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Weston B. Johnson
University of Nebraska-Lincoln

Wajira S. Ratnayake
University of Nebraska-Lincoln, wratnayake2@unl.edu

David S. Jackson
University of Nebraska - Lincoln, djackson1@unl.edu

Kyung-Min Lee
Texas A&M University

Timothy J. Herrman
Texas A&M University

See next page for additional authors

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Authors
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Weston B. Johnson,[1,2] Wajira S. Ratnayake,[3,4] David S. Jackson,[1,2] Kyung-Min Lee,[5] Timothy J. Herrman,[5]
Scott R. Bean,[6] and Stephen C. Mason[7]

ABSTRACT

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Dent corn (*Zea mays* L.) and sorghum (*Sorghum bicolor* L. Moench) sample sets representative of commonly grown hybrids and diverse physical attributes were analyzed for alkaline cooking performance. The influence of kernel characteristics including hardness, density, starch properties (thermal, pasting, and crystallinity), starch content, protein content, and prolamin content on alkaline cooking performance was also determined. Corn nixtamal moisture content was lower for hard, dense kernels with high protein contents; sorghum nixtamal moisture content was lower for kernels with low moisture contents and low starch relative crystallinities. Statistically significant ($P < 0.05$) regression equations showed that corn nixtamal moisture content was influenced by TADD (tangential abrasive dehulling device) index, kernel moisture content, starch content, and protein content; sorghum nixtamal moisture content was influenced by starch relative crystallinity, kernel moisture content, and abrasive hardness index. Pericarp removal was not strongly correlated with kernel characterization tests. Location (environmental) and hybrid (genetic) factors influenced most kernel characteristics and nixtamalization processing variables.

A major food use of corn (*Zea Mays* L.) and sorghum (*Sorghum bicolor* L. Moench) grain (kernel) is commercial production of tortillas, snack chips, and related foods through the alkaline cooking process known as nixtamalization (Serna-Saldivar et al 1988; Suhendro et al 1998; Taylor et al 2006). During nixtamalization, grain is cooked in a lime solution, allowed to steep for 12–16 hr, washed to remove loose pericarp, and stone-ground to produce masa (corn or sorghum dough) (Sahai et al 1999). Masa is then formed and cooked into desired end products.

Nixtamalization efficiencies are of primary concern to processors and depend on three factors: 1) optimizing degree of cook; 2) optimizing pericarp removal; and 3) minimizing dry matter loss (DML) (Shandera et al 1997). During nixtamalization, thermal transfer due to hydration results in partial gelatinization of starch, limited granule swelling, and disruption of starch crystalline structure (Mondragón et al 2004). Degree of cook is determined by the rate at which kernels absorb moisture and it affects masa texture and final product quality (Sahai et al 1999). Slower moisture absorption, which is associated with harder kernels, assures product consistency (Shandera et al 1997). Pericarp removal is the primary purpose of the washing step. Pericarp in masa foods can have an adverse affect on texture, color, and processing properties (Serna-Saldivar et al 1991). However, complete removal of pericarp is not always desirable due to decreased product yield. Grain solids loss during cooking, steeping, and washing increases waste water disposal costs and decreases end product yield (Sahai et al 2000). While these factors are primarily influenced by processing conditions, grain characteristics such as hardness, kernel composition, and starch properties are also critical (Sahai et al 2001a).

Corn and sorghum contain areas of both vitreous and opaque-soft endosperm. The vitreous endosperm is made up of polygonal-shaped starch granules tightly packed in a continuous protein matrix with dispersed protein bodies (Hoseney 1994). The opaque endosperm is made up of spherical starch granules loosely packed in a discontinuous protein matrix with no protein bodies (Hoseney 1994). While vitreousness is not synonymous with hardness, increased amounts of matrix protein and protein bodies have been associated with hardness (Chandrashekar and Mazhar 1999).

Hardness characteristics of corn and sorghum can be evaluated by measuring grinding performance with the Stenvert Hardness Tester (SHT) (Pomeranz et al 1985), resistance to abrasion with the Tangential Abrasive Dehulling Device (TADD) (Shandera et al 1997; Griess et al 2010), true density with a gas pycnometer, and bulk density by measuring the weight of a known volume of grain. Total protein content and the amount of prolamines (aqueous alcohol-soluble proteins) increase hardness (Chandrashekar and Mazhar 1999). Hardness has been associated with decreased DML and nixtamal moisture content (Pflugfelder et al 1988; Shandera et al 1997; Sahai et al 2001a). Harder kernels are often preferred by processors because they absorb moisture more slowly than softer kernels, resulting in decreased processing variability (Shandera et al 1997). Several researchers have concluded that softer kernels lose less pericarp during nixtamalization, but this relationship has not been definitively established (Serna-Saldivar et al 1991; Shandera et al 1997; Salinas et al 2003).

Starch content and functionality are important characteristics related to processing performance. Common starch characterization tests include using a Rapid Visco-Analyser (RVA) which measures pasting properties, differential scanning calorimetry (DSC) which measures thermal properties, by enthalpic transitions and X-ray diffraction which is used to measure the relative amounts of crystalline versus the amorphous forms (relative crystallinity) of starch. Sahai et al (1999) concluded that RVA and DSC were useful techniques in determining starch gelatinization properties relating to nixtamalization. In addition to starch gelatinization, DSC gives information about starch polymer associations by measuring the amount of energy required (enthalpy) to disrupt ordered structure (Tester and Karkalas 2001). Starch crystalline properties, as measured by X-ray diffraction, and thermal stability of structure, as measured by DSC, also are useful in predicting nixtamalization performance (Gomez et al 1989; Sahai et al 2001b; Mondragón et al 2004).

