starches, in which lower values are associated with greater stability (White et al 1989). Highly significant variations in $\%R$ among inbreds suggested differences in stability of starch gels among these genotypes.

The ranges observed for DSC values among the inbreds in this study (Table II) were not as large as those previously observed among mutant sources of maize starch (Brockett et al 1988, Sanders et al 1990, Wang et al 1992). The presence of genetic variations for all DSC parameters in this study suggests that, as with mutant starches, there may be a structural or morphological basis for these differences.

The effect of inbred on AM, HKW, and FD also was significant (Table III). Mean values for HKW and FD varied among inbreds, especially between those of Cateto and Corn Belt sources (Table II). Mean AM ranged from 22.5 to 28.1%, which is similar to the range generally found among nonmutant sources of maize starch.

Highly significant genotype by environment interactions were detected for both HKW and FD, but this interaction was absent for AM. ANOVA also indicated significant inbred by year interactions among all DSC parameters with the exception of ΔH.

TABLE II
Mean Parameters of Differential Scanning Calorimetry Thermograms, 100-Kernel Weight, Days to Flowering, and Amylose Content for 25 Maize Inbreds Grown in 1992 and 1993 at Ames, IA

Inbred	T_o[a] (°C)	T_p[b] (°C)	ΔH[c] (cal/g)	R_n[d] (°C)	R[e] (%)	HKW[f] (g)	FD[g] (days)	AM[h] (%)
PI 186190	63.3	69.5	2.22	12.3	65.3	18.9	82.8	23.3
PI 186216	66.1	71.4	2.59	10.8	59.8	22.4	83.3	25.8
PI 186227	66.5	71.6	2.50	10.2	55.8	12.3	81.5	24.8
PI 186228	67.4	71.6	2.55	8.3	55.8	22.1	76.8	28.1
PI 198904	62.6	68.2	2.23	11.0	57.7	13.2	93.1	24.8
PI 221805	65.4	69.6	2.61	8.4	52.6	18.3	77.5	23.6
PI 303943	63.8	68.0	2.21	8.3	63.5	17.0	88.0	26.4
A619	68.0	72.5	2.71	9.0	56.3	26.5	76.5	25.0
A632	64.6	68.8	2.60	8.3	51.1	22.0	83.4	26.4
A641	65.4	70.8	2.54	10.8	59.0	22.0	76.0	26.4
B73	65.0	69.4	2.54	8.7	54.4	22.6	84.6	24.3
CM105	67.2	72.2	2.59	9.9	56.6	23.3	74.5	25.5
CM145	67.7	72.1	2.76	8.7	53.3	20.5	73.7	27.3
H99	65.5	71.3	2.57	11.5	57.4	24.3	81.9	23.4
MBS7W	63.5	68.1	2.34	9.1	58.9	23.4	90.5	25.5
MBS9W	65.0	69.5	2.46	9.1	56.2	22.5	84.5	25.2
MO17	67.4	71.3	2.68	7.8	53.5	27.0	84.7	27.1
NC250	63.7	68.6	2.32	9.9	59.4	20.3	87.8	26.8
OH43	68.2	72.3	2.82	8.2	54.3	24.4	76.5	26.3
Os420	64.6	69.5	2.71	9.7	51.9	26.6	82.4	25.6
SDp310	65.5	70.1	2.38	9.2	56.6	23.8	79.4	25.5
SDp312	64.4	70.2	2.54	11.7	56.1	20.4	77.0	26.5
W117	66.7	71.2	2.73	9.0	54.6	25.4	80.5	22.5
W64a	64.3	69.5	2.45	10.5	54.3	22.8	80.3	25.5
W845	67.1	71.7	2.78	9.0	50.4	26.0	74.5	26.7
Mean	65.5	70.3	2.53	9.6	56.2	22.0	81.3	25.6
Domestic	65.7	70.5	2.58	9.5	55.3	23.6	80.5	25.7
Exotic	64.9	69.9	2.40	10.0	59.0	17.5	83.6	25.4
Minimum	62.7	68.0	2.21	7.8	50.4	12.3	73.7	22.5
Maximum	68.0	72.5	2.82	12.3	65.3	27.0	93.1	28.1
LSD[i]	1.8	1.3	0.22	1.9	4.7	3.0	2.7	2.3

[a] Gelatinization onset.
[b] Gelatinization peak.
[c] Enthalpy of gelatinization.
[d] Gelatinization range.
[e] Retrogradation (ΔH rerun after seven days under storage at 4°C/ΔH initial run) × 100.
[f] 100-kernel weight.
[g] Flowering date.
[h] Amylose content.
[i] Least significant difference ($\alpha = 0.05$).

