

**PREDICTION OF TORTILLA QUALITY USING MULTIVARIATE
MODELING OF KERNEL, FLOUR, AND DOUGH PROPERTIES**

A Dissertation

by

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ABSTRACT

Advances in high-throughput wheat breeding techniques have resulted in the need for rapid, accurate and cost-effective means to predict tortilla making performance for larger numbers of early generation wheat lines. Currently, the most reliable approach is to process tortillas. This approach is laborious, time consuming, expensive and requires large sample size.

This study used a multivariate discriminant analysis to predict tortilla quality using kernel, flour and dough properties. A discriminant rule (suitability = diameter > 165mm + day 16 flexibility score >3.0) was used to classify wheat lines for suitability in making good quality tortillas. One hundred eighty seven hard winter wheat (HWW) varieties from Texas were evaluated for kernel (hardness, diameter, and weight), flour (protein content, fractions and composition), dough (compression force, extensibility and stress relaxation from TA-XT2i) and tortilla properties (diameter, rheology and flexibility).

The first three principal components explained 58% of variance. Multivariate normal distribution of the data was determined (Shapiro-Wilk $p > 0.05$). PCA identified significant correlation between stress relaxation force and rollability.

Canonical correlation analysis revealed significant correlation between kernel and tortilla properties ($\hat{\rho} = 0.75$), kernel diameter and weight contributed the highest to this correlation. Flour and tortilla properties were highly correlated ($\hat{\rho} = 0.74$). Glutenin to Gliadin ratio (GGratio), IPP and peak time contributed highest to this correlation and

can explain > 60% of variability in tortilla texture (force, distance and work to rupture). The second canonical variate of flour properties is a measure of flour protein content and can explain 26% of the variability in tortilla rollability. Dough and tortilla properties were significantly correlated ($\hat{p} = 0.82, 0.68, 0.54, 0.38$ and 0.29). Dough stress relaxation force after 25 seconds is negatively correlated with tortilla diameter ($r = -0.73$).

Kernel hardness, diameter and weight are the best predictors of tortilla texture after 16 days. Glutenin to gliadin ratio and IPP contributed significantly to tortilla texture. This is the first study to identify the contribution of protein content on tortilla rollability score. Dough extensibility can explain 37% of tortilla rollability. Stress relaxation is the best predictor of tortilla diameter. Tortilla quality variation is attributed to kernel, flour, and dough properties. Logistic regression and stepwise variable selection identified an optimum model comprised of kernel hardness, GGratio, dough extensibility and compression force as the most important variables. Cross-validation indicated 83% prediction efficiency for the model. This emphasizes the feasibility and practicality of the model using variables that are easily and quickly measured. This is the first model that can be used to simultaneously predict both tortilla diameter and rollability. It will be a useful tool for the flat bread wheat breeding programs, wheat millers, tortilla processors and wheat marketers in the United States of America.

DEDICATION

I dedicate this work to my father Prof. Isaac J. Jondiko Ogoche, my brothers and sisters for
their love, patience and encouragement

And

To the loving memory of my mother Rachel Anyango Jondiko (Deceased)

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I wish to express my deepest gratitude to Dr. Joseph Awika, my committee chair, for being my teacher and mentor. He has been a positive influence on my life and career and I am grateful for the opportunities that he provided during my doctorate studies and for helping me to focus on the big picture as I make sound and informed scientific conclusions. Your patience, wisdom and guidance have been a blessing to me.

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statistical analyses and interpretations. She made a significant input to enable this project succeed. She was always a resource to turn to whenever I have professional and personal challenges. Liyi you are my sister and will always be, even if I overworked the equipment or always needed disposable tubes.

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NOMENCLATURE

GGRatio	Glutenin Gliadin Ratio
HMW-GS	High Molecular Weight Glutenin Sub-Units
IPP	Insoluble Polymeric Protein
PCA	Principal Component Analysis
TIA	Tortilla Industry Association
TXE/UVT	Texas Elite/ Uniform Variety Test

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CHAPTER I

INTRODUCTION

Tortilla is currently an integral part of the American diet. According to the tortilla industry association report for 2013, wheat tortilla is the only bakery segment that experienced growth in 2012 and is projected to increase further (TIA 2013). This growth can be attributed to the demographic changes, specifically increase in the Hispanic population, growth in the number of non-Hispanic consumers, and increase in health conscious consumers who prefer tortillas than traditional pan bread. In 2011, there was a significant increase in tortilla exports from USA to Mexico and Canada. Tortilla sales exceeded \$ 11 billion in 2012, affirming consumer preference for its versatility and functional convenience. The main quality attributes related to this functional convenience are tortillas diameter and flexibility over time (shelf stability). Currently, most tortilla processors use trial and error and additives to optimize tortilla quality, which compromise sensory appeal and quality. The health conscious consumers are demanding tortillas with fewer than five added ingredients (clean label), hence the need to increase knowledge of how native wheat flour components contribute to tortilla quality and produce quality products to meet consumer demands with superior sensory appeal. Barros et. al., (2010) used fundamental dough rheology in developing linear tortilla quality prediction models. Such rheological methods are dependent on the shape, size and extrinsic properties of dough which easily creates high variability. Hence, this study explored univariate and multivariate relationships between dough, kernel, flour and tortilla properties for their potential in predicting quality.

Earlier studies by Waniska and others (2004) reported that tortillas of good quality can be produced using wheat flour of intermediate protein content, protein strength and low level of starch damage. Amylose content, ash content, and flour particle size have also been demonstrated to significantly affect tortilla quality (Prasopsunwattana et al. 2009; Whitney et al. 2011). These studies have not adequately established the relationship between protein quality and starch properties that can be used to predict tortilla quality. Breeding targeted at high molecular weight glutenin subunits composition (HMW-GS) can produce wheat varieties with unique protein quality for tortilla production. One of the challenges in identifying wheat varieties that can make good quality tortillas is that the two most important quality parameters tortilla diameter and rollability are usually inversely correlated. Hence, a variety may produce large diameter tortillas that have a low rollability score and vice versa. Varieties possessing null or 2+12 subunits at the Glu-D1 loci produce good quality tortillas (Jondiko et al. 2012a). Zhang and others (2007) reported that dough extensibility is determined by the gliadin quality and the high molecular weight glutenin (HMW) to low molecular weight glutenin (LMW) ratio which impacted the quality of pan bread and Chinese white salted noodles. The relationship between the HMW-GS and low molecular weight glutenin sub-unit (LMW-GS) ratio and tortilla quality has not been fully investigated.

Despite the enormous acceptability and popularity of tortillas the main challenge is that there is no reliable and practical model for prediction of tortilla quality based on grain and flour properties as is the case with pan bread. The quality of tortillas is defined using combination of desirable quality attributes (both diameter and rollability) and

hence, requires a multivariate approach to prediction. Barros et. al (2010) reported that tortilla diameter can be predicted using linear equations comprising of mixing time and dough resistance to extension. However, these linear models did not significantly predict tortilla flexibility which is a critical quality attribute for the unique versatility of tortillas (Alviola and Awika 2010).

The current quality screening methods are time consuming ~21 days, costly, and require at least 1 kg of flour which is not easy to get from early generation pedigrees of wheat for tortilla production.

This study provides information that will increase the understanding of the roles of protein fractions on tortilla quality. The potential of dough properties, in conjunction with kernel and flour parameters to predict tortilla quality were determined using multivariate modeling. In addition, the study investigated whether these multivariate models could be used to reliably classify early to late generation wheat lines for their potential to produce good quality tortillas.

RESEARCH OBJECTIVES

- 1) Determine the univariate relationships between kernel, flour, dough and tortilla properties.
- 2) Determine the effect of polymeric proteins, high molecular weight wheat glutenin (HMW-GS) and low molecular weight wheat glutenin (LMW-GS) content on grain, dough properties and tortilla quality.
- 3) Develop multivariate prediction models for screening of wheat lines developed for tortilla production.

CHAPTER II
RELATIONSHIP BETWEEN KERNEL, FLOUR, DOUGH AND TORTILLA
PROPERTIES

INTRODUCTION

Dough rheology plays an important role in determining the quality of baked products (Lefebvre 2009; Sliwinski et al. 2004). Wheat dough is viscoelastic (Figure 1) and has a nonlinear behavior under steady shear flow (Figure 2). Wheat flour dough is a shear thinning and thixotropic material (Weipert 1990). These attributes result from the complex nature of wheat dough in which starch granules (75-80%) are held together by a protein network (20 – 25%) (Rao et al. 1986; Weipert 1989). This protein network consists of prolamins (gliadins), glutenin and non-gluten proteins (15-20% of the total wheat proteins) such as albumins and globulins (Veraverbeke and Delcour 2002).

Dough rheology can be evaluated using empirical techniques. These include farinograph, mixograph, stress relaxation and texture profile analysis (TPA). However, these techniques provide instrument dependent measurements. Dough mixing time is negatively correlated with the glutenin:gliadin ratio (Barak et al. 2013) and tortilla diameter (Barros et al. 2010; Jondiko et al. 2012a). Dough resistance to extension can also predict the diameter of hot-press tortillas (Barros et al. 2010). Hence, dough mixing time and extensibility are partial predictors of tortilla quality. Farinograph provides the most practical information regarding wheat flour water absorption which is directly related to dough formation and product quality (Tamara et al. 2011). Stress relaxation involves subjecting a dough sample to a specified deformation and the stress required to

maintain the deformation is measured as a function of time (Steffe 1996). Jondiko and co-workers (2012) and (Limanond et al. 2002) reported that relaxation time had a significant role in a linear model for prediction of tortilla flexibility. Stress relaxation is rapid, simple to perform and is suitable for routine quality assurance work. On the flip side, these types of tests have several disadvantages especially for testing nonlinear viscoelastic material (Hibberd and Parker 1975). These demerits include; the sample must be uniform, regular in shape, homogenous and isotropic. For a complex food system such as dough, it is not easy to get homogenous sample. Therefore, these dough rheology measurements require a many experimental repetitions to decrease variability of results compared to measurements on linear viscoelastic materials (Hibberd and Parker 1975; Steffe 1996).

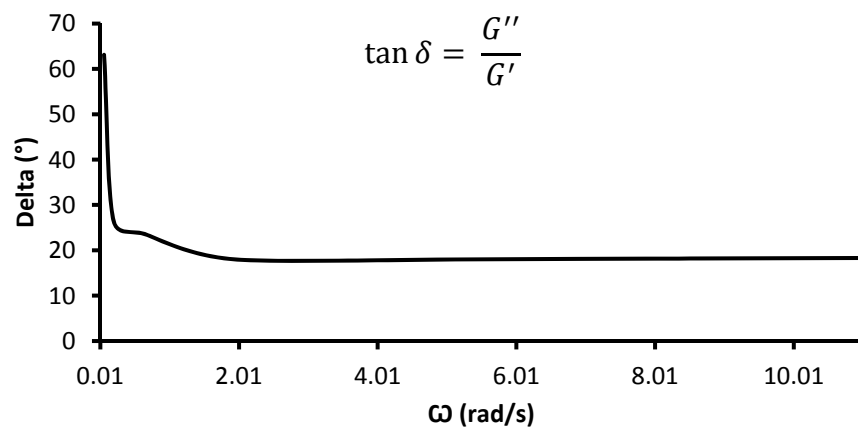


Figure 1. Typical phase angle graph confirming the viscoelastic nature of dough ($0^\circ < \delta < 90^\circ$)

Another category of rheological methods are the fundamental techniques. These techniques measure well-defined properties that are derived from the relationship between stress and strain which are independent of the instruments used (Hibberd and Parker 1975; Steffe 1996). These techniques use small strains on samples that are assumed to be homogeneous. However, the methods are slow and the results are known to have very low correlation with sensory evaluation of foods compared to the empirical tests (Steffe 1996). Though these are not actual measures of rheological parameters they are useful in providing a link between objective mechanical behavior of dough and quality of baked foods (Hibberd and Parker, 1975).

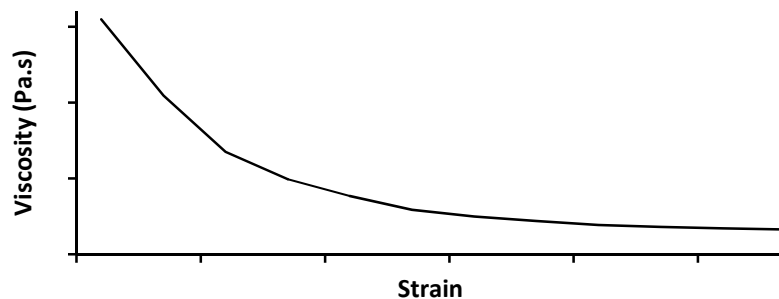


Figure 2. Strain dependent properties of wheat dough

The third categories of rheological methods are the dynamic oscillatory tests which determine the viscoelastic properties simultaneously and are known to have minimal destruction of the sample structural properties. There are three approaches in oscillatory testing; one is the angular frequency sweeps test that provide viscoelastic characteristics of doughs by monitoring both elastic (G') and viscous (G'') moduli. The

second one is the stress sweep tests which determine dough elasticity by analyzing the rheological response of dough during and after application of a constant strain. The third is temperature sweep tests which are useful in determining the thermal stability of the dough structure providing information regarding gelatinization of starch and denaturation of protein.

The relationships between pan bread quality and rheological behavior of wheat flour doughs have been reported (Renzetti et al. 2008; Torbica et al. 2010).

However, there are minimal studies on the link between the dynamic dough properties and the quality of wheat flour tortillas. Jondiko, et al (2012b) demonstrated that tortilla ingredient formulation variations can be investigated using dynamic tests. Dough compression and stress relaxation equilibrium forces measured using the texture analyzer TA-XT2i cannot significantly distinguish treatment differences involving very minor formulation changes compared to dynamic tests that have been reported to be highly sensitive and repeatable (Jondiko et al. 2012b). Dynamic oscillatory tests will yield information on the storage (G') and loss (G'') moduli, tangent delta, dynamic (η') and complex (η^*) viscosity. I hypothesize that these dough parameters are essential in understanding the relationship between dough rheology and tortilla quality. This study explored the correlation between major wheat flour components with the dough rheology. I investigated the relationship between empirical, fundamental, dynamic rheological properties of dough and tortilla quality.

Wheat flour tortilla quality

Tortilla is a complex system in which quality comprises more than one attribute. Good quality tortillas retain flexibility during storage (shelf-stable), have large diameter and high opacity (Waniska et al. 2004). Good tortillas also resist rapid moisture uptake compared to pan bread.

Tortilla diameter and flexibility are strongly related to flour composition and characteristics of starch, protein, and non-starch polysaccharides in flour. In general, flour for tortilla production has low water absorption, minimal gluten strength, and low level of damaged starch (Waniska et al. 2004).

Tortilla diameter is negatively correlated with tortilla flexibility (Alviola et al 2008). Diameter is associated with dough gluten strength and elasticity. Highly elastic dough's that are desirable for pan bread production have been shown to produce small diameter tortillas that have very good flexibility scores over storage. Flour protein content is negatively correlated with tortilla diameter and positively with tortilla flexibility. Tortilla diameter can be predicted using linear models comprising mixing time and dough strength (Barros et al 2010). However, these models cannot be used to reliably predict tortilla flexibility.

Wheat protein composition

Protein in wheat flour is a complex mixture of molecules with varying sizes and structures (Nimmo et al. 1964). These molecules can be categorized as glutenin, gliadin and albumin/ globulin (Mimouni et al. 1998). Glutenin is responsible for the elasticity of dough whereas gliadin is associated with dough extensibility. Higher flour protein

content is essential for longer shelf stability in tortillas but is detrimental to tortilla diameter (Waniska et al. 2004). Addition of wheat protein fractions has been shown to improve tortilla rollability (Pascut et al. 2004).

High protein flours produce pan bread that are generally more shelf-stable, have higher loaf volumes, and softer crumb (Bechtel and Meisner 1954). However, in tortillas high protein content is associated with more shelf-stable but dense tortillas. In recent studies, protein content alone has been shown not to be the only determinant of tortilla shelf stability (Alviola and Waniska 2008). Hence, there is need to comprehensively investigate the roles of various wheat protein fractions in tortilla quality.

Glutenin and gliadin are the proteins responsible for tortilla dough network requirements. Breeding and genetic studies have identified wheat lines with desirable functionality for tortillas. However, the specific compositional attributes that contribute this functionality are not known. Presence of HMW-GS 2+12 (Figure 3) at the Glu-D1 loci produce large diameter tortillas that have good flexibility scores. On the contrary presence of HMW-GS 5+10 (Figure 4) will result in small tortillas that are highly flexible (Jondiko et al. 2012a). This is phenomenon is attributed to the lower number of disulphide bonds resulting from the 2+12 GS compared to the 5+10 HMW-GS (Figure 4). High number of disulphide linkages results in a strong gluten which causes tortilla discs to shrink back after hot-pressing consequently producing small diameter tortillas. Strong gluten structure can be depolymerized using reducing agents (such as cysteine) or proteases to improve dough machinability, extensibility and tortilla diameter. These additives can be counterproductive since the resulting tortillas break easily over storage

(Srinivasan et al. 2000). Recent breeding advancements indicate a potential for elimination of these additives from tortilla production through alternating HMW protein composition (Jondiko et al. 2012a).