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[1] Department of Food Science and Technology, University of Nebraska-Lincoln, Lincoln, NE 68583-0919.

[2] Agricultural Research Division/NE Agricultural Experiment Station, University of Nebraska-Lincoln, Lincoln, NE 68583-0704.

[3] University of Nebraska-Lincoln, The Food Processing Center, 222 Food Industry Complex, Lincoln, NE 68583-0930.

[4] Corresponding author. Phone: (402) 472-2142. Fax: (402) 472-1693. E-mail: wratnayake2@unl.edu

[5] Office of the Texas State Chemist, Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77841-3160.

[6] USDA-ARS, Center for Grain and Animal Health Research, Manhattan, KS 66502. Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by the USDA implies no approval of the product to the exclusion of others that may also be suitable.

[7] Department of Agronomy and Horticulture, University of Nebraska-Lincoln, Lincoln, NE 68593-0915.

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Because kernel hydration largely determines the degree of cook during nixtamalization, measuring kernel hydration is desirable. RVA has been used successfully for this purpose. Almeida-Domiguez et al (1997) found RVA useful in predicting water uptake during nixtamalization. Whalen (1998) used RVA to relate water absorption to corn quality.

We hypothesized that kernel characterization tests were predictive of nixtamalization performance. This would aid in developing better methods for selecting grain for processing. The approach was to develop predictive equations based on characterization tests or to create classification schemes that would allow processors to more accurately assign a particular lot of grain for nixtamalization or some other use. The objective of this study was to identify relationships between corn and sorghum kernel characteristics and nixtamalization performance.

MATERIALS AND METHODS

One hundred dent corn samples consisting of 70 hybrids grown in the states of Illinois, Indiana, Iowa, Kansas, Kentucky, and Nebraska in the United States and 35 sorghum samples consisting of 25 hybrids grown in Manhattan (Kansas), Ithaca (Nebraska), College Station (Texas), and Granger (Texas) were used in this study. Samples were randomly selected from larger sets of 240 corn and 100 sorghum samples. Sample sets were reduced in size (240 to 70, 100 to 35) to create working sample sets of reasonable size for the available resources while maintaining the genetic and environmental diversity of the larger sets. Measured kernel characteristics were first converted into principal component scores using WINSI II software (v.1.0, Foss NIR Systems, Infrasoft International, Silver Spring, MD). Moisture, protein, oil, and starch content were measured using near-infrared transmittance (NIT) (Infratec 1229, Foss North America, Eden Prairie, MN). Density was measured with a multipycnometer (model MVP-6DC, Quantachrome, Boynton Beach, FL). Hardness-associated properties of sorghum consisting of Single Kernel Characterization System (SKCS) hardness, diameter, and weight (Bean et al 2006), abrasive hardness index (Oomah et al 1981), and total protein content (Approved Method 46-30.01, AACC International 2010) were also measured. The first four principal components for corn spectral data accounted for 95% of total variability and were used to create nine clusters of analogous samples. The first two principal components for sorghum hardness-associated data accounted for 73.4% of total variability and were used to create six clusters of analogous samples. Results from maximum likelihood factor analysis indicated that two principal components were sufficient for sample selection. One hundred corn samples were selected using the neighborhood H concept by which the sample with the most neighbors is selected and its neighbors are removed. Sorghum samples were equally selected from each cluster and location; they included both pigmented and nonpigmented samples. The identified corn samples (100) and sorghum samples (35) were stored at –20°C before analysis and processing.

Sample Preparation

Finely ground corn and sorghum flours were prepared by grinding whole kernels through a cyclone sample mill with a 1-mm mesh screen (model 3010-030, Udy, Fort Collins, CO). Coarsely ground flour was produced by grinding whole kernels with a micro-hammer mill (model V equipped with a 2-mm screen, Glen Mills, Maywood, NJ). Whole kernels were used for nixtamalization and physical tests; finely ground flour was used for starch and protein tests, and coarsely ground flour was used for hydration rate test.

Nixtamalization

Nixtamalization of corn and sorghum was adapted from Sahai et al (1999). Samples (200 g) were placed in perforated nylon bags and added to a steam-heated kettle containing a 1% (corn weight basis) lime solution (4:1 water-to-grain ratio) preheated to 94°C. After grain addition, the temperature was brought back to 94°C in 5 min, and cooking continued for an additional 25 min. The lime solution was then quenched with half of the original volume of cold water and steeped 18 hr. The samples were removed from the steep solution and washed with ≈16 L of cold water. Nixtamal moisture was determined by Approved Method 44-15.02 (AACC International 2010). Percent dry matter loss was determined by subtracting the nixtamal dry weight from the dry weight of the original sample. Degree of pericarp removal was determined by staining 8–10 kernels with May-Gruenwald solution and subjectively rating the percent pericarp removed (Serna-Saldivar et al 1991).

Physical Properties

Corn hardness properties were determined using the Tangential Abrasive Dehulling Device (TADD) (model 4E-220, Venebles Machine Works, Saskatoon, SK, Canada) (Shandera 1997) and Stenvert hardness tester (SHT) (GlenMills model V equipped with a 2-mm screen) run at 3,600 rpm (Pomeranz et al 1985). True density for corn and sorghum was measured with a multipycnometer (model MVP-6DC, Quantachrome, Boynton Beach, FL). Bulk density (test weight) was determined by measuring the weight of a known volume of grain using an approved test weight apparatus (USDA 1990) (Seedburo, Chicago, IL).