TABLE III
Mean Squares from the Combined Analysis of Variance Across the 1992 and 1993 Growing Season for Differential Scanning Calorimetry Parameters, Amylose Content, 100-Kernel Weight, and Flowering Date

Source	T_o[a] (°C)	T_p[b] (°C)	ΔH[c] (cal/g)	R_n[d] (°C)	%R[e] (%)	HKW[f] (g)	FD[g] (days)	AM[h] (%)
Year (Y)	0.6	2	0.3**[i]	19**	1885**	2806**	173**	321**
Rep/Y	16**	8**	0.1**	6*	24	16*	1.0	20**
Inbred (I)	10**	8**	0.1**	6**	49**	64**	101**	5*
Cateto vs.								
Corn Belt dent	15**	7**	0.6*	7*	333**	899**	201**	0.03
Exotic	19**	17**	0.2**	17**	75	124**	224**	13**
Domestic	17**	14**	0.1**	8**	51**	40**	181**	9*
I × Y	4**	2**	0.02	5**	25**	24**	17**	2
CV%[j]	2.0	1.4	5.6	13.3	5.8	9.9	2.3	6.2
Repeatability	0.56	0.69	0.80	0.17	0.48	0.62	0.84	0.68

[a]Gelatinization onset.
[b]Gelatinization peak.
[c]Enthalpy of gelatinization.
[d]Gelatinization range.
[e]Retrogradation (ΔH rerun after seven days under storage at 4°C/ΔH initial run) × 100.
[f]100-kernel weight.
[g]Flowering date.
[h]Amylose content.
[i]*, ** = Significant at $P \leq 0.05$, 0.01, respectively.
[j]Coefficient of variation.

TABLE IV
Ranking for Gelatinization Peak (T_p), Range (R_n), and Enthalpy (ΔH) for the Five Highest and Lowest Maize Inbreds, from 1992 and 1993 Based on the Two-Year Mean

T_p	1992	1993	ΔH	1992	1993	R_n	1992	1993
Highest								
A619	18	20	OH43	19	17	PI 186190	20	18
OH43	16	19	W845	16	18	A641	21	5
CM105	15	21	CM145	12	17	PI 186216	15	17
CM145	13	18	W117	17	16	W64a	14	15
W845	14	17	Os420	20	6	PI 186227	18	11
Lowest								
PI 303943	2	1	PI 303943	2	1	Mo17	4	2
MBS7W	1	4	PI 186190	1	2	Oh43	3	4
PI 198904	3	2	PI 198904	3	1	A632	1	12
NC250	7	3	NC250	5	3	PI 186228	7	3
A632	6	5	MBS7W	4	4	PI 303943	3	7

TABLE V
Birefringence End-Point Temperature (BEPT) for Selected Maize Inbreds Grown in 1992

Genotype	BEPT (°C)	BEPT Range (°C)
PI 303943	58.6	4.7
A641	59.8	6.5
ND246	63.2	5.0
OS420	59.7	5.2
A632	61.1	3.9
PI 186190	60.2	5.0
Mean	60.4	5.0
LSD[a]	2.0	NS[b]

[a]Least significant difference ($\alpha = 0.05$).
[b]Not significant.

Repeatability measures (Table III) for DSC values indicated that, with the exception of R_n and %R, most of the variation was due to genotypic differences. In addition, inbreds in the lower one-fifth of mean values over the two years generally ranked consistently in each environment (Table IV).

Experiment 2

Representative DSC thermograms for inbreds evaluated in 1992 exhibiting the highest and lowest T_p, ΔH, and R_n are shown in Figure 1. The inbred ND246, characterized by the greatest T_p in 1992, was not included in the combined analysis in Experiment 1 because of poor growing conditions in 1993. Starch samples from these six inbreds were studied further to determine whether the differences in T_p, ΔH, and R_n reflected differences in functional properties.