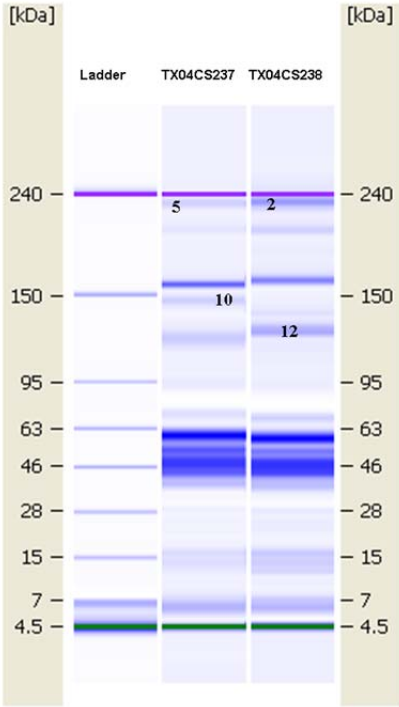


Figure 3. Electrophoretic characterization of Glu D1 HMW-GS for two wheat lines. (TX04CS237 and TX04CS238) indicating the presence of 5+10 and 2+12 (Jondiko et al., 2010)

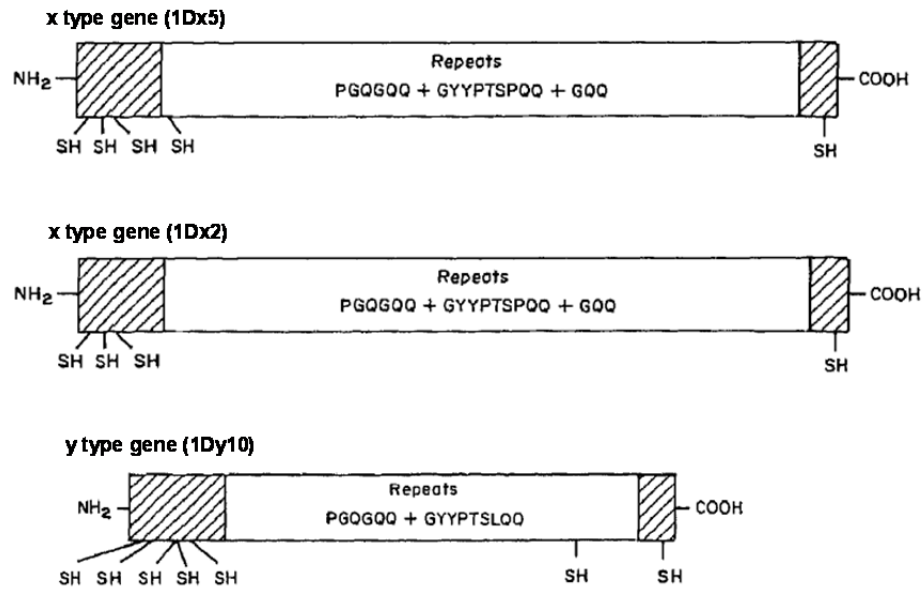


Figure 4. A schematic of the differences in number of disulphide bonds at Glu D1 loci (Suchy et al. 2003)

Polymeric interchain disulphide-linked wheat protein fractions have been reported to significantly affect flour and baking properties. High number of disulphide linkages results in highly elastic dough which is necessary for large pan bread volume. (Gupta et al. 1993; Turner et al. 1965). In tortilla processing high disulphide linkages is not desired for production of large diameter tortillas. Zhang and others (2007) reported that dough extensibility is determined by the gliadin quality and the HMW: LMW ratio which impacted the quality of pan bread and Chinese white salted noodles (Zhang et al. 2007). The relationship between the HMW-GS and LMW-GS ratio and tortilla quality has not been fully investigated.

Johansson et al., (2002) reported that seasonal variation in bread quality is linked to the variation in the amount and distribution of polymeric proteins in Sweden (Johansson et al. 2002). Gupta et al., (1993) reported that the low correlation between dough strength and total protein content is due to the compositional variation of polymeric proteins in wheat. The functional properties of these proteins in bread are known to be affected by the ratio between the HMW-GS and LMW-GS. The composition of the subunits has been confirmed to greatly influence tortilla quality (Jondiko et al. 2012a).

However, the association between polymeric and monomeric proteins and tortilla quality has not been fully understood. We hypothesize that the shelf –stability of tortillas can be greatly influenced by the compositional variation of polymeric proteins fractions and the ratio between HMW-GS and LMW-GS.

MATERIALS AND METHODS

Experimental design

A total of 185 HWW lines were used these included, Advanced (TXE/UVT) generation bread lines including 37 Texas elite (TXE) wheat lines, three uniform variety trial (UVT) (Table 1) and 100 distinctly different lines (Table 2) selected from the TAM111xTAM112 (TAM1112) drought tolerance population planted in the Texas Agricultural Experiment Stations at Etter. Fifty seven identity preserved lines developed for specialty flat bread processing that we harvested from College Station, McGregor and Chillicothe, Tx (TIA) were also used. These lines possessed variations in the allelic composition of the HMW-GS loci GluA1, GluB1 and GluD1.

Kernel properties

The wheat lines were evaluated for hardness, diameter, weight and moisture content using a single kernel hardness tester (SKHT4100). Three hundred kernels were used from each wheat line.

Milling

The grains were tempered (24 hours) to a moisture content of 14% which is optimum for good flour yield during milling. The amount of tempering water was determined based on the grain moisture content (SKHT). The tempered grains were milled using a quad Senior mill (Barbender Instruments, Incorporation, South Hackensack, NJ) to obtain the flour.

Tortilla formulation

The tortilla formulation included 500 g flour from each of the wheat lines and white wheat flour (Cargill Company) was used as control. Other functional ingredients included: 7.5 g salt, 30 g of shortening (vegetable), 3 g sodium bicarbonate, 2.9 g sodium aluminum sulfate, 1.65 g encapsulated fumaric acid, 2.5 g sodium steroyl lactylate, 2 g potassium sorbate, 2.5 g sodium propionate and distilled water. Dough was prepared by mixing dry ingredients in a mixer (model A-200, Hobart Corp, Troy, OH) with a paddle at slow speed (speed 1) for 2 minutes. Shortening (Cargill Company) was added to the dry ingredients and mixed at slow speed (speed 1) for 3 minutes. Amount of water added to the dry ingredients was based on an adjusted value from the Mixograph water absorption. These were mixed using a hook at low speed for 2 minutes. The dough was then mixed at medium speed (speed 2) for the time it will take to reach the

mixograph peak time. The dough was then subjectively evaluated for smoothness, softness, extensibility and force to extend as described by Alviola et. al., (2007). The doughs were proofed for 5 minutes at 32° C and 65-70% relative humidity in a proofing chamber (Model 57638, National Manufacturing Co., Lincoln, NE). Dough temperature were measured and record.

Dough samples were pressed on a stainless steel rounding plate and rated for press rating, then divided and rounded into 36 dough balls (Duchess Divider/Rounder, Bakery Equipment and Service Co., San Antonio, Tx). The dough balls were then rested for 10 minutes at 32° C and 65-70% relative humidity in the proofing chamber. Tortillas were pressed and baked in a three-tier gas-fired oven (Model OP01004-02, Lawrence Equipment, El Monte, CA). The top and bottom platen temperature of the press was set at 400°F (204.4°C). The hot-pressing dwell time was 1.35 sec with a pressure of 1100 psi. The oven temperature 400°F (204°C) and oven dwell time was 30 seconds. The tortillas were cooled on a three-tier conveyor (Model 3106-INF, Superior Food Machinery Inc., Pico Rivera, CA) and individually placed on a sanitized table to cool further. The tortillas were packed in polyethylene bags and stored at ambient temperature (25°C), and sampled at 0, 4, 8, 12 and 16 days after baking. Two batches of each wheat line and control were prepared on separate days and evaluated.

Evaluation of dough properties

Dough development time and tolerance

A mixograph (National Manufacturing Co., Lincoln, NE) was used to estimate dough mixing properties: Mixing time, and tolerance (Figure 5). Ten grams of flour were

used (14% mb) (AACC 2000). Mixing time/ peak time was manually calculated from the mixograph by drawing two midlines from each end of the graph. The point of crossover was marked as the peak time for each wheat line

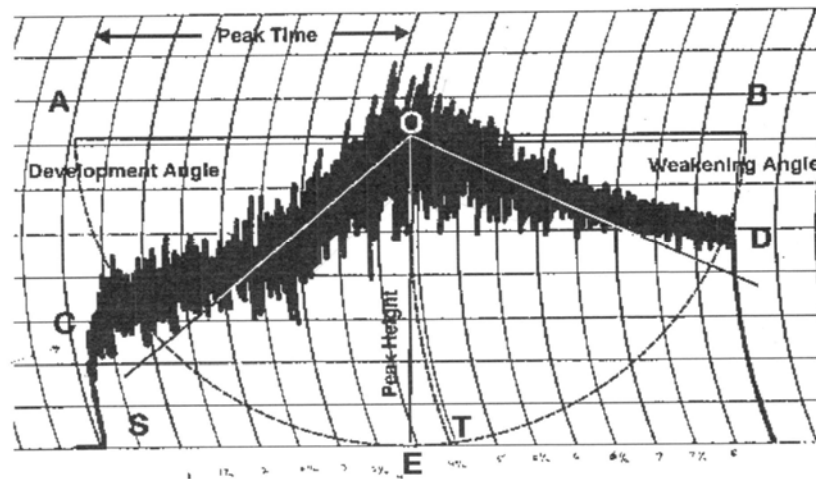


Figure 5. Mixogram identifying dough measurements. (AO - Peak time, OE – Peak Height, AOC – Development angle, BOD – Weakening angle and CED – Mixing tolerance)

Stress relaxation

Stress relaxation tests were conducted by compressing two dough balls (5.2 cm diameter, 2.1 cm height and 45 g weight) on a texture Analyzer (Model TA-XT2, Micro Systems, Scarsdale, NY) after 10 min resting time. A cylindrical probe with a diameter of 10 cm were attached to the texture analyzer arm and calibrated to a distance of 35 mm from the texture analyzer platform. The cylindrical probe compressed the dough balls for 120 Sec. The relaxation force at 25 seconds, 100 seconds, maximum force and relaxation time (Figure 6) were collected (Jondiko et al. 2012a).

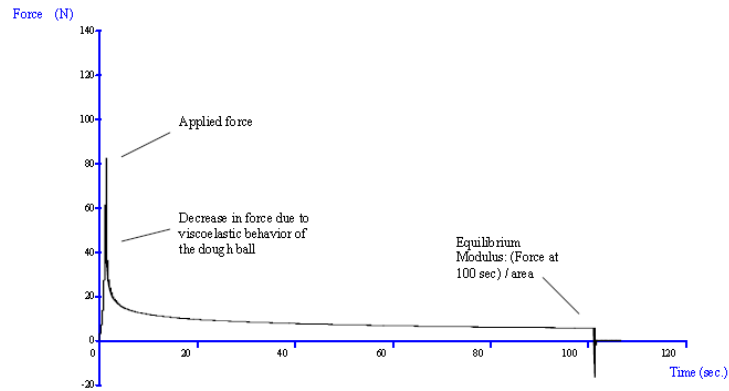


Figure 6. Stress relaxation graph identifying data to be collected

Dough extensibility test

The test was carried out done according to Smewing (1995), which uses the Kieffer dough and gluten extensibility rig (Smewing 1995), followed with modifications by (Barros 2009). After resting the dough balls for 10 min in the proofing chamber, 20 g of one dough ball were weighed and rolled into a cylindrical shape with as little manipulation as possible. The dough press with a grooved base and a top form were used to prepare the samples. Mineral oil was used to aid the removal of the dough strips and avoiding sample adhesion. The dough samples were placed on the grooved base with its length perpendicular to the groove direction. The top was then placed on the grooved base. The dough press was placed in the clamp and screwed down. Excess dough extruding from the sides was removed using a spatula. This process cut the sample into strips. The dough clamp was placed into a plastic bag and left to relax for 40 min at room temperature. After that, the plastic bag was opened, and the clamp released and the dough press removed. Dough strips were removed using a thin spatula and then placed

across the grooved region of the sample plate. The hook probe was then lowered to the surface of the spring loaded clamp. The lever of the spring loaded clamp was lowered and the sample plate inserted into the rig. The handle released slowly and the test conducted. Dough extensibility was defined as the distance the dough strip extends. The maximum force required to extend the dough strip until it breaks is the resistance to extension (Figure 7). Averages of 10 strips were used for each wheat line.

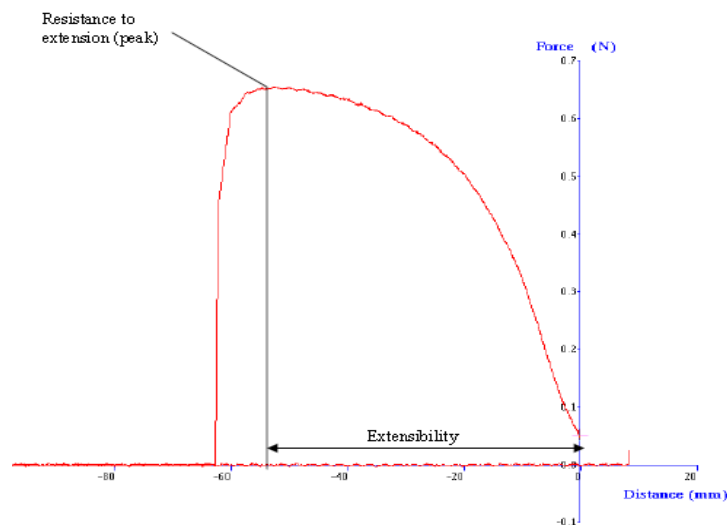


Figure 7. Dough extensibility test measurements (Barros 2009)

Dough compression test

Dough texture was also measured using a dough compression test (Barros 2009; Bejosano et al. 2005), two dough balls (5.2 cm diameter, 2.1 cm height and 45 g weight) were subjected to 70% compression using a 10 cm diameter probe on a texture analyzer

(Model TA-XT2, Micro Systems, Scarsdale, NY). Maximum force (N) was recorded (Figure 8). The tests were carried out at room temperature (~25° C)

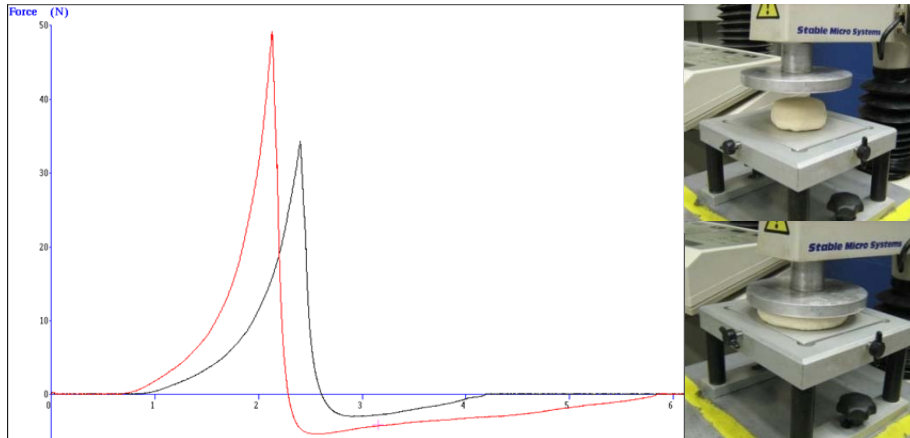


Figure 8. Typical dough compression test graphs and measurements (Barros 2009)

Evaluation of tortilla physical properties

Ten tortillas were selected randomly and weight, diameter, height, opacity, and moisture were determined on the first day after processing (Bello et al. 1991). Rollability and extensibility were measured at 4, 8, 12 and 16 days after production (Alviola and Waniska 2008). The following is a detailed description of the evaluations:

Moisture

Tortilla moisture content were determined using a two-stage procedure in a hot-air oven (AACC 2000). Pre-weighed tortillas were dried for 96 hours after production in ambient conditions followed by a one hour 100° C drying in oven (model 16, Precision Scientific Co. PS, Chicago, IL). Moisture was calculated as a percentage of weight loss

from the drying process (Alviola et al., 2008). Each wheat line was evaluated for moisture on two duplicates.

Weight

Ten randomly selected tortillas (Friend et al. 1995) were weighed using an analytical scale (Ohaus, Houston TX). The values recorded and averaged to obtain the weight of one tortilla

Diameter

Diameters of ten tortillas were measured by using a ruler at two points across the tortilla. These values were recorded to obtain the average diameter of one tortilla (Alviola et al. 2008).

Height

The average Height/ Thickness of a one tortillas were obtained by measuring the height of a stack of ten tortillas using a digital caliper (Chicago Brand 12” Electronic Digital Caliper, Chicago, IL).

Color

Color values L (lightness to darkness), a (red-green) and b (yellow-blue) were measured at two points of each side of two tortillas from each treatment using a Minolta Color Meter (Chroma Meter CR-310, Munilta, Tokyo, Japan).

Specific volume

Tortilla specific volume was determined as follows: Specific volume = $(height) * (\pi r^2)$. Where; height = height of one tortilla weight = weight of one tortilla (g), r=average radius of a tortilla.

Rollability/ flexibility

Tortilla shelf stability was evaluated by the subjective rollability test (Friend et al. 1995), which is a 5 point measure of the cracking and breakage of a tortilla. Two tortillas from each wheat line were wrapped around a 1.0 cm diameter wooden dowel on one side of the tortilla after 4, 8, 12 and 16 days of storage and were allocated a rollability score (RS) according to (Alviola and Waniska 2008; Cepeda et al. 2000; Limanond et al. 2002) on a continuous scale for rollability score: 5 = no cracking; 4 = signs of cracking, but no breaking; 3= cracking and breaking beginning on the surface; 2 = cracking and breaking imminent on both sides; and 1 = unrollable, breaks easily. A RS < 3 (many cracks and breaks on tortilla surface) were indicative of undesirable shelf stability during storage.

Tortilla texture - 2D extensibility

Tortilla textural changes during storage were monitored at day 4, 8, 12 and 16 using the two-dimensional extensibility tests (Barros 2009; Bejosano et al. 2005) on the texture analyzer (model TA-XT2i, Texture Technologies Corp., Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) using the method by (Suhendro et al. 1999). The extensibility test was conducted using the return to start option, at a trigger force of 0.05 N. Pre and post-test speeds were 10.0 mm/s. The test speed was 1.0 mm/s. The modulus of deformation (N/M), force (N), distance (mm) and work to rupture (Nm, area under the curve) were collected (Barros 2009).

Flour protein analysis

Total protein content

Near-infrared reflectance spectrophotometer (Pertten PDA 7000 Dual Array with Grams Software) was used to determine protein and moisture content of the milled flour from the wheat lines. Tortilla flour (ADM Milling Company, Overland Park, Kansas) were used as a control and to calibrate the instrument. Three replicates of each sample were analyzed and the protein and moisture values recorded (AACC 2000).

Polymeric to monomeric protein ratio (Glutenin to Gliadin Ratio)

Protein extraction of proteins followed the method of Gupta et al (1993). Briefly, a 10 mg flour sample were mixed with 1ml 0.05 M Sodium phosphate buffer, pH 6.9, containing 0.5% SDS (w/v) then sonicated for 35 s at power setting 10 W. The sample will then be centrifuged at 15,294 xg for 5 minutes and the supernatant collected (contains total protein) and filtered through 0.45 µm filter and analyzed by size – exclusion HPLC using a 300 x 7.8 mm Biosep S4000 column with gradient system composed of 50% ACN+0.1% TFA (B) and 50% water+ 0.1% TFA (A), 30°C column temp., at a flow rate of 1 ml/min for 30 min run. The chromatograms were manually integrated. The area of the first peak corresponds to total polymeric proteins and the area of the second peak to monomeric proteins. Two replicated of each flour sample were analyzed.