Sorghum hardness-associated properties consisting of SKCS hardness, kernel diameter, and kernel weight were measured as described by Bean et al (2006) using the Perten 4100 SKCS (Perten Instrument, Springfield, IL). Abrasive hardness index (AHI) was determined by progressively abrading the sorghum for 1-min intervals using a TADD (model 4E) equipped with an 80-grit abrasive pad. The abraded sorghum was weighed after 1, 2, 3, and 4 min (Oomah et al 1981). A best-fit line was calculated from a plot of percent kernel removed versus time. AHI was calculated as the inverse of the gradient of the best-fit line and was reported as the time in seconds required to remove 1% of the grain.

Starch Properties

Total starch content of finely ground corn and sorghum flours was determined using a biochemistry analyzer (model 2700 Select, YSI, Yellow Springs, OH, Application Note 322). Samples were first gelatinized by adding 10 mL of 2*N* sodium hydroxide to 0.5-g samples in 70-mL capacity test tubes and heating for 25 min at 94°C and adjusted to pH 4.5 with 2*N* hydrochloric acid and 10 mL of acetate buffer (9.1 g of sodium acetate [Sigma Chemical, St. Louis, MO] and 44.6 mL of glacial acetic acid [Fischer Chemical, Pittsburg, PA] diluted to 1 L). Amyloglucosidase EC 3.2.1.3 (300 units, Sigma) was added and the slurry was incubated for 70 min at 50°C. Enzymatic hydrolysis was stopped by adding 5 mL of 25% trichloroacetic acid (Fischer) and the solution was transferred to a 100-mL volumetric flask and filled to volume with phosphate buffer (40 g of sodium phosphate monobasic, anhydrous [Fisher] and 10 g of sodium phosphate dibasic, anhydrous [Sigma] diluted to 1 L). Glucose concentration was measured with the biochemistry analyzer fitted with an immobilized glucose oxidase EC 1.1.3.4 membrane. Percent total starch (dry basis) was calculated by multiplying the glucose content by a conversion factor of 0.9 (1.1 g of glucose = 1.0 g of starch) expressed as a ratio to the initial sample weight. This value was corrected for the free glucose contained in the sample and amyloglucosidase reagent using appropriate blanks.

Crystalline properties of corn and sorghum starches isolated during wet milling were analyzed using a diffractometer (D-Max/B, Rigaku-Denki, Tokyo, Japan) with Cu-Kα radiation at a voltage of 40 kV at 30 mA. Starches were wetted with ethanol and mounted on a zero background flat-mount quartz sample holder and scanned from $2\theta = 5–30°$ at 3°/min. Diffraction patterns were analyzed using X-ray diffraction peak analysis software (Eva v.9.0, Bruker-AXS, Karlsruhe, Germany). Background was eliminated by connecting a baseline from the start and end points. A smooth curve was fitted under the diffraction peaks to separate crystalline and amorphous regions. Percent relative crystallinity was calculated by dividing the area of the diffraction curve above the smooth curve by the area of the diffraction curve above the baseline (Roe 2000).

Thermal properties of finely ground corn and sorghum flours were determined using DSC (Pyris 1, Perkin Elmer, Norwalk, CT). Samples ($\approx$10 mg) were weighed into stainless-steel sample pans and 55 µL of water was added. Pans were sealed, stored at room temperature overnight, and heated from 25 to 110°C at 10°C/min. Onset temperature, peak temperature, end temperature, and enthalpies were determined. Enthalpies were reported on a dry starch basis (J/g).

Pasting properties of finely ground corn and sorghum flours were determined using RVA (RVA-4, Newport Scientific, Warriewood, Australia). Flour samples (4 g, corrected to 14% moisture) were combined with 25 mL of water in an aluminum canister with a plastic paddle and mixed for 5 min at 160 rpm. The heating profile went from 50 to 95°C in 5 min, held at 95°C for 3 min, cooled to 50°C in 5 min, and held at 50°C for 5 min. Viscosity was measured in centipoise (cP) units. Pasting temperature (°C), peak viscosity (cP), peak temperature (°C), break down (cP), and setback (cP) were determined.

Hydration rate of coarsely ground corn and sorghum flour was determined using RVA. This procedure was based on a method developed by Whalen (1998) to detect differences in corn quality. Coarsely ground flour (8 g, corrected to 14% moisture) was combined with 25 mL of water in an aluminum cup with a plastic paddle. The paddle speed was set at 160 rpm for the entire heating profile except for an initial mix cycle of 10 sec, where it was set at 500 rpm. The heating profile held at 50°C for 5 min, ramped to the pasting temperature (determined previously using finely ground flour) within 3 min, and held at that temperature for 10 min. The hydration rate was determined as the change in viscosity over time from the baseline to a point on the curve corresponding to 5 min after the pasting temperature.

Protein Analysis

Total protein was determined by nitrogen combustion using a nitrogen determinator (Leco FP-528, St. Joseph, MI) according to Approved Method 46-30.01 (AACC International 2010).