BEPT

Loss of birefringence in starch is associated with the disruption of the crystalline structure during gelatinization, and values are often unique among starches from maize endosperm mutants (Shannon and Garwood 1984). The purpose of examining BEPT among the six starches was to determine whether these values agreed with the DSC values T_o and T_p for monitoring the temperature at which gelatinization begins. Mean BEPT and ranges among the six selected inbreds are shown in Table V. Inbreds varied significantly with regard to BEPT, whereas no significant differences were observed for the range of birefringence loss. The inbred PI 303943 had the lowest BEPT (58.6°C) value, and ND246 had the highest value (63.2°C). The BEPT values of these two inbreds correspond to their ranking in T_p, suggesting that these two methods measure similar characteristics of the granules. Loss of birefringence in starch has traditionally been used as an indicator of gelatinization temperature. Wide variations in gelatinization temperature among starches of various maize endosperm mutants and botanical sources also have been observed whereby the BEPT and T_p value have generally agreed well (Biliaderis et al 1980).

Amylography

A typical amylogram from a starch paste using normal (nonmutant) maize starch is shown in Figure 2. Important points recorded during the programmed heating and cooling cycle include

pasting onset (A), peak temperature and viscosity (B), viscosities at 95°C (C), held at 95°C for 30 min (D), 50°C (E), and held at 50°C for 30 min (F). Mean values from amylograms are shown in Table VI. Amylograms from each inbred resembled that of normal maize starch pastes. However, significant differences among inbreds were observed for peak temperature, peak viscosity, and viscosities at 95°C, 95°C hold, and 50°C hold. The inbred PI 303943 had the highest amylogram peak temperature and the lowest peak viscosity measurement, whereas ND246 had the lowest amylogram peak temperature and the highest peak viscosity. Of the six inbreds, PI 303943 and ND246 were selected for high and low T_p, respectively, suggesting that high T_p values are associated with lower amylogram peak temperatures and higher peak viscosities. Although significant differences in viscosities were seen among the inbreds after cooling, and their rankings in viscosity measurement were not consistent throughout the cycle.

Gel Strength

Gel strength measurements were determined by texture analysis of mean values for gel firmness after one and seven days of storage at 4°C are shown in Table VII. Significant differences among inbreds in firmness were observed after one and seven days of storage. Inbreds having high (ND246) and low values (PI 186190) for firmness corresponded to high and low values for T_o, respectively. For all inbreds, there was an increase in gel firmness after storage for seven days. However, the ratio of the gel firmness after seven days of storage varied when compared to that of one-day storage. For example, the ratio was greatest for PI 303943 (2.08) and smallest for Os420 (1.03). These high and low values corresponded to the high and low values for $\%R$, respectively. Because retrogradation results in separation of water from the gel, it might follow that a more unstable gel would result in a more concentrated and firm gel.

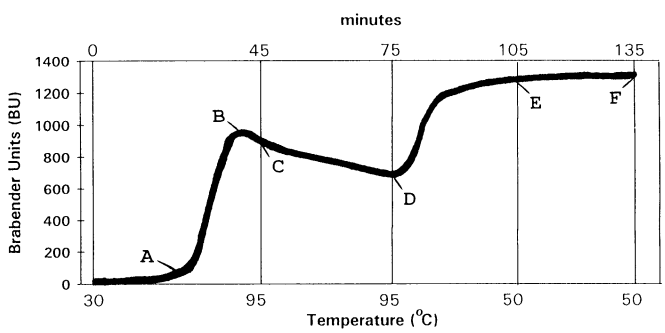

Fig. 2. Amylogram from normal (nonmutant) maize starch showing pasting onset (A), peak temperature and viscosity (B), viscosities at 95°C (C), held at 95°C for 30 min (D), 50°C (E), and held at 50°C for 30 min (F).

Correlation Analysis

Simple correlation coefficients of DSC parameters with functional properties of the six selected inbreds are shown in Table VIII. Values for T_o and T_p were significantly correlated with BEPT. The significant correlation of BEPT with T_o and T_p demonstrates that these methods measure similar properties of the starch. Measurement of gelatinization onset by DSC rather than BEPT, however, would be expected to be a more accurate measure because more granules are represented. The temperature range of birefringence loss did not correlate with the range of the DSC endotherm.

A significant negative correlation ($r = -0.92$) occurred between viscosity peak temperature and T_p. A significant positive correlation ($r = 0.86$) also was detected for the viscosity at 95°C and T_p. Significant positive correlations occurred between gel firmness at one and seven days of storage with T_o and seven-day storage and T_p. Although not significant, the correlation between $\%R$ and the ratio of gel firmness ($r = 0.66$) suggests that these may both be measures of gel stability. Wang et al (1992) examined starches from a selected group of eight maize endosperm mutants and found significant relationships between DSC parameters and amylograph values. For example, significant correlations existed between ΔH and pasting T_o ($r = -0.82$), peak viscosity ($r = 0.85$), and viscosity at 95°C ($r = 0.79$). These results demonstrate that DSC values can be used to identify measurable differences in functional properties among nonmutant maize starch in addition to mutant sources of maize starch, although the relationships between DSC and functional properties differ.