Insoluble polymeric protein content (%IPP)

Protein extraction (Bean et al, 1998) were done on 100 mg of flour to which 1 ml 50% 1-propanol was added and vortexed for 5 min, and then centrifuged for 5 min at

12,000 xg. The supernatant (containing soluble monomeric and polymeric proteins) were discarded. This process was repeated twice to ensure maximum removal of soluble protein polymers. The pellet containing the insoluble polymeric proteins were lyophilized. The pellet protein content was determined by nitrogen combustion method (LECO analysis). Insoluble polymeric protein percentage (%IPP) were calculated by a conversion factor of 5.7 and divided by the total flour protein content.

Extractable (%EPP) and un-extractable polymeric protein content (%UPP)

Ten milligram (10 mg) of flour was suspended in 1 ml of 0.05 M sodium phosphate buffer (pH 6.9), containing 0.5% sodium dodecyl sulphate (SDS) and shaken on a vortex for 30 min. The mixture then centrifuged for 5 min at 16,595 xg. The supernatant (containing extractable polymeric protein - EPP) were collected and filtered (0.45 µm) and analysed by size – exclusion HPLC as described above. The pellet were mixed with 1 ml sodium phosphate buffer and sonicated for 25 sec at 10 watt output. The mixture were centrifuged at 16,595 xg/5 min, the supernatant collected and filtered as above then analyzed using the SE-HPLC as described above. The percentages of extractable and unextractable polymeric protein were calculated as [peak 1 area (extractable)/peak 1 area (total)] x 100 and [peak 1 area (unextractable)/peak 1 area (total)] x 100 respectively. Peak 1 (total) refers to the sum of peak 1 (extractable) and peak 1 (unextractable) (Figure 9).

High molecular weight and low molecular weight glutenin sub-units analysis

HMW-GS and LMW-GS were quantified using RP-HPLC. 100 mg flour was mixed with 1ml sodium iodate buffer (0.3M sodium iodate + 7.5% isopropanol)

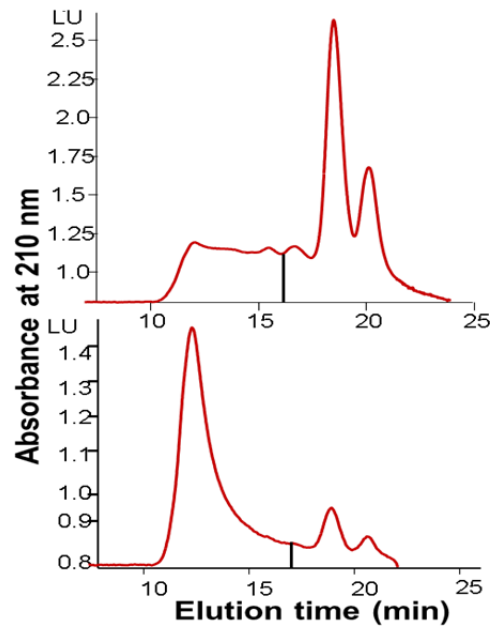


Figure 9 A typical SE-HPLC chromatogram of total, extractable and unextractable proteins.

and vortexed for 15 min. The mixture was centrifuged for 5 min at 15,294 xg. The supernatant containing gliadins were discarded. To the pellet 1ml water were added then shaken for 5 min and centrifuged as above. The pellet were mixed with 1 ml 50% isopropanol containing 2% BME and vortex for 30 minutes, and then centrifuged for 5 min. at 15,294 xg. The supernate was collected (contains glutenins). 600 ul of the glutenin extract were alkylated with 40 μ l 4-vinylpyridine for 15 min at 60°C. The resulting sample was injected into a Phenomenex column C 18 250 x 4.6, 5 μ diameter and 300 Å pore size. The solvent flow rate was 1.0 ml/min and composed of water (A) and acetonitrile (B), both containing 0.1% TFA. The gradient was as follows: 0-3 min from 25% B to 35% B, 3-24 min increased to 53%B, the gradient decreased to 25% B at

25 min and kept at 25% B until 29 min. Detection of protein peaks were carried out by UV detector at 200 nm. The area of the curve corresponding to HMW-GS and LMW-GS (Figure 10) contents were determined by manual integration and the HMW/ LMW-GS ratio calculated (Cinco-Moroyoqui and MacRitchie 2008; Fu and Kovacs 1999; Suchy et al. 2003).

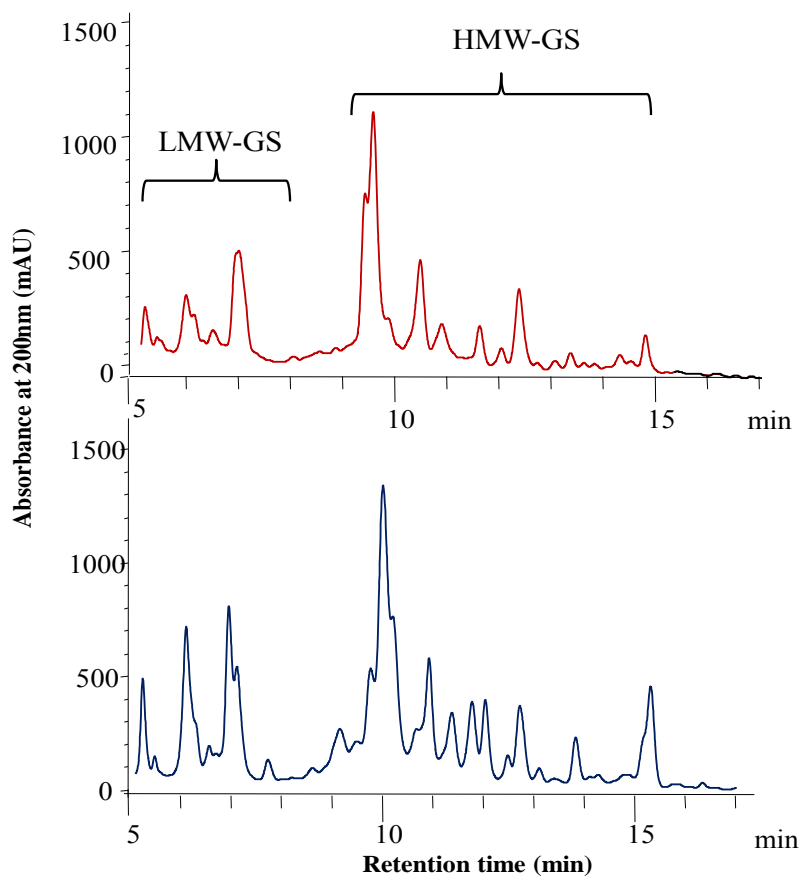


Figure 10. Typical RP-HPLC chromatograms of High Molecular Weight and Low Molecular Weight Glutenin Subunits for two wheat lines

Data Analysis

Data analysis were done using SAS version 9.2 (SAS Institute, Cary, NC).

Means and standard deviations were derived using SAS 9.2. Pearson's correlations were performed to investigate the relationships between the protein fraction contents and tortilla quality variables.

RESULTS AND DISCUSSION

Univariate relationship between kernel, flour, dough and tortilla properties

The means, standard deviation, minimum and maximum values for kernel, protein, dough and tortilla properties for the wheat lines are given in Table 1.

The overall mean for grain hardness index was 65.8 (Table 1). The mean hardness for the TIA, TXE/UVT and TAM111x112 lines were 67, 80 and 60 respectively (Table 2) hardness ranged between 38 and 89. Majority of the lines from the TXE/UVT population had the highest hardness index (Table 2). The kernel diameter ranged between 2.3 – 3.1 mm with an overall mean of 2.5 mm (Table 1). Lines from Chillicothe, College Station and McGregor (TIA) had the low hardness index (Table 2). This can be attributed to the variation in high molecular weight glutenin sub-units at loci GluA1, GluB1 and GluD1, they were specifically developed to have the unique protein quality for production of specialty flat breads which require lower gluten strength compared to wheat lines used for pan bread processing which must have high hardness index that has been shown to provide the high protein strength to hold the crumb cells in place during and after baking.

Kernel weight means were 28, 31 and 25 g for TIA, TXE/UVT and TAM1112 lines respectively. Kernels from TAM1112 had significantly lower weights than kernels from both TIA and TXE/UVT (Table 2). Kernel hardness, diameter and weight were significantly ($P < 0.05$) negatively correlated with dough elasticity, relaxation force after 25 sec (F_{25}) and after 100 sec (F_{100}). The correlations coefficients ranged between $-0.24 - 0.32$ (Table 3). Dough extensibility was positively correlated with kernel properties ($P < 0.05$). This was expected owing to the diversity in the wheat lines. Kernels with high hardness index produced highly extensible doughs that were less elastic and had gluten structure that relaxed quicker and would likely produce large diameter tortillas.

Dough extensibility (mm) averages were 83 mm (TIA), 71 mm (TXE/UVT) and 56 mm (TAM1112). Majority of the least extensible lines were from the TAM1112 population (Table 2). These lines were specifically bred for drought tolerance study and the kernels had very high protein content (probably due to drought conditions) and high gluten strength which was more elastic compared to TIA lines. TIA lines on the other hand had highly extensible doughs. These lines possess deletions at the GluA1, B1 and D1 which caused the doughs to be more extensible (Jondiko et al. 2012a).

Dough elasticity (the resistance to extension) ranged between 0.1 – 1.2 N (Table 1). The work to extend was between 17.5 and 20.9 N.mm (Table 2). As expected the TIA lines were the least elastic. The average elasticity for the TXE/UVT lines was similar to TIA and significantly lower than lines from TAM1112 lines (Table 2). The high elasticity was probably due to the harsh drought conditions.

The dough compression force for the TXE/UVT and TAM111x112 lines ranged from 36 to 170 (N), with an average of 90 (N). The compression force varied widely among these lines (Table A1). Compression force was negatively correlated with tortilla diameter and specific volume (Table 4). Hence, doughs that required high force to compress had strong or elastic gluten structure that could not retain its shape during tortilla pressing. This is also supported by the significant positive correlation ($P < 0.05$) between compression force both modulus of deformation ($r = 0.37$) and force to rupture ($r = 0.28$) meaning that the tortillas were less brittle and required high force to rupture after 16 days.

The mean stress relaxation time was 1.74 (min) and ranged between 1.2 and 2.7 min (Table 1). This is the time it takes for the maximum compression force of dough to decay to 36.8% of its initial value. Doughs that had lower relaxation time had gluten structure that could not resist the compression for a longer time and hence could produce tortillas with large diameter. This is confirmed by the significant correlation between tortilla diameter and relaxation time (Table 4).

Table 1
Overall means, standard deviations (Std Dev) and ranges for all wheat lines used

Variable	¹ N	Mean	Std Dev.	Minimum	Maximum
Kernel Properties					
Hardness (Index)	185	66.8	10.8	38.2	89.9
Diameter (mm)	185	2.5	0.1	2.3	3.1
Weight (g)	185	27.3	3.3	21.0	38.1
² Mixo_time (Sec.)	185	3.4	1.1	1.5	7.5
Protein Quality					
Protein content (%)	185	14.3	0.9	11.9	16.4
³ GGRatio	171	0.7	0.1	0.5	0.99
⁴ H_L_GS_Ratio	176	0.3	0.1	0.3	0.6
⁵ IPP (%)	183	46.4	7.9	12.2	72.2
Dough Properties					
*Elasticity (N)	175	0.5	0.2	0.1	1.2
Extensibility (mm)	175	66.2	24.0	23.1	136.2
Work to extend (N.mm)	173	19.4	4.7	4	31.1
Relaxation time (Sec.)	185	1.7	0.3	1.2	2.7
⁶ F_25 (N)	185	8.4	1.3	5.3	12.4
⁷ F_100 (N)	185	5.7	1.2	3.2	11.0
Compression force (N)	183	93.6	25.8	35.8	170.3
Tortilla properties					
Diameter (mm)	185	165.1	8.7	136.1	184.0
Specific volume (cm ³ /g)	185	1.6	0.2	0.8	2.1
Lightness (L – value)	185	82.1	2.0	72.6	85.6
Rollability	180	3.14	0.8	1.3	5.0
Gradient	155	0.9	0.2	0.5	1.4
Force	155	8.8	3.0	3.8	17.5
Distance	155	16.2	3.7	8.3	24.7
Work	155	57.8	34.1	13.3	202.5

¹Total number of lines evaluated, entries with missing data were deleted. ²Dough development time from mixograph peak time, ³Glutenin to Glidinin Ratio, ⁴High Molecular Weight to Low Molecular Weight Glutenin Sub-Unit Ratio, ⁵Insoluble polymeric protein content, ⁶Stress Relaxation Force after 25 Sec. and after ⁷100 Sec. compression. *Elasticity means the force recorded as resistance to extension in dough extensibility test.

Table 2
Means, standard deviations (Std Dev) and ranges for variables measured on the TIA, TXE/UVT and TAM1112 wheat lines¹

Variables	Location		
	TIA	TXE_UVT	TAM1112
Kernel properties			
Hardness (Index)	66.7 ± 10.6b	80.3 ± 4.8a	59.6 ± 5.5c
Diameter (mm)	2.6 ± 0.1a	2.6 ± 0.1b	2.4 ± 0.1c
Weight (g)	28.1 ± 2.4a	30.8 ± 2.2a	25.0 ± 1.8b
Flour properties			
Protein content (14% mb)	14.9 ± 0.7a	13.0 ± 0.7c	14.6 ± 0.3b
Mixo time (min)	2.9 ± 1.1c	3.2 ± 1.0b	3.8 ± 1.1a
Glutenin to Gliadin Ratio	0.6 ± 0.1a	0.8 ± 0.1a	0.7 ± 0.1b
H_L_GS_Ratio	0.4 ± 0.1a	0.3 ± 0.03b	0.3 ± 0.04c
IPP (%)	41.4 ± 5.6c	43.9 ± 4.5b	49.7 ± 8.2a
Dough rheology			
Elasticity (N)	0.3 ± 0.2b	0.4 ± 0.2b	0.6 ± 0.2a
Extensibility (mm)	82.9 ± 27.1a	71.1 ± 19.3b	55.9 ± 18.4c
Work to extend (N.mm)	17.5 ± 4.7b	18.1 ± 3.4b	20.9 ± 4.6a
Relaxation time (min)	1.6 ± 0.4c	1.8 ± 0.1b	1.7 ± 0.3a
² F ₂₅ (N)	8.4 ± 1.3ab	7.8 ± 1.2b	8.7 ± 1.3a
³ F ₁₀₀ (N)	6.0 ± 1.9a	5.2 ± 1.0b	5.9 ± 1.0a
Compression force (N)	104.1 ± 17.5a	89.4 ± 21.4b	91.0 ± 29.2b
Tortilla properties			
Tortilla diameter	169.4 ± 7.9a	164.9 ± 8.2ab	163.0 ± 8.4b
Tortilla specific volume (cm ³ /g)	1.7 ± 0.2a	1.5 ± 0.2c	1.6 ± 0.2b
Lightness (L)	82.5 ± 1.2a	82.2 ± 1.8a	81.9 ± 2.2a
Rollability (Score)	3.4 ± 1.0a	3.2 ± 0.5a	3.0 ± 0.7a
Gradient (N/mm)	0.8 ± 0.2c	0.9 ± 0.1b	0.9 ± 0.2a
Force (N)	6.1 ± 1.5c	8.5 ± 2.2b	10.7 ± 2.7a
Distance (mm)	11.4 ± 1.2c	16.4 ± 1.8b	19.0 ± 2.3a
Work (N.mm)	24.6 ± 7.8c	50.6 ± 14.6b	82.1 ± 32.7a
N	57	40	100

¹ Average from all lines planted in each location. Values followed by the same letter in each row are not significantly different (P < 0.05)

²Insoluble polymeric protein content, ³Stress Relaxation Force after 25 Sec. and after ⁷100 Sec. compression

The relaxation force after 25 sec of compression (F_25) range was 5.3 – 12.4 (N), whereas the relaxation force after 100 sec of compression was between 3.2 and 8.6 (N) (Table 1). F_25 and F_100 were highly correlated with of tortilla texture ($r > 50$) (Table 4). Hence, doughs had high relaxation forces produced tortillas that required longer distance to rupture and high force to rupture and were more flexible after 16 days.

Table 3
Pearson correlation coefficients between dough rheology, kernel and flour properties

	Dough Extensibility			Stress Relaxation			Compression Force
	Elasticity	Extensibility	Work	¹ RT	² F_25	³ F_100	
Kernel Properties							
Hardness	-0.32*	0.26*	-0.26*	0.06	-0.25*	-0.24*	0.01
Diameter	-0.26*	0.25*	-0.17	0.04	-0.26*	-0.26*	-0.07
weight	-0.26*	0.23*	-0.18	0.06	-0.29*	-0.28*	-0.05
Flour properties							
Mixo_Peak time	0.27*	-0.37**	0.05	-0.05	0.38**	0.38**	0.11
Protein content	0.32*	-0.20*	0.29*	-0.11*	0.16	0.16	0.00
Glutenin to Gladin ratio	-0.15	0.22*	-0.09	-0.03	-0.26*	-0.23*	0.02
⁴ HMW to LMW GS Ratio	0.02	-0.04	-0.12	0.00	-0.03	-0.02	0.04
⁵ IPP%	0.35	-0.21	0.31	0.09	0.31	0.32	-0.01

¹Stress relaxation time, ²Stress Relaxation Force after 25 Sec. and after ³100 Sec. compression
⁴High Molecular Weight to Low Molecular Weight Glutenin Sub-Unit Ratio, ⁵Insoluble polymeric protein content

Tortilla diameter ranged between 136 and 184 mm with an overall mean of 165 mm (Table I). Tortilla diameter was significantly correlated with all the dough properties (Table 4). Stress relaxation force F_25 and F_100 were highly negatively correlated with tortilla diameter ($r > - 0.70$). This is because the doughs that had high relaxation forces had strong gluten matrix which retained its structure and could not spread into a large

disc during tortilla hot pressing resulting in small diameter tortillas. However, tortilla rollability score after 16 days of storage which is a critical quality parameter was not significantly correlated with any of the dough rheological properties (Table 4). The tortilla rollability scores were between 1.3 and 5.0 (Table I). The correlation between tortilla diameter and both mixograph peak time ($r = -0.35$) and dough elasticity ($r = -0.46$) were lower than previously reported $r = -0.87$ and -0.86 respectively (Barros et. al 2010). This was likely due to the large sample size and diverse set of samples used in this study ($N = 185$) which is almost 10 fold what was used in the earlier study. The large sample size was more representative of the diversity in dough properties of wheat varieties.