Prolamin (kafirin and zein) contents (db) were determined by removing albumin and globulin proteins and extracting kafirin or zein with 60% tertiary-butanol + 5% β-ME + 0.5% sodium acetate (Bean et al 2000). A Coomassie assay kit (Plus Protein, Pierce, Perbio Science, Rockford, IL) was used for kafirin and zein quantification. Supernatant (50 µL) obtained from extraction was transferred into microcentrifuge tubes and 1.5 mL of Coomassie reagent was added. The solution was mixed (Vortexgenie 2, Scientific Instruments, Bohemia, NY). Absorbance was measured at 595 nm using a spectrophotometer (Beckman DU 530, Fullerton, CA). A standard curve was generated using commercial zein or laboratory purified kafirins (0.2 to 8 mg/mL) to measure the protein concentration in unknown samples. A linear response ($y = 0.150x - 0.0041$ for corn, and $y = 0.2196x + 0.0303$ for sorghum) across the standard curve was seen with $R^2 = 0.99$ between concentration and absorbance at 595 nm.

Statistical Analyses

Nixtamalization was performed in triplicate for all corn and sorghum samples. All kernel characterization tests were performed in duplicate except for kernel moisture and corn TADD tests performed in triplicate. Simple correlations among all quality factors and processing variables were computed using Proc Corr (v.8.02, SAS Institute, Cary, NC). Stepwise and multiple regression analysis methods were used to develop equations for predicting nixtamalization performance with significant independent variables (NCSS 2004, Visual Statistical Systems, Kaysville, UT). Independent variables for regression equations are listed in order of greatest effect on R^2 values. Tables I and II show regression equations with $R^2 > 0.40$ containing significant ($P < 0.05$) independent variables. One-way ANOVA was used to assess differences between locations and hybrids for a subset of five hybrids grown in three locations. Multiple mean comparisons between hybrids and locations were performed using the LSD test ($P < 0.05$).

RESULTS AND DISCUSSION

Grain Characterization

Summary data of kernel characterization tests performed on corn and sorghum are shown in Table III. Wide ranges (difference between highest and lowest values observed for the set of samples) and high coefficients of variations for both corn and sorghum variables indicate diversity in the selected sample sets.

Nixtamalization Results

Table IV shows the nixtamalization results for both corn and sorghum. Ranges and coefficients of variation for dry matter loss and nixtamal moisture content were larger for sorghum than for corn. A nixtamal moisture content of 48–50% is recommended to produce masa with acceptable plasticity, cohesiveness, and machin-

TABLE I

Corn Wet Milling and Nixtamalization Stepwise Regression Equations for $R^2 > 0.40$

Wet Milling	Starch yield (%) = 0.0013 (RVA breakdown) + 0.600 (starch content) – 0463 (RVA pasting temperature) – 10.70 (true density) + 70.56	($P < 0.05$, $R^2 = 0.76$)
	Wash solids yield (%) = 0.407 (RVA pasting temperature) – 0.192 (starch content) – 0.0013 (RVA breakdown) – 0.136 (kernel moisture content) –1.94	($P < 0.05$, $R^2 = 0.64$)
	Bran yield (%) = –0.276 (starch content) – 0.0004 (RVA peak viscosity) + 0.104 (kernel moisture content) + 29.62	($P < 0.05$, $R^2 = 0.46$)
	Steep solids yield (%) = 11.33 (true density) – 0.1069 (DSC enthalpy) – 8.09	($P < 0.05$, $R^2 = 0.56$)
Nixtamalization	Nixtamal moisture (%) = 0.276 (TADD loss) + 0.468 (kernel moisture content) – 0.255 (starch content) – 0.301 (protein content) + 45.21	($P < 0.05$, $R^2 = 0.64$)

TABLE II

Sorghum Wet Milling and Nixtamalization Stepwise Regression Equations for $R^2 > 0.40$

Wet Milling	Starch yield (%) = –2.16 (protein content) + 0.0020 (RVA hydration rate) + 85.25	($P < 0.05$, $R^2 = 0.58$)
	Steep solids yield (%) = –0.126 (abrasive hardness index) + 0.0222 (SKCS hardness) + 0.132 (protein content) + 4.48	($P < 0.05$, $R^2 = 0.54$)
	Bran yield (%) = 1.84 (protein content) – 0.200	($P < 0.05$, $R^2 = 0.46$)
Nixtamalization	Nixtamal moisture (%) = 1.65 (relative crystallinity) – 1.21 (abrasive hardness index) + 47.07	($P < 0.05$, $R^2 = 0.63$)

ablilty (Gomez et al 1991). Mean nixtamal moisture content for corn in this study was slightly higher than recommended levels, but was very acceptable for distinguishing differences between samples. DML and degree of pericarp removal were similar to other results reported (Serna-Saldivar et al 1991; Sahai et al 2000). Corn samples had acceptable nixtamal moisture contents (48–50%), good pericarp removal (>50%), and reasonably low dry matter loss (<5%): Syngenta WX7663, Pioneer 33P67, 33R77, 34A31, 3497, 36B08, Horizon 7373, Garst 8383, and Mycogen 2A812. Several sorghum hybrids had nixtamal moisture contents of 48–50% with DML <5% (03bott121, 02CS5864*5863, 03bott214, 96C1642*1643, 02CS5804*5803, ATX399*KS115, 03bott092, and 99CS2118*2117).

Nixtamal Moisture Content

The extent to which kernels hydrate during the nixtamalization process affects degree of cook (Sahai et al 1999, 2001a). Degree of cook influences masa texture, ease of processing, and final product quality (Sahai et al 1999).