CONCLUSIONS

Genotypic differences in thermal properties of starch were found among the 25 inbreds grown in 1992 and 1993. With the exception of ΔH, inbred by year interactions existed among these materials. The high sensitivity of DSC in identifying differences in thermal properties of starch among nonmutant genotypes suggests that it may be a useful aid in plant breeding programs designed to select for desirable starch types.

Starches of inbreds grown in 1992 and selected for high and low T_p, R_n, and ΔH revealed that differences in these DSC values reflected differences in functional properties. Significant differences were observed for BEPT, viscosities, gel firmness, and gel stability. Several significant correlations existed between the DSC values T_o and T_p and values for viscosity peak temperature and viscosity at 95°C with texture analyzer values for gel firmness measured after one and seven days of storage at 4°C. These results suggest that thermal properties measured by DSC can be used to predict functional properties of starches among nonmutant sources of maize. Practical applications of DSC in breeding for improved starch characteristics might include the screening of nonmutant maize germ plasm or improvement of nonmutant maize populations through recurrent selection. Future studies are required to determine whether the magnitude of variation is of economic importance.

TABLE VI
Pasting Properties of Starches Measured from Selected Maize Inbreds Grown in 1992 by Amylography

Genotype	Pasting Onset (°C)	Peak Temperature (°C)	Brabender Viscosity Units (BU)					
			Peak Viscosity	At 95°C	95°C Hold	At 50°C	Set Back	50°C Hold
PI 303943	72.6	95.0	740	740	800	1,365	565	1,380
A641	73.4	93.6	820	820	680	1,320	640	1,300
ND246	82.0	89.9	990	960	900	1,320	420	1,205
Os420	77.0	94.2	870	865	760	1,380	620	1,355
A632	77.0	94.3	795	790	660	1,340	680	1,315
PI 186190	79.4	92.1	940	920	730	1,250	520	1,140
Mean	76.9	93.2	854	854	755	1,329	574	1,083
LSD[a]	NS[b]	2.6	84	93	125	NS	...	124

[a] Least significant difference ($\alpha = 0.05$).
[b] Not significant.

TABLE VII
Gel Strength Measurements After One and Seven Days of Storage for Starch Pastes from Selected Maize Inbreds Grown in 1992

Genotype	Firmness (g)		
	1 Day	7 Days	RF[a]
PI 303943	46.1	96.0	2.08
A641	41.6	54.1	1.30
ND246	63.9	71.8	1.12
Os420	38.8	40.1	1.03
A632	39.4	44.2	1.12
PI 186190	34.8	44.5	1.27
Mean	44.1	58.4	...
LSD[b]	3.5	65.2	...

[a]Ratio of gel firmness (7-day grams − Force/1-day grams − Force).
[b]Least significant difference ($\alpha = 0.05$).

TABLE VIII
Simple Correlations Among Differential Scanning Calorimetry (DSC) Parameter Means, Amylograph Points, and Gel Strengths for Starch of Six Selected Maize Inbreds Grown in 1992

	DSC Parameters				
	T_o[a] (°C)	T_p[b] (°C)	R_n[c] (°C)	ΔH[d] (cal/g)	R[e] (%)
Brabender values					
Pasting onset	0.62	0.71	0.20	−0.03	−0.17
Viscosity peak temperature	0.57	−0.92**[f]	−0.02	−0.25	−0.23
Peak viscosity	0.35	0.78	0.15	0.37	−0.14
95°C	0.49	0.86*	0.21	0.29	−0.09
95°C Hold	0.65	0.51	0.10	−0.25	0.26
50°C	0.24	−0.38	0.60	−0.68	−0.31
50°C Hold	−0.09	−0.62	0.35	−0.53	−0.24
Gel strengths					
1-day storage	0.84*	0.69	0.18	−0.31	0.38
7-day storage	0.83*	0.82*	0.64	−0.17	−0.23
RF[g]	−0.33	−0.55	−0.64	−0.13	0.66
Birefringence					
End point	0.84*	0.84*	0.29	−0.17	0.01
Range	−0.35	0.51	0.83	0.78	0.11

[a]Gelatinization onset.
[b]Gelatinization peak.
[c]Enthalpy of gelatinization.
[d]Gelatinization range.
[e]Retrogradation.
[f]*, ** = Significant at $P \leq 0.05$ and 0.01.
[g]Ratio of gel firmness.