Objective tortilla textural properties; modulus of deformation, force, distance and work to rupture were negatively correlated with dough extensibility and positively correlated with elasticity, relaxation forces F_{25} and F_{100} (Table 4). This implies that tortilla shelf stability can be predicted using these dough properties with a higher precision compared to the subjective tortilla rollability scores.

This study identified significant correlations such as between dough extensibility and tortilla distance to rupture (Table 4) which has not been reported before.

Table 4
Pearson correlation coefficients between dough rheology and tortilla properties

	Dough Extensibility			Stress Relaxation			Compression Force
	Elasticity	Extensibility	Work	RT	F_25	F_100	
Tortilla Diameter	-0.46**	0.38**	-0.19*	-0.19*	-0.73**	-0.71**	-0.17*
Specific volume	-0.29*	0.24*	-0.05	-0.09	-0.47**	-0.46**	-0.26*
Lightness	-0.04	0.16	0.06	0.12	-0.19	-0.17	-0.35
Flexibility (Day 16)	0.15	-0.04	0.14	0.11	0.13	0.14	0.01
Modulus of deformation	0.40**	-0.45**	-0.03	-0.03	0.50**	0.50**	0.37**
Force to rupture	0.54**	-0.48**	0.18	0.04	0.62**	0.60**	0.28*
Distance to rupture	0.50**	-0.28**	0.42**	0.11	0.47**	0.45**	-0.07
Work to rupture	0.60**	-0.43**	0.33*	0.06	0.61**	0.60**	0.13

** Correlation is significant at both $p < 0.01$ and $p < 0.05$ and * Correlation is significant at only $p < 0.05$

The role of flour protein fractions in kernel and tortilla quality

The flour protein content (14 % m.b) measured using the NIR for the TIA, TXEUVT and TAM1112 lines were 14.9, 13.0 and 14.6 % respectively (Table 2). Kernel diameter and weight were highly negatively correlated with protein content $r = -0.60$ and -0.68 respectively (Table 5). Hence, kernels with small diameter and weight had high protein content, because under drought conditions the kernels did not produce a lot of starch in its endosperm during maturation.

Both tortilla diameter and rollability were not correlated with protein content. This implies that flour protein content is not a reliable predictor of tortilla diameter and rollability. However, tortilla specific volume and objective tortilla texture properties force, distance and work to rupture were positively correlated with flour protein content (Table 5). This means that a large proportion of the variability in tortilla shelf stability is attributed to the flour protein content.

The correlations between tortilla rollability and both distance ($r = 0.25$) and force ($r = 0.21$) to rupture were significant ($P < 0.05$). Hence, these objective tortilla texture measurements can be optimized to identify a range that can be used to determine which lines produced good or poor quality tortillas other than using subjective rollability scores as a determinant of wheat functionality performance.

IPP% was negatively correlated ($P < 0.05$) with kernel hardness ($r = -0.23$), diameter ($r = -0.32$) and weight ($r = -0.30$). Tortilla diameter and texture were significantly correlated with flour IPP content (Table 5).

Table 5
Pearson correlation coefficients between protein fraction content, kernel properties and tortilla quality variables

	Protein content	Glutenin to Gliadin ratio	HMW to LMW GS Ratio	IPP%
Kernel Properties				
Hardness	-0.71**	0.63**	0.24*	-0.23*
diameter	-0.60**	0.64**	0.20*	-0.32*
weight	-0.68**	0.68**	0.28*	-0.30*
Tortilla Properties				
Tortilla Diameter	0.01	0.05	0.04	-0.23*
Specific volume	0.17*	-0.02	-0.21	-0.03
Lightness	-0.01	0.01	0.15	0.05
Flexibility (Day 16)	-0.06	0.06	0.27*	0.07
Modulus of deformation	-0.04	-0.05	-0.02	0.20*
Force to rupture	0.22*	-0.23*	-0.11	0.33*
Distance to rupture	0.46**	-0.30*	-0.16	0.32*
Work to rupture	0.35*	-0.27*	-0.14	0.39**

** Correlation is significant at both $\rho < 0.01$ and $\rho < 0.05$

* Correlation is significant at only $\rho < 0.05$

The negative correlation between tortilla diameter and IPP means that the insoluble polymeric proteins play a significant role in increasing the elasticity of gluten matrix producing in small diameters tortillas during processing. IPP can be used to explain 23 % of variability in tortilla diameter but cannot significantly explain the variability in tortilla rollability. On the other hand high IPP content results in production of highly flexible tortillas that require longer distance to rupture; IPP was responsible for up to 39 % of the variability in tortilla texture based on correlation on Table 5. This underscores the need to explore the role of other protein fractions on tortilla shelf stability.

The overall mean for glutenin to gliadin ratio (GGRatio) was 0.71 with a standard deviation of 0.11 (Table 1). The advanced lines TXE/UVT had the highest GGRatio whereas the lines possessing variations in HMW-GS GluD1, B1 and D1 (TIA lines) had the lowest GGRatio (Table 2). Glutenin to gliadin ratio was strongly positively correlated with kernel hardness ($r = 0.63$), diameter ($r = 0.64$) and weight ($r = 0.68$) (Table 5). The high content of glutenin proteins can be associated with the high amount of endosperm starch content resulting in large diameter and weight compared to elevated gliadin content this is because glutenins may be the main structure that holds the starch granules in the endosperm. GGRatio was negatively correlated ($P < 0.05$) with tortilla force, distance and work to rupture (Table 5). Glutenin is highly polymeric and is associated with the elasticity of dough (Veraverbeke and Delcour 2002). Hence, high glutenin content contributed significantly to the flexibility of tortillas via the increased number of disulphide bonding in the polymeric glutenins that produced highly elastic

dough. As expected the lines from TIA had the highest high molecular weight to low molecular weight glutenin sub-units ratio (HLMW-GSRatio). This is attributed to the genetic variation in the Glu1 GluB1 and GluD1 loci. This lines produced tortillas with significantly larger diameters and acceptable tortilla rollability scores (Jondiko et al. 2012a). The HLMW-GSRatio was significantly correlated with kernel hardness, diameter and weight ($P < 0.05$) (Table 5). It was the only variable that was significantly correlated with tortilla rollability ($r = 0.27$). The positive correlation with tortilla rollability means that high molecular weight glutenins provides a large number of noncovalent hydrogen bonding sites that form highly elastic dough which makes tortillas flexible. Hence, this indicates that of all the protein properties the high molecular weight to low molecular weight glutenin sub-units ratio could be a better predictor of tortilla rollability. However, it was not significantly correlated with the objective tortilla texture variables (Table 5).

CHAPTER SUMMARY AND IMPLICATIONS

Wheat protein fractions play a significant role in the tortilla making functionality of wheat flour. Environment and genetics contributed significantly to the variation in the tortilla making performance of the wheat lines used in this study. Flour protein and IPP contents explained variability in both dough rheological properties and objective tortilla texture over storage. Tortilla diameter can explained using a combination of flour IPP content, dough elasticity, extensibility, compression force, stress relaxation force and relaxation time. Whereas 27% of the variability in tortilla rollability can be predicted

using the ratio between high molecular weight glutenin sub-units and low molecular weight glutenin sub-units.

The glutenin to gliadin ratio is a good measure of dough extensibility and stress relaxation force. This ratio can be used to predict objective tortilla texture after 16 days of storage.

It is clear from this study that the measure of the tortilla making functionality is dependent on kernel, flour and dough properties. This is because the most important tortilla quality parameters diameter and shelf stability after 16 days of storage (rollability and objective textural properties) cannot be predicted simultaneously using univariate relationships of the kernel, flour and dough variables. Hence, we proceed to explore the potential of predicting tortilla making performance of the wheat varieties using multivariate analysis.

CHAPTER III

MULTIVARIATE MODELING OF TORTILLA QUALITY PREDICTION

INTRODUCTION

Wheat flour tortillas are valuable and highly appreciated breads in the USA due to their exceptional functional and sensory properties. To consumers, the definition of good quality tortilla encompasses its ability to retain flexibility and be large enough to wrap food. Due to the negative correlation between tortilla diameter and flexibility it is not possible to predict each of the desired quality parameters independently using linear models (Barros et al. 2010). Advances in high-throughput wheat breeding techniques have resulted in the need for quick means to predict tortilla making performance for large number of wheat lines in early generation. Currently, the most reliable methodology requires processing of tortilla, and this has a number of drawbacks. The main disadvantage of this approach is that it is laborious, time consuming, expensive and requires large sample size of approximately 2 kg of seed, which is normally not attainable from early generation lines.

Furthermore, in recent years research has developed instrumental techniques for rapid evaluation of wheat kernel and flour properties for tortilla quality attributes. Barros and others (2010) reported that dough rheological measurements, mixograph peak time and resistance to extension, can be used to predict tortilla diameter for a given wheat line. However, these measurements did not significantly predict tortilla flexibility. Despite having a high correlation with tortilla diameter, most of the flour properties did

not feature in the linear stepwise multiple regression models for tortilla flexibility and diameter (Barros et al. 2010).

Multivariate statistical methods designed to elicit information from simultaneous measurements of many variables acquired from wheat kernel, flour, and dough have a potential to predict the quality of tortilla, especially the diameter of the end product and flexibility during storage. These methods include principal component analysis (PCA), canonical correlation analysis and discriminant analysis (DA).

PCA are linear combinations of original measured variables with properties that capture the variation in a special manner (Johnson et al. 2007). In PCA, the PC's are new variables (principal component scores) that are uncorrelated, account for variation, possess geometric properties, and are used to point a researcher to dominant combinations of variables. The PRINCOMP procedure was used to perform principal component analysis. The correlation matrix of all the variables was used to create eigenvalues, eigenvectors and standardized principal component scores. The PCA plots were used to explore and summarize the linear relationships among the kernel, flour, dough and tortilla properties. In this study PC's were used in statistical data screening to identify outliers. These new variables were used as input for graphing and plotting to reveal abnormalities in the data set prior to analysis. In addition, the PC scores were analyzed individually to check for normality of the variables and independence of the experimental units (wheat lines).

Canonical correlation analysis (Ramos et al.) is a generalization of multiple correlations used in multiple regression problems (Johnson et al. 2007). CCA is used in

studies where there are many predictor variables and many variables to predict and a lot of relationships exist amongst the variables in each set. Canonical correlation analysis can be used for comparing two or more sets of variables (Johnson et al. 2007). The CANCECORR procedure was used to test if the canonical correlations ($\hat{\rho}$) were equal to zero using the F approximation statistics (Rao 1973). Each of the groups of variables (kernel, flour, dough and tortilla) was tested for multivariate normal distribution using the Shapiro-Wilk statistic (Davis 1979).

MATERIALS AND METHODS

The variables were examined in pairs. One pair involved kernel properties and tortilla properties. The second pair included dough and tortilla properties, a third pair included flour and tortilla properties and the last grouping comprised of examining correlations between all the kernel, flour and dough variables as a group versus tortilla properties. Since CCA variables are independent of each other, examining them in groups will enable us to identify useful correlations within the grouped variables, some of which have not been reported. A clear example would be identification of significant correlations between tortilla flexibility and tortilla textural properties which have traditionally been known to have no or very low correlation (Alviola and Awika 2010; Alviola 2007; Barros et al. 2010; Waniska et al. 2004).

Discriminant analysis (DA) was used to separate our experimental units (wheat lines) into distinct sets or groups based on the quality of tortillas they produce. The lines were classified into two predetermined groups; one group were lines that are good for tortillas and the second group were lines that are not good for tortilla production.

Kernel properties (hardness, diameter, and weight), flour properties (protein content, protein fractions, protein composition and pentosan content), and dough rheology (mixograph peak time, compression force, extensibility, loss modulus, storage modulus) were measured on 127 diverse wheat lines all of which were processed into tortillas. An extra set of 57 lines from three locations (College Station, McGregor and Chillicothe) harvested in 2009 were evaluated for kernel, flour, dough and tortilla properties and used to train and calibrate all the prediction models. Tortilla properties measured included: tortilla appearance (diameter, lightness, and specific volume), shelf stability parameters (rollability and rheological properties during punch analysis measured by texture analyzer).

Multivariate methods were used to first, identify the correlation between wheat quality properties and tortilla quality properties and, secondly, to develop models that were used to classify these wheat lines into groups representing good/bad tortilla making abilities.

Statistical Analysis

Data analysis (Principal component, canonical correlation and discriminant) analysis were done using SAS version 9.2 (SAS Institute, Cary, NC).

Data description and classification criterion

Kernel, flour, dough and tortilla properties from a total of 185 wheat lines were used. Of this sample set, 57 wheat lines possessing variations in high molecular weight glutenin sub-units (HMW-GS) at the Glu A1, Glu B1 and Glu D1 were used to train and calibrate the prediction models. This was based on the evidence that these lines had the

unique protein quality to produce tortillas of superior quality (Jondiko et al. 2012a). A total of 185 lines were evaluated for kernel, flour, dough and tortilla properties described in chapter II and used to cross validate the prediction models. A total of 23 variables were used in this study.

A suitability of categorical numbers 1 or 0 was assigned to each wheat line, where varieties with “1” were suitable to make quality tortillas (Diameter \geq 165 cm and day 16 rollability score \geq 3.0). Wheat lines not meeting this description were assigned “0” meaning they were not suitable to make quality tortillas.

Principal component analysis (PCA)

The data were standardized in order to eliminate the effect of differences in units of measurement. Principal component analysis (PCA) was conducted on these standardized variables using variance-covariance matrix to extract the eigenvalues. Principal component (PC) plots were examined to identify potential outliers. PCA was also used to determine if the data was following a multivariate normal distribution.

Canonical correlation analysis

Canonical correlation analysis was carried out on grouped variables to examine the potential correlations that have not been reported using traditional univariate analysis of the variables. The variables were grouped as kernel properties (hardness, weight and diameter), flour properties (protein, GGRatio, H_L_MW_GS_Ratio and IPP), dough properties (mixograph peak time, compression force, extensibility, elasticity, stress relaxation time (RT), relaxation force after 25 sec and 100 sec, F_25 and F_100 respectively) and canonically correlated against tortilla quality variables (diameter,

specific volume, lightness, rollability, modulus of deformation (gradient), force, distance and work to rupture at day 16 of storage).

Canonical correlations were used to find linear combinations of the kernel, flour, dough and tortilla variables which have maximum correlation with each other.

Canonical analysis were used to evaluate whether one group of variables would explain the desired tortilla quality variables better than others variables.

Discriminant analysis using logistic regression

In order to predict if these 185 wheat lines would be suitable to make good quality tortillas, a discriminant analysis were carried out on suitability of making good tortillas using all the wheat kernel, flour, and dough property variables. Variable selection was conducted to finalize the logistic regression model as a discriminant tool. The efficiency of the models was evaluated by apparent error rate discriminant rule using the formula below.

$$\textit{Apparent Error} = \frac{\textit{Number of lines misclassified}}{\textit{Total number of wheat lines used in the study}}$$

RESULTS AND DISCUSSION

Kernel properties

Kernel hardness ranged from 39 – 87 hardness index (Figure 11a). The overall mean was 65.8 with a standard deviation 10.8. Hence, the sample set covered a large range of hardness distribution. The test for normality of hardness index indicated a large skewness towards higher hardness index. This is concurrent with the fact that these lines were selected for as hard red winter wheat in the breeding program.

Kernel diameter ranged between 2.3 mm and 3.1 mm with a mean of 2.5 mm and standard deviation of 0.15 (Figure 11c). Kernel weight averaged 27.3 g with a distribution from 22 – 38 g (Figure 11b). Overall; this data covered a diverse range of wheat samples and was good for development of a prediction model for tortilla quality.

Flour properties

Flour protein content (as is) average was 14.32 % with a standard deviation of 0.88 (Figure 12a). The lowest and highest protein contents were 12.0 and 16.5 respectively. Glutenin to gliadin ratio (GGRatio) was the only protein fraction that followed a perfectly normal distribution (Kolmogorov-Smirnov $p = 0.141$). GGRatio ranged between 0.48 and 0.96 with a mean and standard deviation of 0.67 and 0.12 respectively (Figure 12b). The wheat lines used had a high molecular weight to low molecular weight glutenin sub-unit ratio between 0.25 and 0.58 with a mean of 0.33 (Figure 12c). The histogram of insoluble polymeric protein (Schipper and Weipert) shows that the wheat lines had a considerably well distributed variation in IPP from 12 to 72 % (Figure 12d). Hence, the data was representative of a diverse wheat population.