For corn, three RVA parameters were correlated with corn nixtamal moisture content: peak viscosity, breakdown, and hydration rate (Table V). Nixtamal moisture content was lower for hard, dense corn kernels with high protein content. Figure 1 shows RVA profiles of representative high (56%) and low (47%) nixtamal moisture content samples. Both peak viscosity and breakdown (peak viscosity minus trough viscosity) were lower for samples that absorbed less water during nixtamalization (profile B in Fig. 1). Lower peak viscosity may be due to either lower starch content or relatively lesser degree of starch granule hydration and swelling during pasting. This observation was in agreement with Salinas et al (2003). The higher protein content of kernels could cause less moisture absorption during nixtamalization, likely contributing to low RVA peak viscosity. This is potentially due to the "inhibitory effect" of increased matrix protein that could reduce the degree of starch granule hydration and swelling. It is also likely that this inhibitory effect is also responsible for the apparent increase in pasting temperatures observed for samples that absorbed less moisture during nixtamalization (e.g., profile B in Fig. 1) though the correlation between pasting temperature and nixtamal moisture content was only $r = -0.269$. Figure 2 shows RVA hydration rate profiles of the same low and high nixtamal moisture samples in Fig. 1. An important difference between the standard RVA profiles in Fig. 1 and the hydration rate profiles in Fig. 2 was the size of the flour particles. The flour used to measure hydration rate was ground with a hammer mill. The temperature was ramped and held at the pasting temperature determined previously for that particular sample using the standard RVA profile and held for several minutes. This had the effect of magnifying hydration differences between samples as larger particles took longer to hydrate and gelatinize. Viscosity increases were likely inhibited by the presence of increased matrix protein in larger particles as harder kernels tend to yield larger particles when grinding with the hammer mill (Pomeranz et al 1985) (profile B). Kernel hydration during cooking (measured as nixtamal moisture content) was also hindered by starch-protein associations. Because hard, dense kernels with higher protein content absorbed less moisture during nixtamalization, had lower RVA peak viscosities and breakdowns, and had decreased RVA hydration rates (Table V), RVA may be useful in screening samples for use in nixtamalization.

For sorghum, nixtamal moisture content increased with DSC enthalpy and starch relative crystallinity (Table V). This may be due to some fundamental difference in starch structure that affects hydratability during cooking or strong interaction between starch and protein in sorghum.

TABLE III

Simple Statistics of Corn and Sorghum Characterization Test Results

Test Variable	Corn Mean ± SD[a]	Corn Range	Corn %CV[b]	Sorghum Mean ± SD[a]	Sorghum Range	Sorghum %CV[b]
Moisture (%)	14.2 ± 1.8	10.4 – 17.4	12.4	11.8 ± 1.4	9.3 – 14.1	11.7
Protein (%db)[c]	7.7 ± 1.0	4.2 – 10.0	12.6	12.2 ± 1.1	10.3 – 14.9	8.9
Prolamin (%tp)[d]	46.7 ± 7.3	32.6 – 76.6	15.7	61.8 ± 10.9	42.5 – 81.8	17.6
Starch (%db)[e]	72.2 ± 1.6	67.6 – 74.8	2.2	71.3 ± 2.0	66.4 – 74.8	2.8
Starch relative crystallinity (%)	14.7 ± 0.7	12.4 – 16.2	4.7	15.6 ± 0.7	14.2 – 17.8	4.4
True density (g/cm^3)	1.321 ± 0.027	1.225 – 1.371	2.1	1.412 ± 0.013	1.390 – 1.439	0.9
Test weight (g/cm^3)	57.9 ± 2.5	51.1 – 62.4	4.3			
100 kernel weight (g)	30.1 ± 4.6	18.2 – 41.7	15.3			
SHT time of grind	20.6 ± 4.5	12.5 – 32.5	21.8			
TADD (% remaining)	24.0 ± 6.0	7.3 – 39.0	24.7			
SKCS kernel weight (mg)				30.5 ± 6.7	22.6 – 49.6	22.0
SKCS kernel diameter (mm)				2.0 ± 0.3	1.5 – 2.7	16.3
SKCS hardness scale				82.5 ± 8.4	63.6 – 94.2	10.2
Abrasive hardness index[f]				14.1 ± 2.3	10.6 – 20.4	16.2

[a] Standard deviation.
[b] Percent coefficient of variation.
[c] Percent dry basis.
[d] Percent of total protein.
[e] Relative scale based on soft wheat = 50, hard wheat = 100.
[f] Seconds required to abrade 1% of the kernel with TADD.

TABLE IV

Nixtamalization Results for Corn and Sorghum

Yield	Corn Mean ± SD[a]	Corn Range	Corn %CV[b]	Sorghum Mean ± SD[a]	Sorghum Range	Sorghum %CV[b]
Dry matter loss (%)	5.0 ± 1.2	1.1 – 8.3	24.9	2.8 ± 1.4	0.3 – 6.8	48.8
Nixtamal moisture content (%)	52.1 ± 2.0	47.0 – 57.7	3.8	49.6 ± 2.5	45.8 – 59.5	5.0
Pericarp removal (%)	37.0 ± 10.1	16.7 – 66.7	16.2	–	–	–

[a] Standard deviation.
[b] Percent coefficient of variation.