LITERATURE CITED

BILIADERIS, C. G., MAURICE, T. J., and VOSE, J. R. 1980. Starch gelatinization phenomena studied by differential scanning calorimetry. J. Food Sci. 45:1669.

BROWN, P. 1966. Genetic control of starch granule development in maize endosperm. MS thesis. Department of Horticulture. Pennsylvania State University: University Park, PA.

BROCKETT, E., THOMPSON, D., DAVIS, T., and BOYER, C. 1988. Gelatinization characterization of starch from *du, wx, ae* and *ae wx* endosperm of sweet corn inbred Ia 5125. Starch/Staerke 40:175.

CAMPBELL, M. R, POLLAK, L. M., and WHITE, P. J. 1992. Effect of planting date on maize starch thermal properties by using differential scanning calorimetry. Cereal Foods World 37:57.

DAYNARD, T. B., and DUNCAN, W. G. 1969. The black layer and grain maturity in corn. Crop Sci. 9:473.

DONOVAN, J. W., LORENZ, K., and KULP, K. 1983. Differential scanning calorimetry of heat-moisture treated wheat and potato starches. Cereal Chem. 60:381.

HALLAUER, A. R., and MIRANDA, J. B. 1981. Heredity variance; Mating designs. Pages 45-114 in: Quantitative Genetics in Maize Improvement. Iowa State University Press: Ames, IA.

KATZ, F. R. 1991. Natural and modified starches. Pages 315-327 in: Biotechnology and food ingredients. I. Goldberg and R. Williams, eds. Van Nostrand Reinhold: New York.

KNUTSON, C.A. 1986. A simplified colorimetric procedure for determination of amylose in maize starches. Cereal Chem. 63:89.

KRUEGER, B. R., KNUTSON, C. A., INGLETT, G. E., and WALKER, C. E. 1987. A differential scanning calorimetry study on the effect of annealing on gelatinization behavior of corn starch. J. Food Sci. 52:715.

LI, J., BERKE, T. G., and GLOVER, D. V. 1994. Variation for thermal properties of starch in tropical maize germ plasm. Cereal Chem. 71:87.

SANDERS, E. B., THOMPSON D. B., and BOYER, C. D. 1990. Thermal behavior during gelatinization and amylopectin fine structure for selected maize genotypes as expressed in four inbred lines. Cereal Chem. 67:594.

SAS. 1990. SAS user's guide: Statistics. The Institute: Cary, NC.

SCHOCH, T. J. 1942. Fractionation of starch by selective precipitation with butanol. J. Am. Chem. Soc. 64:2957.

SHANNON, J. C, and GARWOOD, D. L. 1984. Genetics and physiology of starch development. Pages 25-86 in: Starch: Chemistry and Technology. R. Whistler, J. BeMiller, and E. Pachall, eds. Academic Press: Orlando, FL.

STEEL, G. D., and TORRIE, J. H. 1980. Procedures of Statistics: A Biometrical Approach, 2nd ed. McGraw-Hill: New York.

STEINKE, J. K., and JOHNSON, L. A. 1991. Steeping maize in the presence of multiple enzymes. I. Static batchwise steeping. Starch/Staerke 68:7.

STEVENS, D. J., and ELTON, G. A. H. 1971. Thermal properties of the starch/water system. Part I. Measurement of heat of gelatinization by differential scanning calorimetry. Starch/Staerke 23:8.

TAKAHASHI, S., MANANGAT, C., and SEIB, P. A. 1989. Acetylated and hydroxypropylated wheat starch: Paste and gel properties compared with modified maize and tapioca starches. Cereal Chem. 66:499.

WANG, Y., WHITE, P., and POLLAK, L. 1992. Thermal and gelling properties of maize mutants from the OH43 inbred line. Cereal Chem. 69:328.

WHITE, P. J., ABBAS, I. R., and JOHNSON, L. A. 1989. Freeze thaw stability and refrigerated storage retrogradation of starches. Starch/Staerke 41:176.

WHITE, P. J., ABBAS, I. R., POLLAK, L. M., and JOHNSON, L. A. 1990. Intra- and interpopulation variability of thermal properties of maize starch. Cereal Chem. 67:70.

WHITE, P. J., POLLAK, L. M., and BURKHART, S. 1991. Thermal properties of starches from corn grown in temperate and tropical environments. (Abstr.) Cereal Foods World 36:524.

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