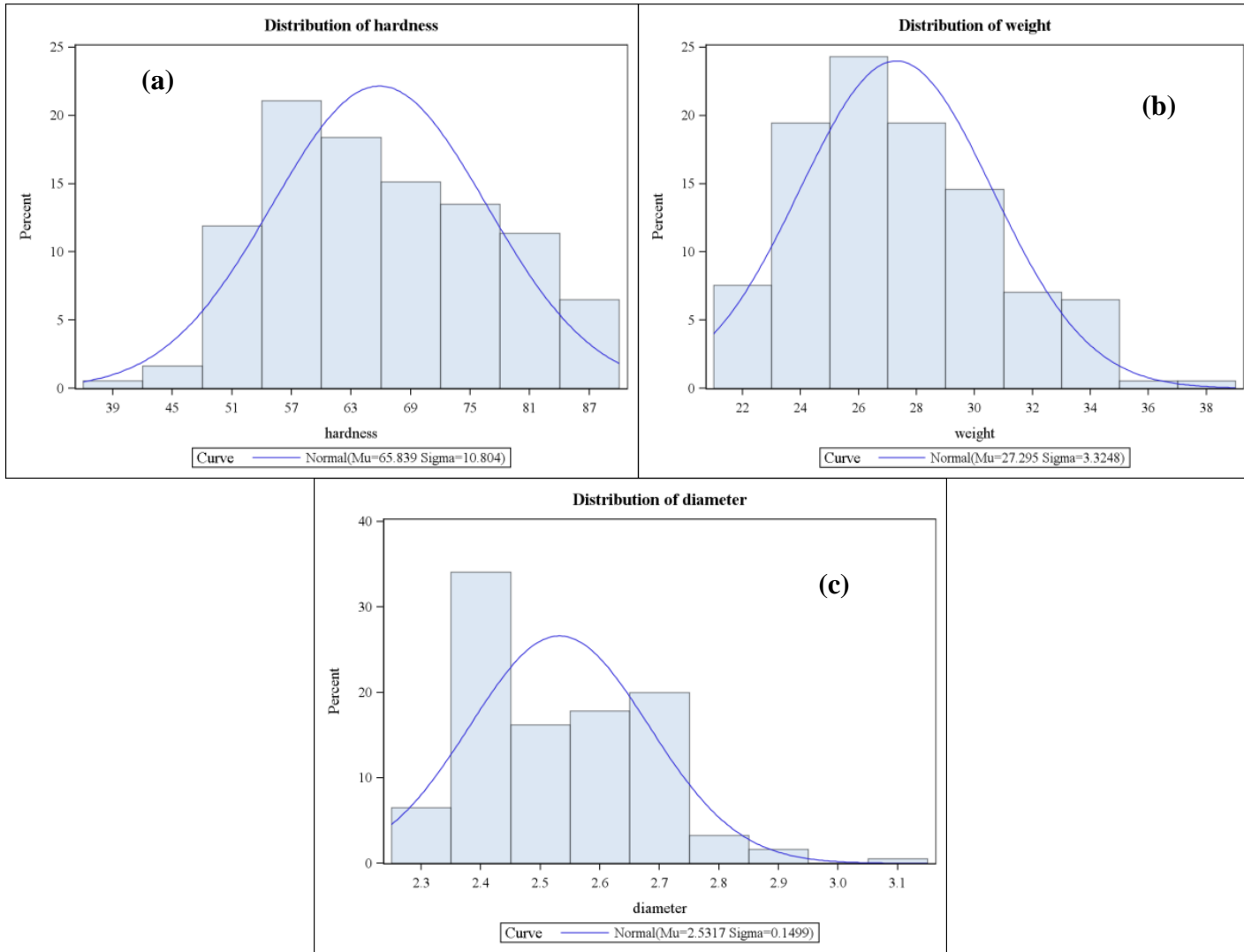


Figure 11. Histogram and normal density curves for kernel properties

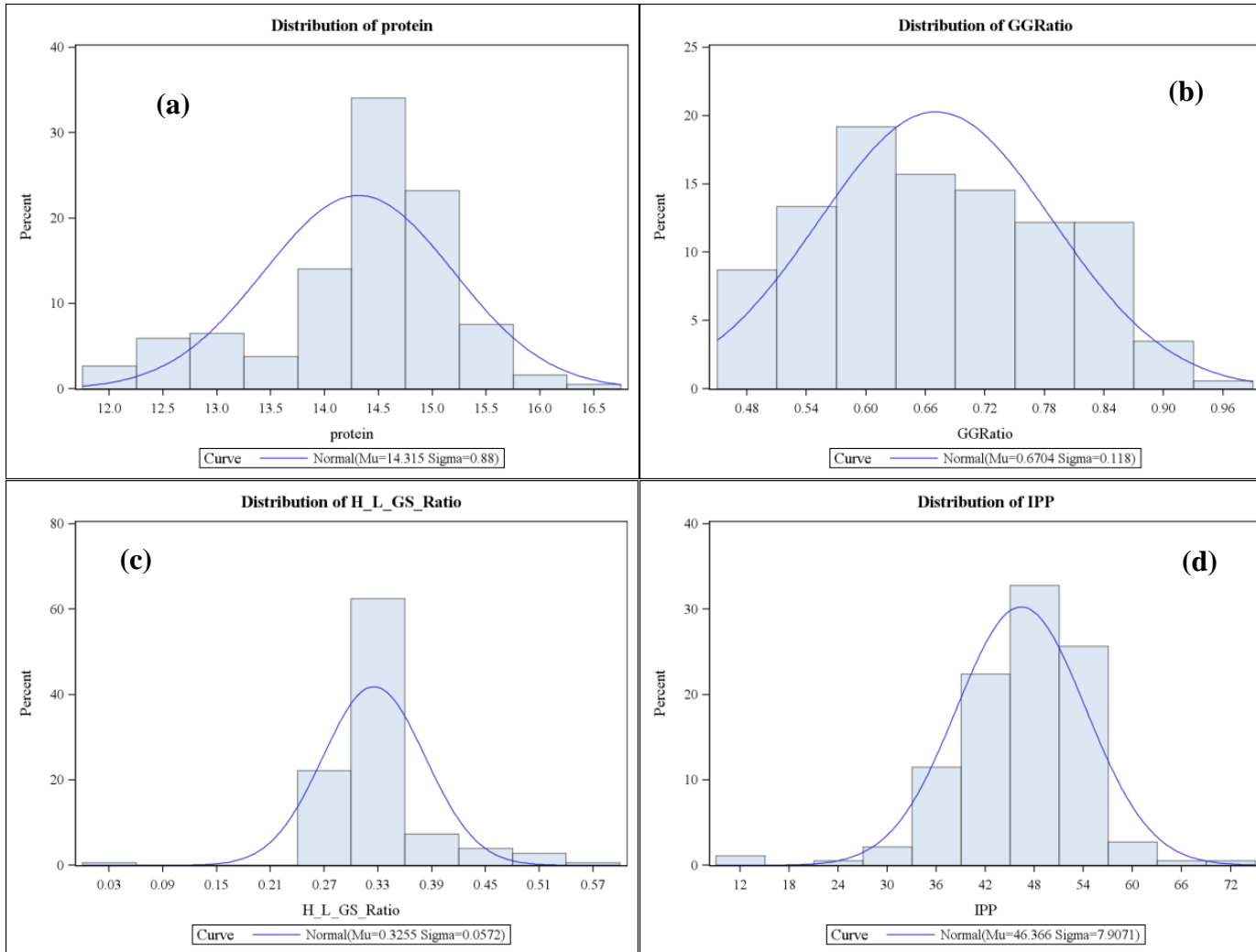


Figure 12. Histogram and normal density curves for protein, glutenin to gliadin ratio (GGRatio), high molecular weight to low molecular weight glutenin sub-unit ratio (H_L_GS_Ratio) and IPP

Dough properties

Dough extensibility test

Dough extensibility (mm) average was 66.2 mm with a standard deviation of 24 (Figure 14a). The dough extensibility (DistanceD) data had a normal distribution (Kolmogorov-Smirnov $p = 0.07$). The least extensible dough has an extensibility of 23 mm where as the most extensible was 136 mm (Figure 13a). Dough elasticity (ForceD) ranged between 0.18 and 1.14 N (Figure 13b). The distribution of work to extend is shown in Figure 13c. The dough properties of the sample size used was diverse and can be used in making inferences regarding the tortilla making performance of wheat varieties.

Dough stress relaxation test

The stress relaxation time overall mean was 1.70 (min) and ranged between 1.2 and 2.7 min (Figure 14a). The relaxation force after 25 sec of compression (F_25) range was 5.3 – 12.4 (N) (Figure 14b). Whereas the relaxation force after 100 sec of compression was between 3.2 and 8.6 (N) (Figure 14c). These were within the expected ranges for both flat and pan bread making.

Dough compression force

Dough compression force ranged from 38 to 173 (N), with an average of 94 (N) (Figure 14d). The compression force varied widely among these lines with a normal distribution (Kolmogorov-Smirnov $p > 0.150$) (Table Appendix). The samples were representative and can be used to develop a prediction model for tortilla quality.

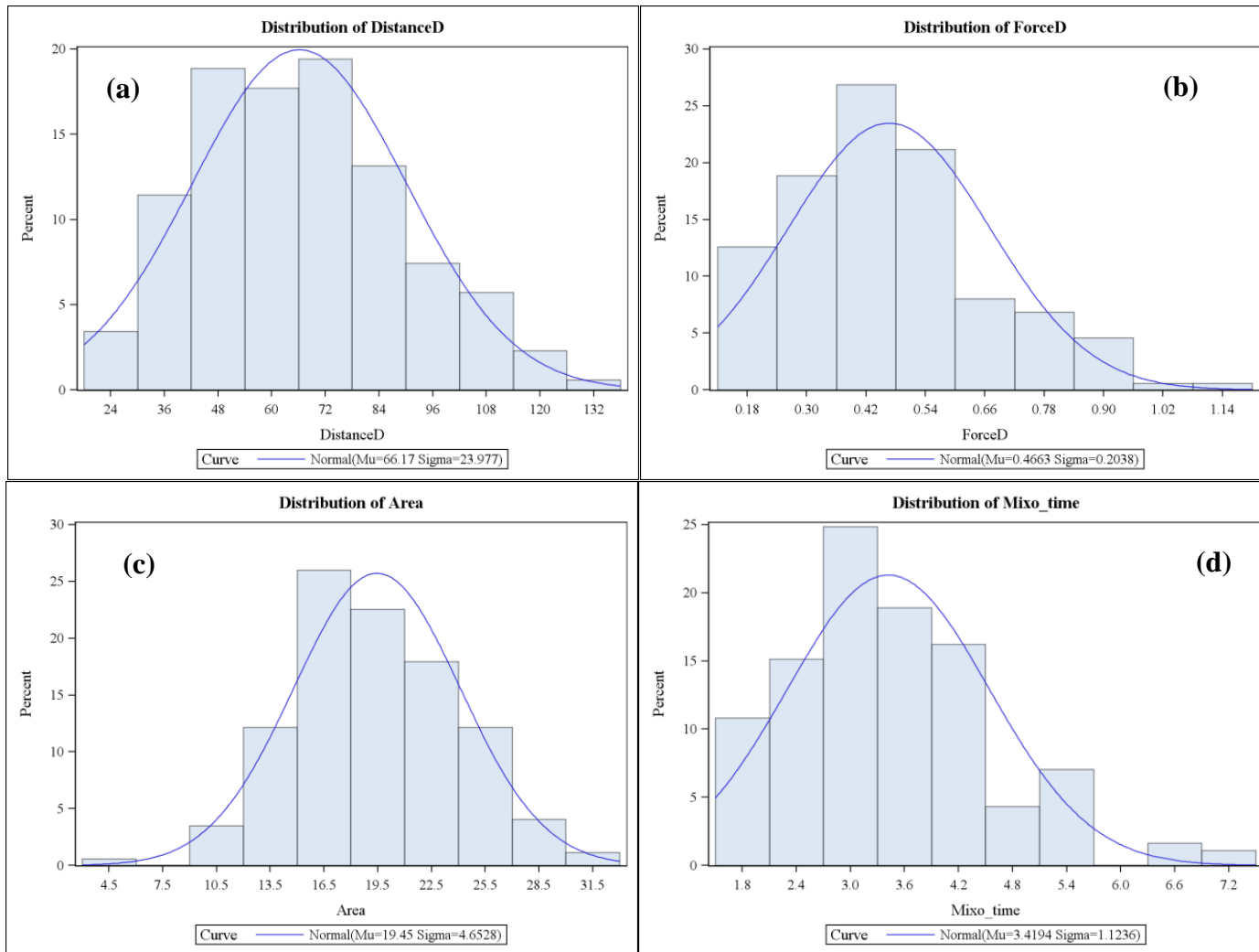


Figure 13. Histogram and normal density curves for dough extensibility variables and peak time for all the wheat lines used to develop the prediction models. Dough extensibility (DistanceD), elasticity (ForceD). Work to extend and mixograph peak time (Mixo_time)

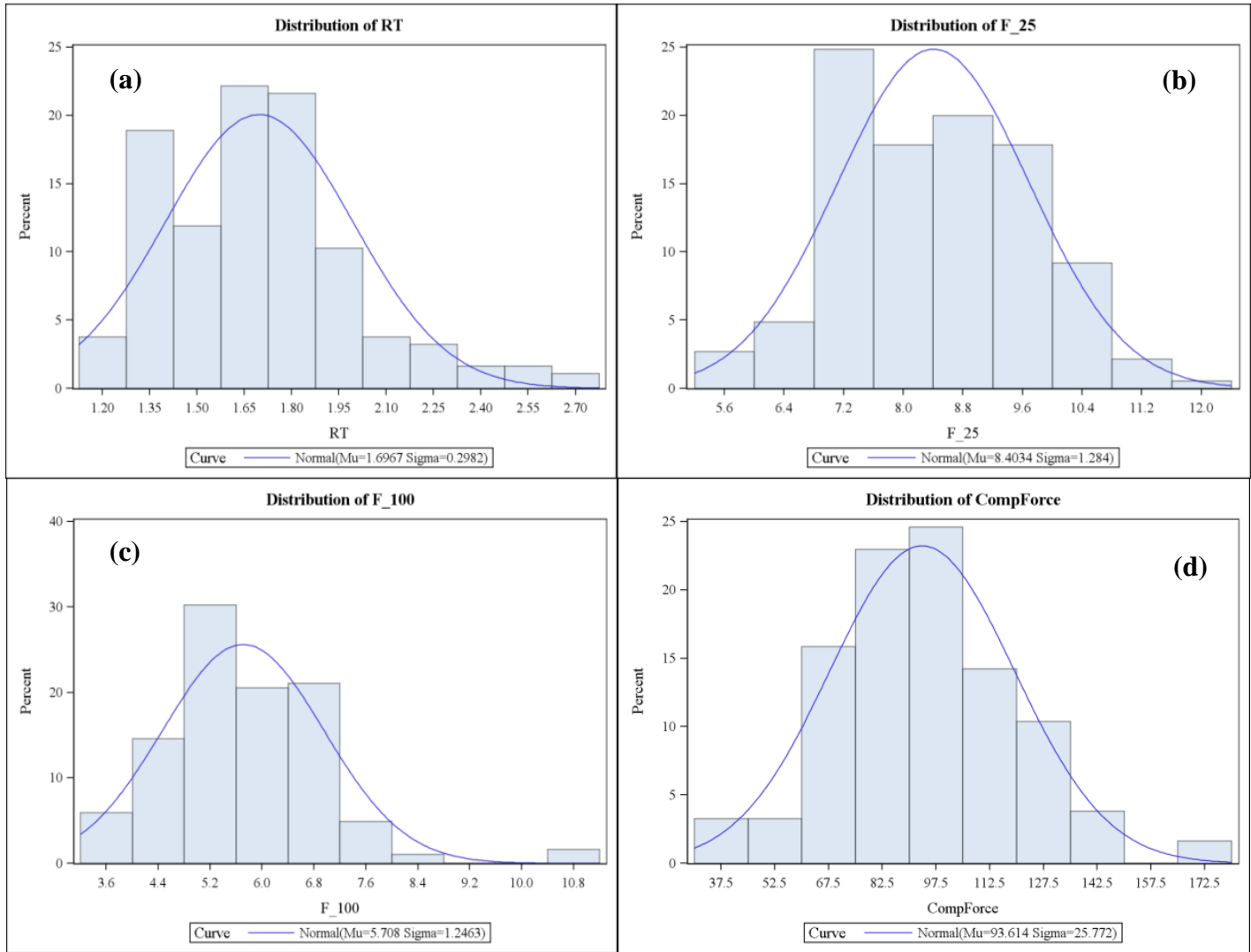


Figure 14. Histogram and normal density curves for dough stress relaxation and compression force test variables

Tortilla physical properties

Tortilla diameter (Figure 15a), rollability (Figure 15b) and specific volume (Figure 15c) had a significant normal distribution ($P > 0.01$). The lowest rollability score was 1.3 and the highest was 5.0 (Figure 15b). Tortilla Lightness ranged between 72 and 56 (Figure 16d). Based on the wide ranges in tortilla properties the data used was representative of the tortilla processing performance of wheat varieties and can be used to make predictive decisions regarding tortilla making performance of wheat varieties.

Tortilla textural properties

Tortilla modulus of deformation and force to rupture had a normal distribution ($p > 0.01$). The modulus of deformation (Gradient) data ranged from 0.54 to 1.38 N/mm (Figure 16a). Force to rupture (Force) was between 3.75 – 17.25 N (Figure 16b). Distance to rupture (Distance) was widely distributed with an average of 16.3 mm and a standard deviation of 3.7 mm (Figure 16c).

A majority of the samples used required between 25 and 100 N.mm work to rupture (work) with an overall average of 58 N.mm to rupture after 16 days of storage (Figure 16d). The tortilla texture data was representative of a diverse wheat population in terms of tortilla processing functionality and shelf stability (Jondiko et al., 2012; Alviola and Awika 2012; Barros et al., 2010; Waniska et al., 2004).