Stepwise regression analysis for corn indicated that nixtamal moisture content was significantly dependent on (listed in order of contribution to R^2) TADD hardness, kernel moisture content, starch content, and protein content ($P < 0.05$, $R^2 = 0.64$)

$$\text{Corn nixtamal moisture content} = 0.276\ (\text{TADD}) + 0.468\ (\text{kernel moisture content}) - 0.255\ (\text{starch content}) - 0.301\ (\text{protein content}) + 45.21 \tag{1}$$

Close protein-starch associations in hard endosperm, as shown by increased protein content and TADD hardness, might have inhibited moisture absorption during nixtamal cooking. Kernels also appeared to hydrate easier during cooking if they had higher moisture content to begin with.

Stepwise regression analysis for sorghum indicated that nixtamal moisture content was significantly dependent on (listed in order of contribution to R^2) starch relative crystallinity, kernel moisture content, and abrasive hardness index ($P < 0.05$, $R^2 = 0.63$)

$$\text{Sorghum nixtamal moisture content} = 1.65\ (\text{starch relative crystallinity}) - 1.21\ (\text{kernel moisture content}) - 0.634\ (\text{abrasive hardness index}) + 47.07 \tag{2}$$

As shown by simple correlations between nixtamal moisture content and sorghum starch relative crystallinity, the amount of starch ordered structure may affect moisture absorption during nixtamalization. Kernel hardness (abrasive hardness index) inhibited kernel hydration during cooking.

DML and Pericarp Removal

Correlation analysis and stepwise regression analysis revealed no significant ($P < 0.05$) relationships between corn pericarp removal and any kernel characterization test (Table V), confirming the results of Shandera et al (1997). Serna-Saldivar et al (1991) found only a poor negative correlation between pericarp removal and hardness ($r = -0.32$). Pericarp removal is likely influenced by pericarp properties such as thickness and resistance to alkali steeping that were not measured in this study. Kernel characterization tests used in these studies did not give much information about pericarp properties.

Sahai et al (2001a) used correlation and stepwise regression analysis to evaluate DML. They reportedly found no significant corre-

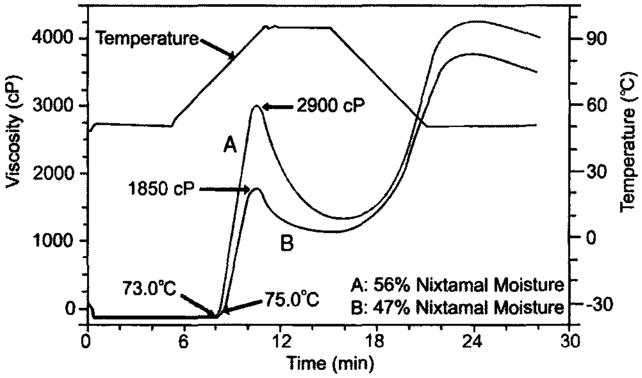

Fig. 1. RVA profiles of corn with high and low nixtamal moisture contents.

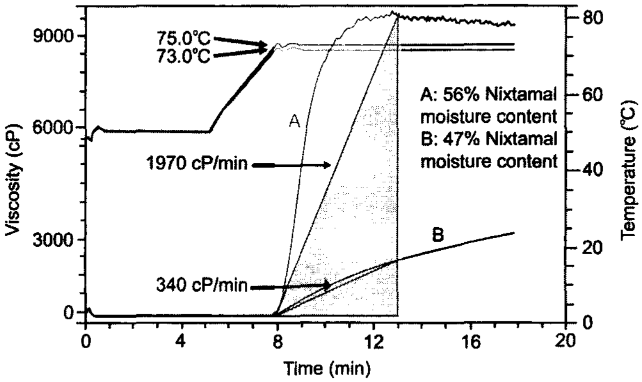

Fig. 2. RVA hydration rate profiles of corn samples containing high and low nixtamal moisture contents.

TABLE V
Corn and Sorghum Nixtamalization Performance Correlated with Kernel Characterization Tests[a]

	Corn			Sorghum	
Physicochemical Tests	**Dry Matter Loss**	**Nixtamal Moisture**	**Pericarp Removal**	**Dry Matter Loss**	**Nixtamal Moisture**
Moisture content	–0.248	ns	ns	ns	–0.466
Protein content	ns	–0.307	ns	ns	ns
Prolamin content	ns	ns	ns	ns	ns
Starch content	ns	ns	ns	ns	ns
Starch relative crystallinity	ns	ns	ns	0.375	0.583
True density	ns	–0.614	ns	ns	ns
Test weight	ns	ns	ns		
100 kernel weight	ns	ns	ns		
SHT time of grind	ns	ns	ns		
TADD[b]	ns	–0.677	ns		
SKCS kernel weight				ns	ns
SKCS kernel diameter				ns	ns
SKCS hardness scale[c]				ns	ns
Abrasive hardness index[d]				ns	ns
RVA					
Onset temperature	ns	ns	ns	ns	0.429
Peak temperature	ns	ns	ns	ns	0.386
Enthalpy	ns	0.389	ns	ns	0.562
End temperature	ns	–0.268	ns	ns	ns
DSC					
Initial pasting temperature	ns	–0.269	ns	ns	ns
Peak viscosity	ns	0.523	ns	ns	ns
Breakdown	ns	0.570	ns	ns	ns
Setback	ns	ns	ns	ns	–0.337
Hydration rate	ns	0.584	ns	ns	ns

[a] Correlation value r at $P < 0.05$ level.
[b] Tangential Abrasive Dehulling Device.
[c] Relative scale based on soft wheat = 50, hard wheat = 100.
[d] Seconds required to abrade 1% of the kernel with TADD.

lations between kernel characteristics and DML. Stepwise regression analysis showed that processing conditions had a major impact on DML while DML was only slightly affected by kernel characterization tests (breakage susceptibility and hardness index). In the current study, stepwise regression analysis revealed no relationship between DML and corn and sorghum characterization tests. DML was only slightly correlated with corn moisture content and sorghum starch relative crystallinity (Table V). The results suggest that DML measurements may be confounded by the presence of pericarp (Pflulgfelder et al 1988) because pericarp removal is difficult to predict.