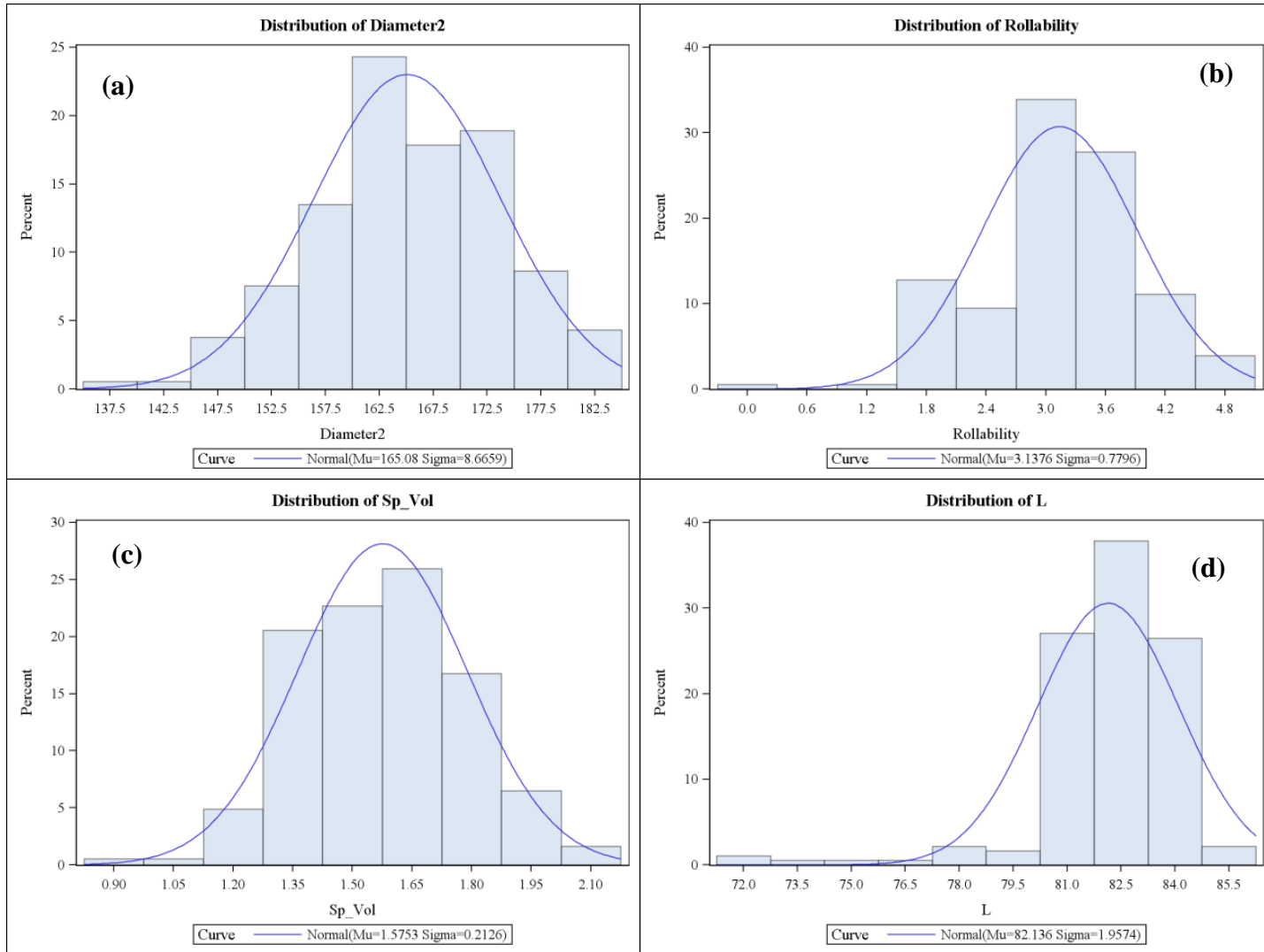


Figure 15. Histogram and normal density curves for tortilla diameter, rollability specific volume and lightness

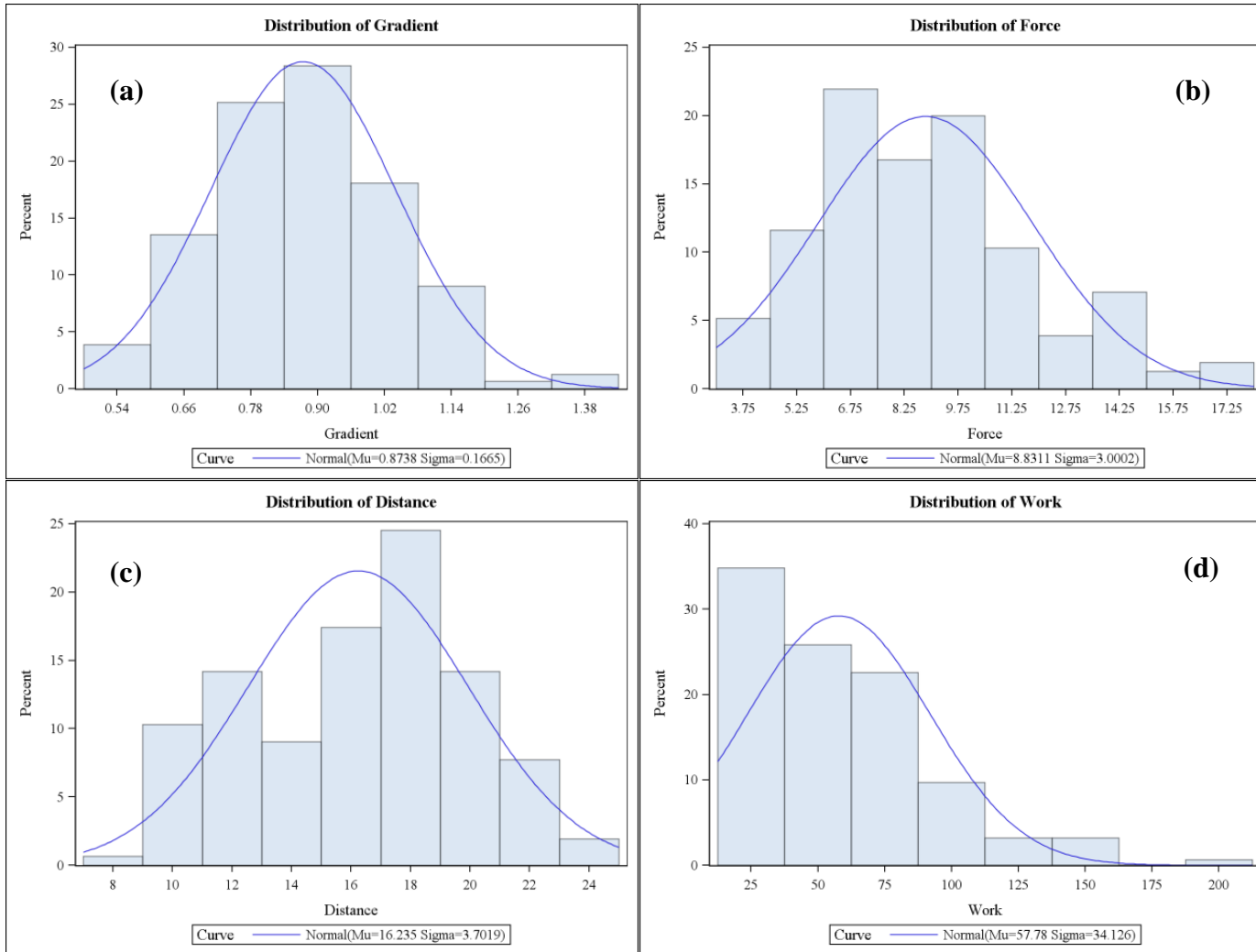


Figure 16. Histogram and normal density curves for objective tortilla texture variables. Gradient refers to modulus of deformation, force is the force to rupture, distance is the distance required to rupture the tortillas and work is the work required to rupture the tortillas as described in chapter II.

Principal component analysis (PCA)

A total of 23 principal components were produced from the variance covariance matrix of the data (Table 6).

Table 6
Principal component analysis eigenvalues of the covariance matrix using standardized variables

PC	Eigenvalue	Proportion	Cumulative
1	7.97	0.34	0.34
2	2.93	0.13	0.47
3	2.74	0.12	0.58
4	1.77	0.08	0.66
5	1.31	0.06	0.72
6	1.05	0.05	0.76
7	0.86	0.04	0.80
8	0.71	0.03	0.83
9	0.57	0.02	0.85
10	0.52	0.02	0.87
11	0.48	0.02	0.90
12	0.44	0.02	0.91
13	0.41	0.02	0.93
14	0.31	0.01	0.95
15	0.28	0.01	0.96
16	0.25	0.01	0.97
17	0.23	0.01	0.98
18	0.17	0.01	0.98
19	0.12	0.01	0.99
20	0.09	0.00	0.99
21	0.07	0.00	1.00
22	0.05	0.00	1.00
23	0.02	0.00	1.00

The first principal component (PC1) explained 34.1% of the total variance, PC2 explained 12.6%, and the third principal component explained 11.7%. The total variance in the data is 23.34 which is the sum of the eigenvalues (Table 6). 90% of the variance in the data set was attributed to the first 11 principal components (PCs).

The first 3 principal components (PCs) explained 58.4% of the variance (Table 6). The first 10 PCs explained between 2 – 80% of the variance proportion (Figure 17).

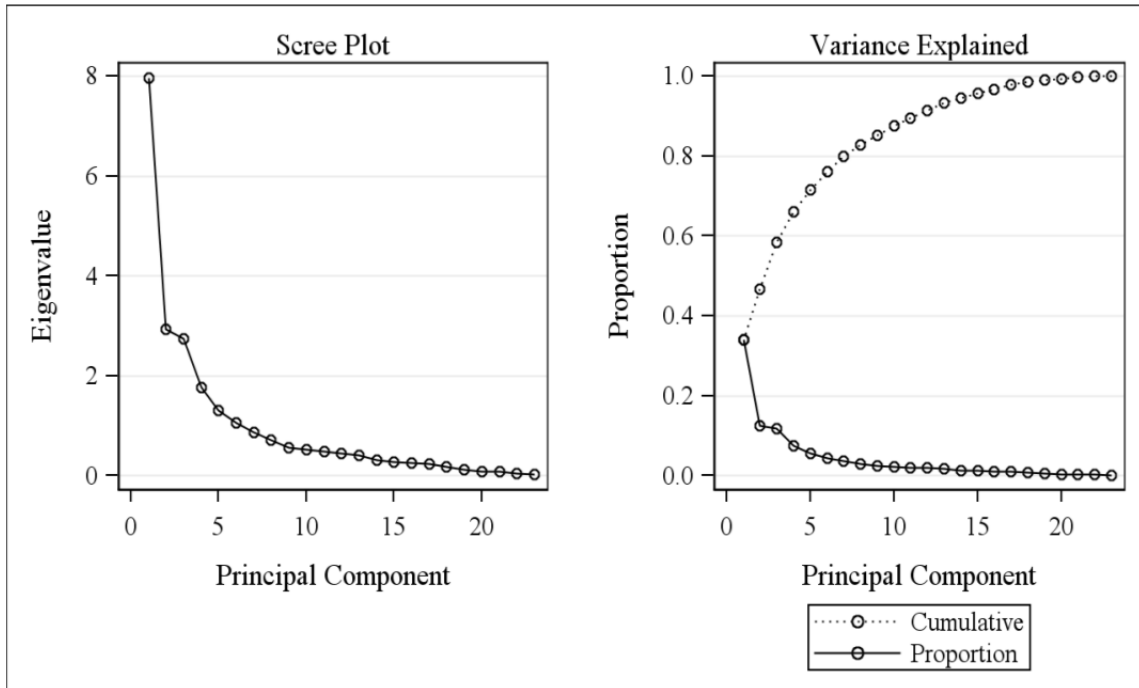


Figure 17. Scree plot of the principal components and the proportion of variance explained

The first component is a measure of dough properties (mixograph peak time - Mixo time, elasticity - forceD, extensibility - distanceD, stress relaxation force after 25 sec - F_25, and 100 sec. - F_100), flour IPP content, tortilla diameter, specific volume and tortilla texture (modulus of deformation, force, distance and work to rupture after 16 days of storage), since the first eigenvector (Prin1) shows approximately equal loadings for these variables (+ or - ~ 0.20) (Table VII).

The second principal component (Prin2) is a measure of overall kernel properties since the second eigenvector had high positive loadings on variables of kernel hardness and diameter (Table 7)

The third eigenvector had high positive loadings on flour protein content and a high negative loading on glutenin gliadin ratio (GGRatio). Variability in tortilla diameter was explained using PC1 and PC2 whereas tortilla rollability was attributed to PC3, PC4 and PC5 with loadings > 0.20 (Table 7).

Data normality

Test for normality revealed a multivariate normal distribution of the data using the first 3 principal components ($p < 0.05$) (Table 8). The Shapiro-Wilk statistic p – value must be greater than 0.05 and have a $W > 0.95$ in order to conclude that the principal components were derived from a data set that had a multivariate normal distribution.

Table 7
The loadings for the first ten principal components for all the 23 variables

Variables	Eigenvectors									
	Prin1	Prin2	Prin3	Prin4	Prin5	Prin6	Prin7	Prin8	Prin9	Prin10
hardness	-0.04	0.38*	-0.21*	-0.23*	0.30*	-0.14	0.31*	0.09	-0.28*	0.14
diameter	-0.19	0.40**	0.16	0.13	0.02	0.27	-0.14	-0.03	0.10	-0.28*
weight	-0.14	0.44**	-0.03	0.10	0.05	0.32*	-0.15	-0.05	0.19	-0.24*
protein	-0.07	-0.35*	0.45**	-0.07	0.04	0.20*	-0.03	0.06	-0.05	-0.21*
Mixo_time	0.23*	0.08	0.05	0.11	-0.05	-0.47**	0.24*	-0.44**	0.38*	-0.25*
GGRatio	0.06	0.21*	-0.51**	-0.06	-0.05	0.24*	0.06	0.02	0.13	0.05
H_L_GS_Ratio	-0.08	0.17	0.23*	0.26*	-0.04	0.18	-0.03	-0.59**	0.01	0.59**
IPP	0.22*	-0.08	0.02	0.04	0.14	-0.02	0.24*	-0.11	0.22*	0.04
ForceD	0.30*	-0.08	0.00	0.13	-0.12	0.42**	0.37*	0.07	-0.16	-0.07
DistanceD	-0.27*	-0.01	0.09	-0.21*	0.27*	-0.20*	-0.40**	0.08	0.27*	0.13
Area	0.16	-0.05	0.06	-0.02	0.51**	0.23*	0.14	0.25*	0.50**	0.12
RT	0.12	-0.01	-0.17	0.63**	0.27*	-0.20*	-0.21*	0.16	-0.13	-0.04
F_25	0.24*	0.09	0.28*	0.12	0.09	0.13	-0.08	0.06	-0.23*	0.12
F_100	0.22*	0.18	0.29*	0.29*	0.05	-0.17	0.05	0.28*	0.08	-0.04
CompForce	0.03	0.23*	0.27*	-0.24*	-0.37*	-0.03	0.19	0.19	0.27*	0.00
Diameter2	-0.31*	-0.21*	-0.12	0.09	0.00	-0.03	0.21*	0.01	0.14	0.15
Sp_Vol	-0.27*	-0.19	0.00	0.11	-0.10	0.12	0.09	0.09	0.11	0.14
L	-0.09	-0.12	-0.10	0.24*	-0.14	0.07	0.09	0.22*	0.27*	0.30*
Rollability	0.07	0.06	0.23*	-0.28*	0.42**	0.02	0.14	-0.11	-0.16	0.15
Gradient	0.25*	0.20*	0.05	-0.08	-0.27*	-0.17	-0.16	0.30*	0.01	0.39*
Force	0.32*	-0.01	-0.04	-0.15	-0.11	0.05	-0.30*	0.02	0.06	0.13
Distance	0.27*	-0.19	-0.22*	-0.12	0.11	0.14	-0.17	-0.22*	0.09	-0.05
Work	0.32*	-0.12	-0.08	-0.12	-0.06	0.15	-0.31*	-0.11	0.12	-0.02

* Loadings between 0.20 – 0.40 means acceptable variance explained. ** Loadings above > 0.40 – Implies high variance explained

Hence, the experimental design and number of wheat lines used was highly representative producing tortillas with large and diverse quality attributes from small to large diameter tortillas and also tortillas with low to high rollability scores. Multivariate normality allowed us to carry out multivariate analysis of the data set leading to robust and reliable findings and conclusions.

Table 8
Test for normality

Test	Shapiro-Wilk Statistic		p Value	
Prin1	W	0.986	Pr<W	0.2200*
Prin2	W	0.970	Pr<W	0.4591*
Prin3	W	0.981	Pr<W	0.0781*

Dough rheology, tortilla texture and diameter trends from PC1 and PC2 plot

The plot of the first two components (Figure 18) identifies sample trends based on the dough rheology and tortilla texture. Wheat lines 138, 139, 159, and 167 (Extreme right of PC1 and PC2 Plot) had higher average mixo_time, elasticity, IPP%, modulus of deformation, tortilla force, distance and work to rupture. This means that these lines required longer time for gluten development, produced highly elastic doughs that resulted in small diameter tortillas that were very shelf stable and flexible after 16 days, compared to lines 52, 67 and 82 which have lower dough and tortilla rheological properties. However, these three lines (52, 67 and 82) produce highly extensible doughs,

tortillas with very large diameters and specific volume this is evident from their location on the negative side of PC1 (Figure 18).

This phenomenon is supported by the actual data (Table A1) where lines 52, 67 and 82 tortilla diameters are 182, 179 and 184 mm respectively whereas lines 138, 139, 159 and 167 tortillas have 136, 150, 150 and 149 mm diameter respectively. (Table A1).

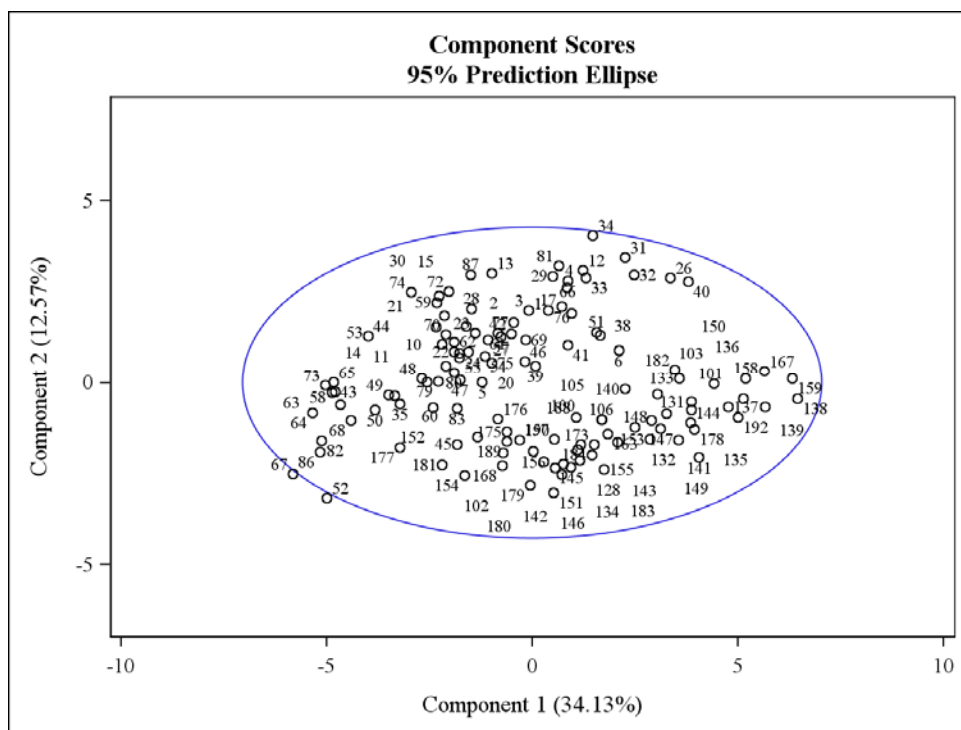


Figure 18. Plot of principal component 1 (component 1) and principal component 2 (component 2)

Dough rheology, tortilla texture and diameter trends from PC1 and PC3 plot

The plot of the first and third components identified wheat line entry number 43 as a potential outlier in the first component (Figure 19). This is attributed to the fact that

line 43 had low dough elasticity (0.16 N), average mixo_time (2.25 min), IPP% (35.79 %), modulus of deformation (0.65), tortilla force (5.34 N), distance (15.25 mm) and work (26.9 N.mm) to rupture coupled with higher extensibility (90.7 mm), tortilla diameter (182 mm) and specific volume (1.82). Lines 138, 139, 159 and 167 which were located on the extreme right of the PC1 vs PC3 plot had similar kernel, protein, GGRatio, H_L_MW_GS_Ratio compared to line 43. However these four lines had significantly high dough elasticity (> 0.7 N) compared to 0.2 N for entry 43 (Table A1).

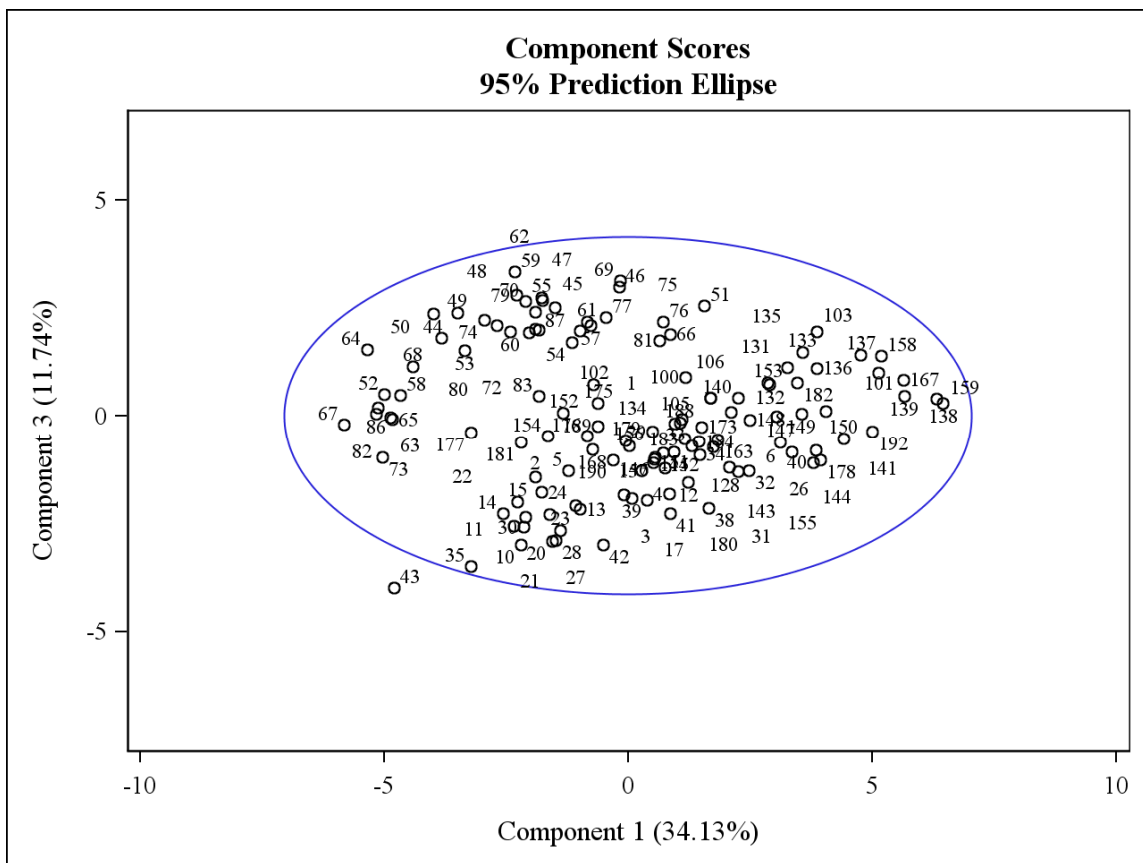


Figure 19. Plot of the first (component 1) and the third (component 3) principal components

The plot of PC1 and PC3 has the potential to identify unique wheat lines that can be used in processing of tortillas despite having high flour protein content. This is because PC1 explains dough extensibility properties whereas PC3 explains tortilla rollability and distance to rupture. Most of the lines from College Station, McGregor and Chillicothe (lines 44 – 88) are located on the left side of the PC1 vs PC3 plot. Doughs from these lines had low elasticity and produced tortillas with large diameter and acceptable rollability scores (Table A1). This confirms the findings that variation in allelic composition can be used to significantly improve tortilla making functionality of wheat varieties (Jondiko et al. 2012a).

Screening the whole data set using a plot of PC1, PC2 and PC3

After validating that the first three principal components were from a multivariate normal distribution, we conclude that lines 84 and 85 were outliers since they were located outside the 95% prediction ellipse of PC1, PC2 and PC3 (Figure 20). These two lines were eliminated from the data set. A new pairwise component score plot of the first three components justified the elimination of these two lines as discussed below (Figure 20). Based on this rationale proceeding analysis were conducted without lines 84 and 85.

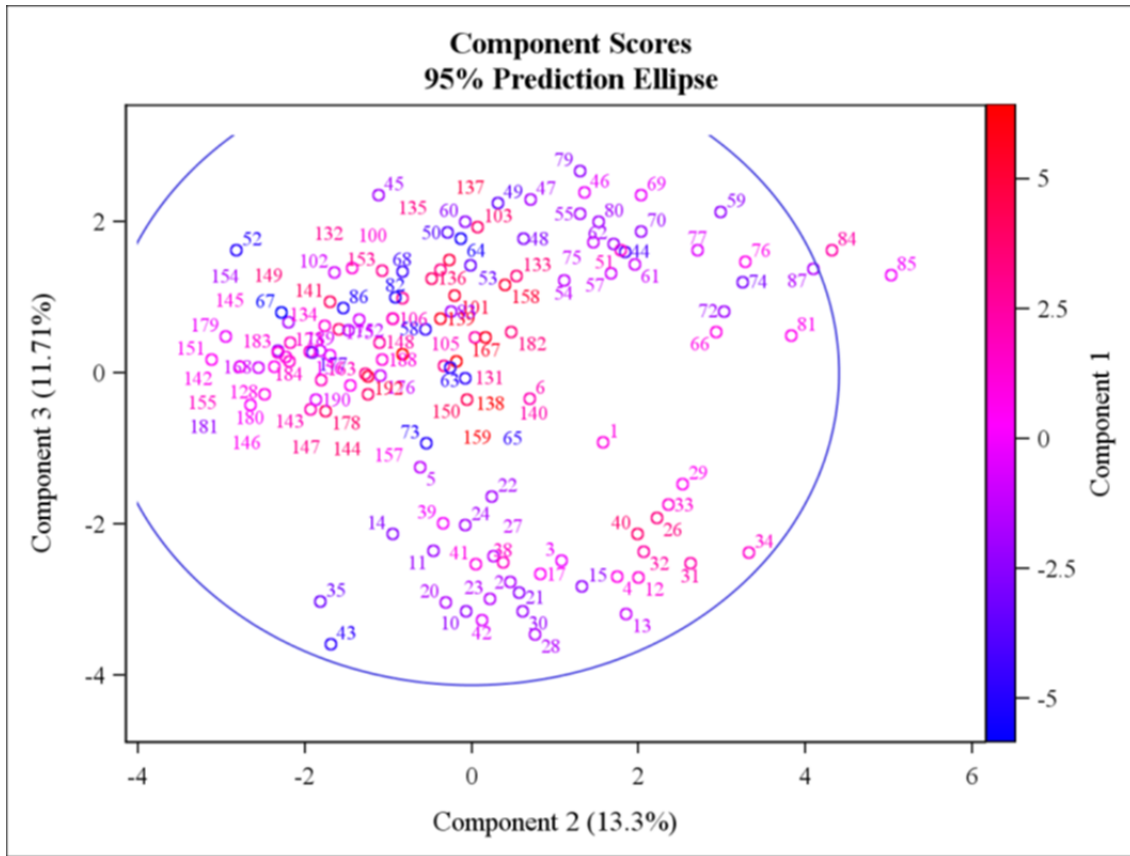


Figure 20. First three PCA Plot identifying potential outliers using standardized data

Identification of outliers

A plot on 95% predicted ellipse of the data using the first 3 PCs after removal of the identified outliers (Figure 21) revealed that the data follows a multivariate normal distribution. This is because all the remaining lines fit well into the ellipse (Figure 21). Hence, a significantly large proportion of the variance in the dataset was explained by the first, second and third principal components.

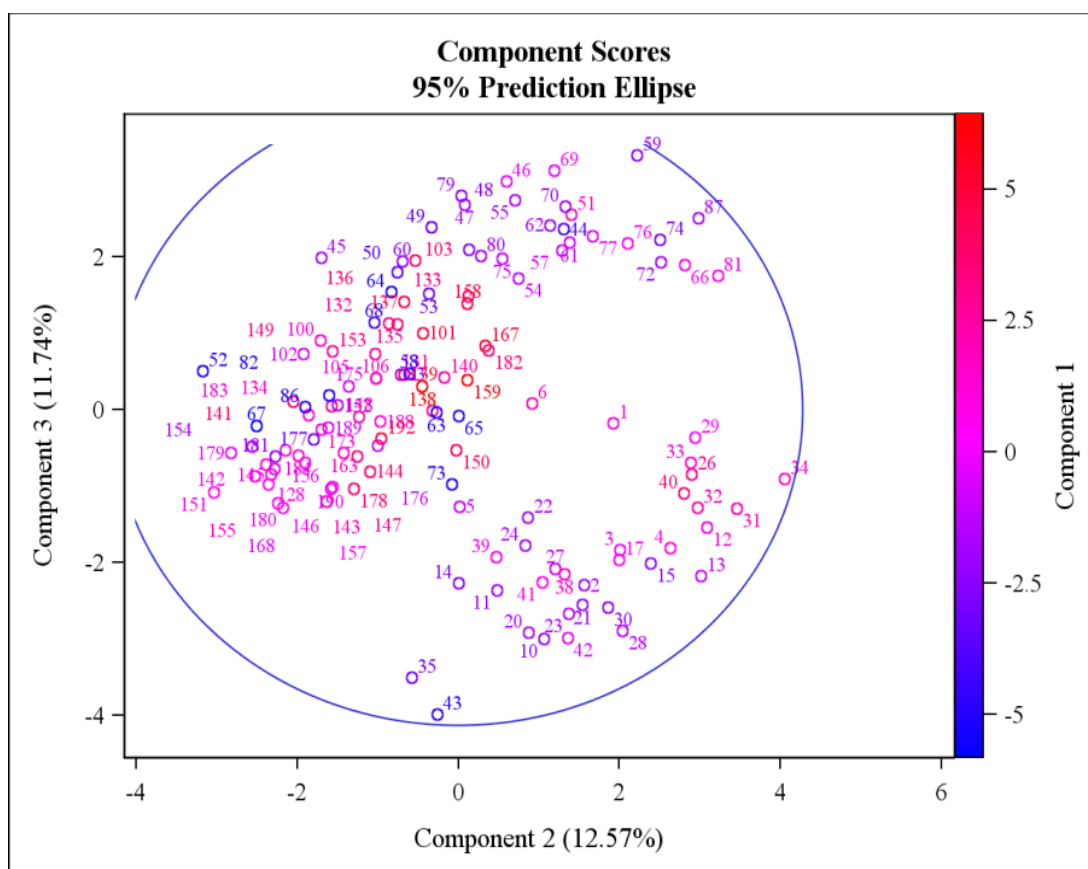


Figure 21. First three PCA Plot after removal of outliers

Principal component plots for the kernel, flour, dough and tortilla quality variables

All kernel variables positively correlate with the second principal component. This is because the kernel properties (weight, hardness and diameter) were located on the positive scale of PC2 (Figure 22).. Tortilla diameter and specific volume correlated negatively and evenly with the first principal component (They are located close to each other on the plot of PC1 vs PC2 of variables). Tortilla rollability correlated positively and evenly with both the first and second principal components (Figure 22) ; was located at equal distance from the origin. Rollability was closely related with dough rheology

variables, compression force, stress relaxation time, F_25, F_100 and tortilla modulus of deformation.

Tortilla diameter was negatively correlated with kernel properties (hardness, diameter and weight) because they are located in opposite quadrants on the PC1 vs PC2 plot (Figure 22). This is a new finding because the univariate correlation between tortilla diameter and kernel properties hardness ($r = 0.05$), diameter ($r = 0.05$) and weight ($r = 0.11$) were not significant ($P < 0.05$).

The plot of PC1 vs PC2 (Figure 22) were located on the opposite sides of the plot indicating a negative correlation between tortilla diameter and tortilla texture variables (modulus of deformation – gradient, distance, force and work to rupture) after 16 days of storage. This is in agreement with univariate analysis from which tortilla diameter was significantly correlated with modulus ($r = -0.58$), force to rupture (-0.72), distance to rupture (-0.48) and work to rupture (-0.68).

Dough elasticity (ForceD), work to extend, insoluble polymeric polymer (IPP %) content of flours and tortilla objective texture variables (force, distance and work to rupture) can be explained together. This was consistent with the significant univariate correlation coefficients ($P = <.0001$) between these variables as discussed in chapter II.

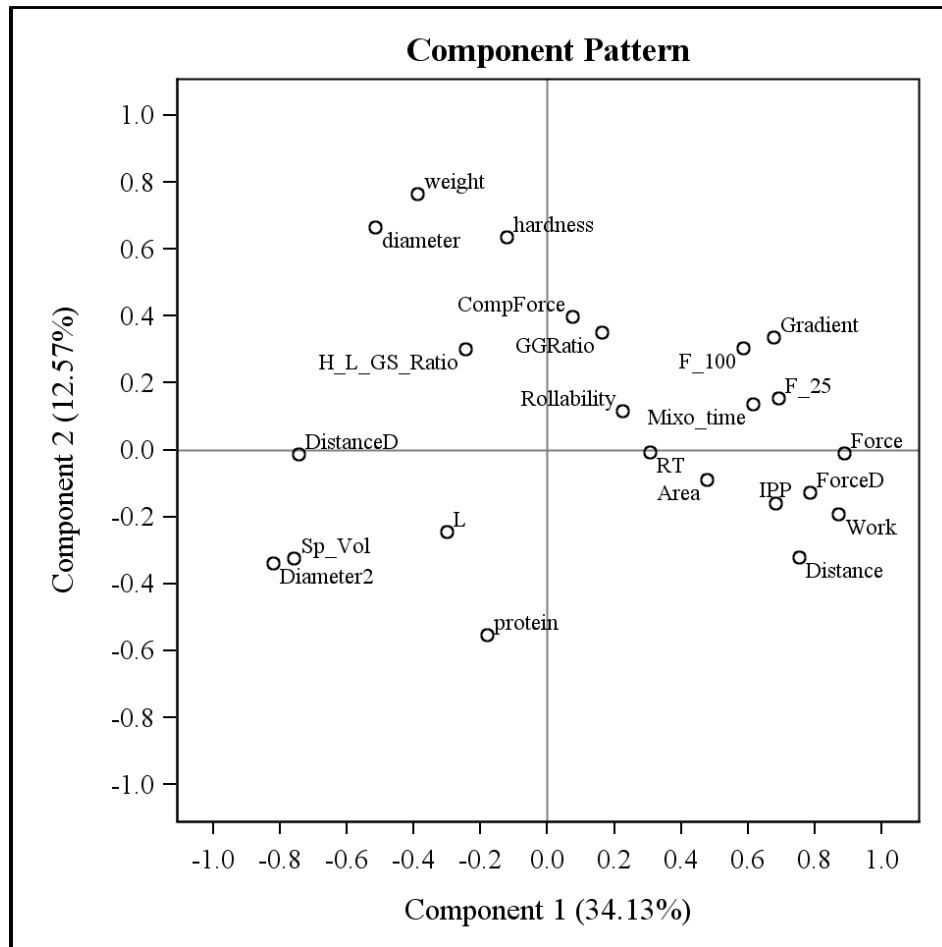


Figure 22. Component patterns of principal components 1 and 2

Variable patterns on a plot of the first and third principal components

The most striking information from the plot of the first and third principal components is that flour protein content was negatively correlated with kernel hardness and the glutenin to gliadin ratio (Figure 23). This confirms the fact that protein content is not a reliable predictor of kernel properties and flour performance for tortilla making (Waniska et. al., 2004).

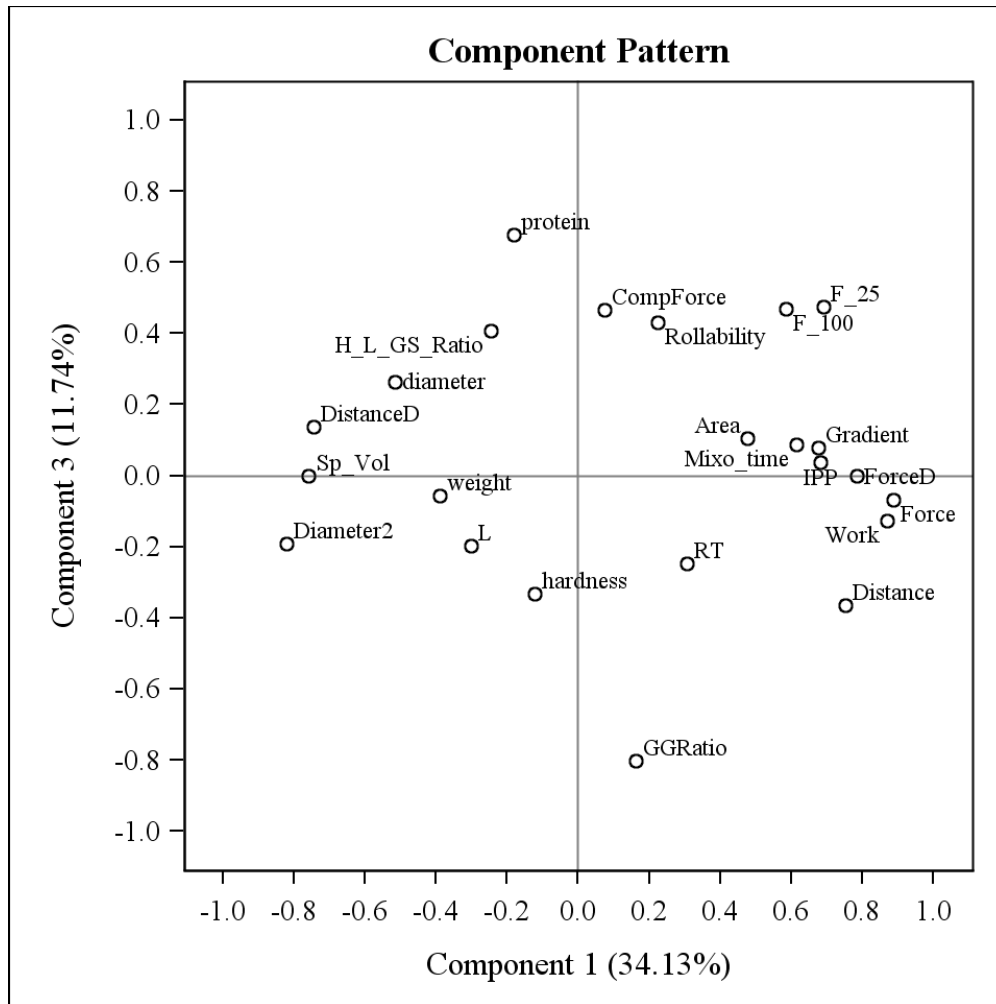


Figure 23. Component patterns of principal components 1 and 3.