Genetic and Environmental Effects

A major goal of this study was to assemble corn and sorghum sample sets representative of the genetic and environmental diversity of grain actually available for use by industry. The study, however, was not specifically designed to reveal genetic and environmental differences. Nevertheless, it was possible to separate out five sorghum hybrids that were grown near Ithaca, Nebraska, Manhattan, Kansas, and College Station, Texas, to partially evaluate genetic and environmental effects.

The five sorghum hybrids grown in Texas, Nebraska, and Kansas varied ($P < 0.05$) in starch contents, SKCS hardnesses, SKCS kernel weights, SKCS kernel diameters, RVA setbacks, and RVA hydration rates (Table VI). Hybrids also differed significantly in nixtamal moisture content but not dry matter loss. Average nixtamal moisture contents and dry matter losses were the not different among the growth locations (Table VII). A high variation was observed in SKCS kernel weight and diameter; the lowest values in 02CS5804*5803 and the highest values in ATX399*KS115. Texas-grown sorghum had more protein compared to Nebraska and Kansas sorghum. Abrasive hardness index was also higher for Texas-grown sorghum. Deposition of starch was restricted when growth temperatures are elevated (Tester and Karkalas 2001). Climatic data from May to August 2003 for weather observation stations near Ithaca, Nebraska, Manhattan, Kansas, and College Station, Texas is shown in Table VIII (National Climatic Data Center. Available online at http://www7.ncdc.noaa.gov/SerialPublications/LCDPubs?action=getstate&LCD=hardcode). Average daily

TABLE VII
Means of Location × Sorghum Characterization and Wet Milling Results for Five Sorghum Hybrids Grown at Three Locations[a]

	Location		
Variable	Kansas	Nebraska	Texas
Moisture (%)	11.2a	13.9b	11.2a
Protein (%db)[b]	11.5a	12.2a	14.0b
Prolamin (%db)[c]	64.7a	70.2a	47.6b
Starch (%db)[b]	72.1a	72.3a	69.5a
Starch relative crystallinity (%)	15.5a	15.6a	15.7a
True density (g/cm^3)	1.419a	1.417a	1.404a
SKCS[d] kernel weight (mg)	32.0a	31.7a	34.8a
SKCS kernel diameter (mm)	2.0a	1.9a	2.3a
SKCS hardness[e]	81.9a	85.3a	82.4a
Abrasive hardness index[f]	13.5ab	12.6a	15.1b
DSC			
Onset temperature (°C)	70.1a	70.3a	74.5b
Peak temperature (°C)	76.8a	77.1a	79.6b
Enthalpy (J/g)	6.12a	6.12a	5.3b
End temperature (°C)	84.8a	84.6a	87.6b
RVA			
Pasting temperature (°C)	75.4a	78.6b	80.7b
Peak viscosity (cP)	2,738a	2,410b	1,516c
Breakdown (°C)	1,463a	1,178b	340c
Setback (cP)	2,377a	2,639a	2,122a
Hydration rate (cP/min)	759a	1,048a	757a
Dry matter loss (%, db)[b]	3.0a	2.6a	3.6a
Nixtamal moisture content (%)	49.4a	48.5a	49.3a

[a] Values followed different letters are significantly different at $P < 0.05$.
[b] Percent dry basis.
[c] Percent of total protein.
[d] Single kernel characterization system.
[e] Relative scale based on soft wheat = 50, hard wheat = 100.
[f] Seconds required to abrade 1% of the kernel using TADD.

TABLE VI
Means of Hybrid × Sorghum Characterization and Nixtamalization Results for a Subset of Five Sorghum Hybrids Grown at Three Locations[a]

	Hybrid				
Variable	03bott121	03bott214	02CS5864*5863	02CS5804*5803	ATX399*KS115
Moisture (%)	12.2a	12.1a	12.0a	11.8a	12.2a
Protein (%db)[b]	12.9a	12.4a	12.2a	12.0a	13.3a
Protein (%db)[c]	64.2a	56.8a	56.9a	64.0a	62.2a
Starch (%db)[b]	70.2ab	71.4ab	73.3a	72.2ab	69.3b
Starch relative crystallinity (%)	15.5a	15.3a	15.4a	15.9a	15.8a
True density (g/cm^3)	1.420a	1.398b	1.425a	1.420a	1.404b
SKCS[d] kernel weight (mg)	36.0b	28.4a	26.0a	24.8a	48.9c
SKCS kernel diameter (mm)	2.40a	1.90b	1.68b	1.74b	2.62a
SKCS hardness[e]	88.2a	78.4b	94.0a	88.3a	67.1c
Abrasive hardness index[f]	13.6a	12.2a	14.5a	14.6a	13.9a
DSC					
Onset temperature (°C)	71.7a	71.3a	71.4a	72.1a	71.7a
Peak temperature (°C)	78.0a	76.9a	77.8a	79.1a	77.5a
Enthalpy (J/g)	5.6a	5.7a	5.8a	6.4a	5.8a
End temperature (°C)	85.8a	84.9	85.5a	87.2a	85.2a
RVA					
Pasting temperature (°C)	79.3a	79.9a	78.2a	76.4a	77.5a
Peak viscosity (cP)	2,086a	2,105a	2,364a	2,282a	2,270a
Breakdown (°C)	902a	882a	1,103a	1,003a	1,079a
Setback (cP)	2,460ab	2,868a	2,272b	2,086b	2,211b
Hydration rate (cP/min)	968a	1,105a	988a	259b	953a
Dry matter loss (%db)[b]	3.4a	3.8a	3.1a	2.7a	2.6a
Nixtamal moisture content (%)	47.9a	50.0a	48.9ab	49.5ab	49.0ab

[a] Means with different letters are significantly different ($P < 0.05$) within the same row.
[b] Percent dry basis.
[c] Percentage of total protein.
[d] Single kernel characterization system.
[e] Relative scale based on soft wheat = 50, hard wheat = 100.
[f] Seconds required to abrade 1% of the kernel using TADD.

TABLE VIII
Climatic Data for Weather Observation Stations Near Sorghum Growing Locations (2003)

	College Station, TX				Topeka, KS				Lincoln, NE			
	May	June	July	August	May	June	July	August	May	June	July	August
Avg maximum daily temp (°F)	88.6	90.7	92.2	95.1	75.7	82.7	94.3	93.8	72.2	80.1	92.1	90.6
Avg minimum daily temp (°F)	68.6	71.4	73.9	74.5	52.7	61.4	70.0	68.1	46.6	57.9	66.0	64.8
Total monthly precipitation (in.)	0.6	6.6	4.1	4.5	3.7	3.7	0.7	6.3	3.6	6.8	1.4	1.1

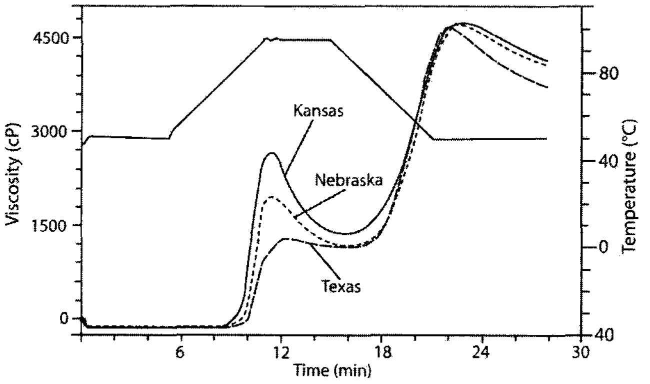

Fig. 3. RVA profiles of identical sorghum hybrids grown at different locations.

maximum temperature from May to August was 5.0°F higher in Texas compared to Kansas and 7.9°F higher in Texas compared to Nebraska. Average daily minimum temperature was 9.1°F higher in Texas compared to Kansas and 13.3°F higher in Texas compared to Nebraska. Total precipitation from May to August 2003 was 0.35 in. higher in Texas compared to Kansas and 0.71 in. higher in Texas compared to Nebraska.

Elevated growing season temperature likely contributed to the decreased starch content, increased protein content, and increased hardness found in Texas sorghum, similar to results of Griess et al (2010), who found lower yield, higher starch, lower protein, and harder kernels in the most stressful dryland environments than in irrigated and less stressful dryland environments.

Elevated growth temperature has also increased gelatinization temperature and enthalpy (Tester and Karkalas 2001). Gelatinization temperature (as measured by DSC peak temperature and RVA pasting temperature) and DSC enthalpy were higher for Texas sorghum (Table VII). Figure 3 shows RVA pasting profiles for identical hybrids grown in Texas, Kansas, and Nebraska. Pasting temperature was highest for Texas sorghum and lowest for Kansas sorghum; peak viscosity was lowest for Texas sorghum and highest for Kansas sorghum. Protein contents for the particular Texas, Nebraska, and Kansas sorghum samples shown in Fig. 3 were 14.7, 11.7, and 10.8%, respectively. Increased endosperm matrix protein may have interfered with starch hydration and swelling (Ratnayake et al 2007).

CONCLUSIONS

Hard, dense corn kernels with high protein contents were more desirable for nixtamalization due to lower nixtamal moisture contents; sorghum nixtamal moisture content was lower for kernels with high moisture content and low starch relative crystallinity. Regression equations for both corn ($R^2 = 0.76$, $P < 0.05$) and sorghum ($R^2 = 0.58$, $P < 0.05$) may be useful in estimating nixtamal moisture content.

While laboratory nixtamalization is still the best method for predicting industrial nixtamalization performance, individual characterization tests may be useful in screening samples for use in nixtamalization. TADD hardness, RVA, starch content, and protein content appear to be the best characterization tests for predicting corn nixtamal moisture content. Measuring total kernel starch content requires skill and experience, making it less practical for use in estimating nixtamal moisture content. Due to simple operation and relatively rapid response, total protein, TADD hardness, and RVA tests may be the most practical and useful tests for predicting industrial nixtamal moisture content.

A subset of five sorghum hybrids grown in three locations showed that genetic and environmental factors influenced most kernel characteristics. Nixtamalization processing variables were not significantly different when separated by growing location.

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