Variable patterns on a plot of the second and third principal components

Overall conclusion from the component patterns PC1 vs PC2, PC1 vs PC3 and PC2 vs PC3 of all property variables was that the variance in tortilla diameter (Diameter2) is represented highly by PC1; whereas tortilla rollability at 16th day of storage (Rollability) is explained evenly by PC1 , PC2 and PC3. Tortilla diameter, lightness and specific volume can be predicted together whereas tortilla rollability can be

predicted using dough rheology variables (compression force, stress relaxation time, F₂₅, F₁₀₀, dough peak time (mixo_time), and tortilla modulus of deformation (gradient) (Figure24).

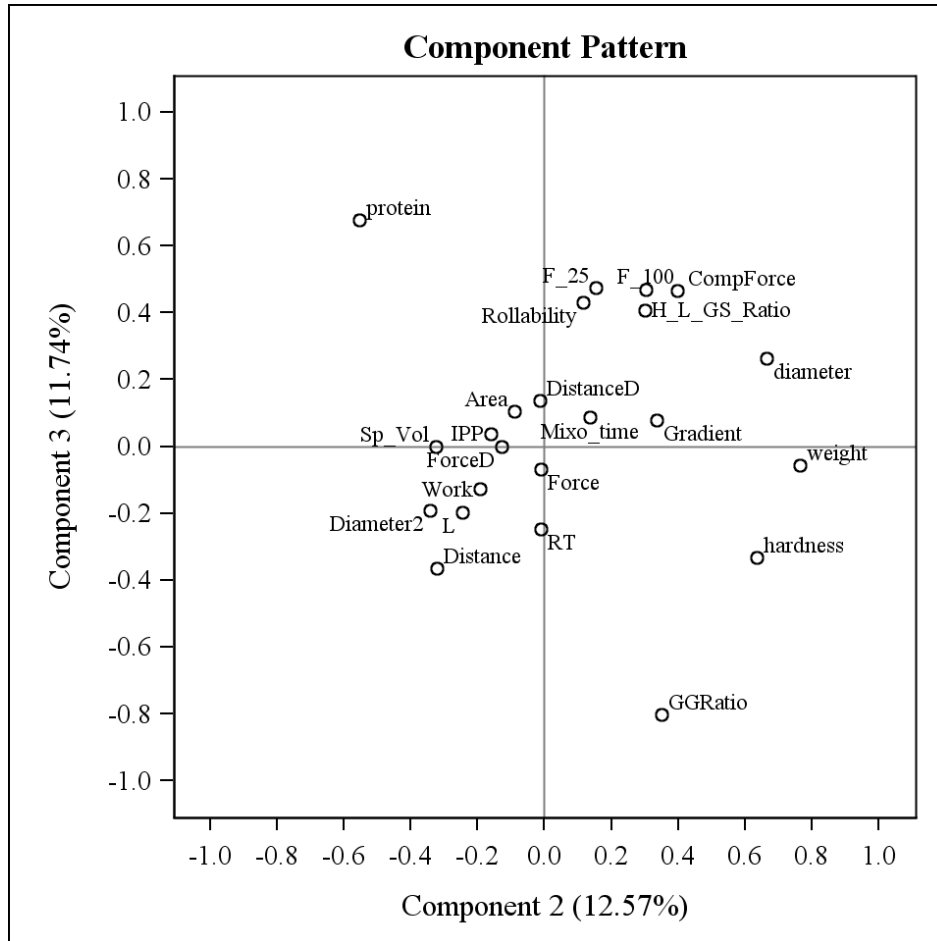


Figure 24. Component patterns of principal components 2 and 3.

Canonical correlation analysis

Canonical correlation analysis between kernel and tortilla properties

Canonical correlation analysis was used to determine whether the kernel variables are related in any way to the tortilla variables. The canonical correlation analysis of the grain properties (hardness, weight and diameter) and tortilla properties (diameter, rollability, modulus of deformation, force, distance and work to rupture) was done using the standardized data. The first two canonical correlation coefficients ($\hat{\rho}$) (ρ^{hat}) were highly significant ($P < 0.0001$).

The first significant canonical correlation, $\rho_1^{\text{hat}} = 0.75$ is the highest possible correlation between any linear combination of the wheat kernel variables and any linear combination of the tortilla quality variables. The first pair of variables were:

$$\hat{U}_1 = -0.08 \text{ Hardness} - 1.63 \text{ Diameter} + 0.86 \text{ Weight}$$

$$\hat{V}_1 = 0.41 \text{ Diameter}^2 - 0.15 \text{ Rollability} + 0.04 \text{ Gradient} + 0.81 \text{ Force} + 1.24$$

Distance - 0.81 Work (Table 9).

Kernel weight and diameter had high canonical correlation coefficients 0.86 and -1.63 respectively with \hat{U}_1 . Hence, \hat{U}_1 represented kernel physical appearance (diameter and weight) (Table 9). \hat{V}_1 is representative of tortilla textural properties force and distance to rupture. This means that a grain with small diameter and high weight produced tortillas that required longer distance to rupture i.e. highly flexible compared to tortilla made using kernels with large diameter and have low weight.

The second canonical correlation, $\rho_2^{\text{hat}} = 0.53$. Its canonical variable \hat{U}_2 was an indicator of kernel hardness since it had a high coefficient (0.99) (Table 9). The paired variables were:

$$\hat{U}_2 = 0.99 \text{ Hardness} - 0.43 \text{ Diameter} + 0.27 \text{ Weight.}$$

$$\hat{V}_2 = -0.71 \text{ Diameter} + 0.12 \text{ Rollability} + 0.48 \text{ Gradient} + 0.46 \text{ Force} + 1.80 \text{ Distance} - 3.02 \text{ Work.}$$

\hat{V}_2 represents tortilla diameter and texture (distance and work to rupture). The second canonical correlation analysis implies that grains with high hardness index produced tortilla with small diameter, longer distance to rupture and very low work to rupture meaning that these tortillas were dense and less fluffy. This could be due to the fact that hard grains produce highly elastic gluten structure that shrinks into a small tortilla during processing.

Table 9
Canonical correlation between grain and tortilla variables

Standardized Canonical Coefficients for the grain Variables			
	\hat{U}_1	\hat{U}_2	\hat{U}_3
Hardness	-0.08	0.99	0.49
Diameter	-1.63	-0.43	1.07
Weight	0.86	0.27	-1.89
Standardized Canonical Coefficients for tortilla Variables			
	\hat{V}_1	\hat{V}_2	\hat{V}_3
Diameter	0.41	-0.71	0.36
Rollability	-0.15	0.12	0.97
Gradient	0.04	0.48	0.63
Force	0.81	0.46	-1.56
Distance	1.24	1.80	-0.34
Work	-0.81	-3.02	1.84

Canonical correlation analysis between flour and tortilla properties

The first three canonical correlations were significant ($P < 0.05$) with $\rho^{\text{hat}} = 0.74$, 0.47 and 0.38 for the first, second and third canonical coefficients. The first coefficient:

$$\hat{U}_1 = -0.08 \text{ Protein} + 0.42 \text{ Mixo_time} + 0.44 \text{ GGRatio} - 0.27 \text{ H_L_GS_Ratio} + 0.45 \text{ IPP} \text{ (Table 10).}$$

Dough peak time, GGRatio and IPP have similar loadings meaning that \hat{U}_1 coefficient represents a combination of peak time, GGRatio and IPP. Correlations between tortilla variables and canonical variables of flour properties (Table 10) indicate that $> 60\%$ of variability in tortilla texture (Force, distance and work) is attributed to the first canonical correlation \hat{U}_1 . This means that wheat lines with high peak time, GGRatio and IPP produced tortillas that required high force, distance and work to rupture. These tortillas also had small diameters because of a negative correlation between \hat{U}_1 and tortilla diameter (Table 10).

The second canonical correlation, $\hat{U}_2 = 0.95 \text{ Protein} - 0.02 \text{ Mixo_time} + 0.11 \text{ GGRatio} - 0.17 \text{ H_L_GS_Ratio} + 0.47 \text{ IPP}$.

Protein had the largest loading and hence the second correlation is an indicator of flour protein content. The correlations between tortilla variables and the second canonical correlation show that 26% of variability in tortilla flexibility is attributed to \hat{U}_2 (Table IX). Hence, protein content is responsible for about 26% variability in tortilla rollability. This is an important relationship that has not been reported before. The univariate correlation between protein content and rollability was not significant ($r = -0.06$). However, canonical correlation analysis identifies the relationship between tortilla

flexibility and flour protein. This can be attributed to the subjective nature of the rollability test whose effect is corrected using the canonical variables leading to an effective way to identify the contribution of IPP on tortilla rollability.

Table 10
Canonical correlation between flour and tortilla variables

Standardized Canonical Coefficients for the flour Variables					
	\hat{U}_1	\hat{U}_2	\hat{U}_3	\hat{U}_4	\hat{U}_5
Protein	-0.08	0.95	0.39	0.88	-0.13
Mixo_time	0.42	-0.02	-0.46	0.76	0.56
GGRatio	0.44	0.11	0.73	0.90	-0.56
H_L_GS_Ratio	-0.27	-0.17	-0.47	0.39	-0.77
IPP	0.45	0.47	-0.23	-0.63	-0.61
Correlations Between the tortilla Variables and the Canonical Variables of the flour Variables					
	\hat{U}_1	\hat{U}_2	\hat{U}_3	\hat{U}_4	\hat{U}_5
Diameter	-0.49	-0.03	0.26	0.01	0.02
Rollability	0.01	0.26	-0.16	-0.07	-0.01
Gradient	0.48	-0.06	-0.21	-0.01	0.05
Force	0.63	0.14	-0.10	-0.03	0.01
Distance	0.65	0.19	0.09	0.02	-0.01
Work	0.61	0.22	-0.05	0.02	0.00

Canonical correlation analysis between dough and tortilla properties

The first five canonical correlations were significant ($P < 0.05$) with $\rho^{\text{hat}} = 0.82$, 0.68, 0.54, 0.38 and 0.29 for the first, second, third fourth and fifth canonical coefficients respectively. The first coefficient (\hat{U}_1) is an indicator of stress relaxation force (F_25 and F_100) these two variables have highest loading on Table 11. \hat{U}_1 was negatively correlated with tortilla diameter ($r = -0.73$) (Table 11).

Table 11
Canonical correlation between dough and tortilla variables

Standardized Canonical Coefficients for the dough Variables						
	\hat{U}_1	\hat{U}_2	\hat{U}_3	\hat{U}_4	\hat{U}_5	\hat{U}_6
Elasticity	0.13	0.29	0.94	1.71	-1.05	0.07
Extensibility	0.05	-0.36	1.33	0.88	-1.06	-0.46
Area	-0.06	0.55	0.17	-0.76	0.75	0.37
RT	-0.14	0.18	-0.56	-0.10	-0.41	-0.54
F_25	0.56	0.26	-0.03	-0.92	-1.03	-0.44
F_100	0.43	-0.76	0.30	0.83	1.09	-0.25
CompForce	0.26	-0.22	-0.38	-0.31	-0.40	0.55

Correlations Between the tortilla Variables and the Canonical Variables of the dough Variables						
	\hat{U}_1	\hat{U}_2	\hat{U}_3	\hat{U}_4	\hat{U}_5	\hat{U}_6
Diameter	-0.73	-0.24	0.10	0.05	0.00	0.03
Rollability	0.34	0.10	0.37	-0.19	0.06	0.05
Gradient	0.58	0.05	-0.21	0.09	0.07	0.09
Force	0.51	0.42	-0.13	0.05	-0.06	0.06
Distance	0.16	0.65	-0.04	0.07	0.02	-0.01
Work	0.43	0.51	-0.04	0.13	-0.05	0.02

This implies that wheat lines that produced doughs with high relaxation force after 25 and 100 seconds of compression produced tortilla with small diameters signifying strong gluten structure of doughs. This implies that stress relaxation force indicates more elastic dough structure that could shrink back during processing or require a higher force to spread during hot pressing.

Tortilla rollability had the highest correlation with the third coefficient (\hat{U}_3) of dough variables (Table 11). This coefficient is representative of dough elasticity with the highest loading (0.94) on Table 11. The third coefficient was correlated with tortilla rollability (0.37) (Table 11). Univariate linear correlation between dough elasticity and

rollability was $r = 0.15$ (Table 4). Hence, canonical correlation reveals that doughs with high elasticity produce tortilla that retain their flexibility over storage and have high rollability score. This is consistent with finding by Waniska and others (2004).

Discriminant analysis

Discriminant rule was established. The response (Suitability) criterion was equal to one (= 1) for each line that was good for tortilla processing and Zero (0) for each of the lines that had inferior tortilla processing ability. Variable selection carried out using forward, backward and stepwise variable selection resulted into three models. Forward selection procedure resulted in model with 11 of the 23 variables (Table 12):

Predicted suitability = $-1.584 - 0.05 * \text{Hardness} + 9.734 * \text{Diameter} - 0.496 * \text{Weight} - 1.137 * \text{Protein} + 7.274 * \text{GGRatio} - 9.2731 * \text{H_L_GS_Ratio} - 0.053 * \text{IPP} + 1.5851 * \text{ForceD} - 0.171 * \text{Area} + 1.354 * \text{F_25} + 0.029 * \text{CompForce}$ (Table 12).

The three kernel properties (hardness, diameter and weight), four flour protein variables (protein, GGRatio, H_L_GS_Ratio and IPP) and four dough rheology variables (Extensibility, work to extend, relaxation force after 25 sec and compression force) were used to classify and cross validate the model using five nearest neighbors (Table Appendix). The apparent error rate for this model was 0.30 (Table 14). This model correctly classified 67% and 72% of the Good and Poor wheat lines with an overall accuracy of 70%.

Table 12
Forward selection procedure analysis of maximum likelihood estimates

Parameter	DF	Estimate	Standard	Wald Chi- Square	Pr > ChiSq	Exp(Est)
Intercept	1	-1.584	8.32	0.04	0.85	0.21
Hardness	1	-0.050	0.03	3.48	0.06	0.95
Diameter	1	9.734	4.50	4.68	0.03	16878.63
Weight	1	-0.496	0.21	5.82	0.02	0.61
Protein	1	-1.137	0.42	7.20	0.01	0.32
GGRatio	1	7.274	3.06	5.64	0.02	1442.90
H_L_GS_Ratio	1	-9.273	5.39	2.96	0.09	0.00
IPP	1	-0.053	0.04	2.02	0.16	0.95
ForceD	1	1.585	1.74	0.83	0.36	4.88
Area	1	-0.171	0.07	6.44	0.01	0.84
F_25	1	1.354	0.31	19.19	<.0001	3.87
CompForce	1	0.029	0.01	7.08	0.01	1.03

Backward elimination procedure selected 9 variables for the prediction model (Table 13). The backward elimination model was as follows:

Predicted suitability = $-5.3 - 0.052 \cdot \text{Hardness} + 9.92 \cdot \text{Diameter} - 0.479 \cdot \text{Weight} - 1.075 \cdot \text{Protein} + 7.846 \cdot \text{GGRatio} - 9.976 \cdot \text{H_L_GS_Ratio} - 0.185 \cdot \text{Area} + 1.41 \cdot \text{F_25} + 0.028 \cdot \text{CompForce}$ (Table 13).

This model had a slightly lower apparent error rate (0.26) compared to the forward selection model. It missclassified 31 % and 22 % of the good and poor wheat lines respectively. It correctly classified 74 % of the wheat lines (Table 14).

Table 13
Backward elimination procedure analysis of maximum likelihood estimates

Parameter	DF	Estimate	Standard	Wald Chi- Square	Pr > ChiSq	Exp(Est)
Intercept	1	-5.300	7.76	0.47	0.49	0.01
Hardness	1	-0.052	0.03	3.90	0.05	0.95
Diameter	1	9.920	4.39	5.10	0.02	20339.65
weight	1	-0.479	0.20	5.70	0.02	0.62
protein	1	-1.075	0.41	6.76	0.01	0.34
GGRatio	1	7.846	2.92	7.24	0.01	2554.50
H_L_GS_Ratio	1	-9.976	5.45	3.35	0.07	0.00
Area	1	-0.185	0.06	8.58	0.00	0.83
F_25	1	1.410	0.28	24.92	<.0001	4.10
CompForce	1	0.028	0.01	6.71	0.01	1.03

Table 14
Classification summary for cross-validation using five nearest neighbors

From Type	Forward selection			Backward elimination			Stepwise selection		
	GOOD	POOR	Total Error	GOOD	POOR	Total Error	GOOD	POOR	Total Error
GOOD	41	20	61	42	19	61	46	8	54
	67.21	32.79	100	68.85	31.15	100	85.19	14.81	100
POOR	25	63	88	19	69	88	13	60	73
	28.41	71.59	100	21.59	78.41	100	17.81	82.19	100
Total	66	83	149	61	88	149	59	68	127
	44.3	55.7	100	40.94	59.06	100	46.46	53.54	100
Priors	0.41	0.59		0.41	0.59		0.43	0.57	
Overall Apparent Error rates									
Rate	0.33	0.28	0.30	0.31	0.22	0.26	0.15	0.18	0.17

Stepwise variable selection model had seven variables:

Predicted suitability = - 10.149 -0.189*Weight + 10.583*GGRatio - -0.052*IPP
-0.219*Area + 1.401*F_25 + 0.355*F_100 + 0.021*CompForce (Table 15).

This model had one variable each from the kernel properties (Weight), dough extensibility test (Work to extend), and compression test (Compression force) plus two

variables from both the flour protein fractions (GGRatio and IPP) and stress relaxation tests (F_25 and F_100). The stepwise model had a significantly lower apparent error rate (0.17) compared to forward and backward elimination models. It correctly classified 83% of the wheat varieties based on their tortilla processing functionality (Table 14).

Table 15
Stepwise selection procedure analysis of maximum likelihood estimates

Parameter	DF	Estimate	Standard Error	Wald Chi-Square	Pr > ChiSq	Exp(Est)
Intercept	1	-10.149	3.35	9.18	0.00	0.00
Weight	1	-0.189	0.08	5.99	0.01	0.83
GGRatio	1	10.583	2.39	19.53	<.0001	39446.00
IPP	1	-0.052	0.03	2.30	0.13	0.95
Area	1	-0.219	0.06	11.82	0.00	0.80
F_25	1	1.401	0.32	19.39	<.0001	4.06
F_100	1	0.355	0.24	2.17	0.14	1.43
Compression force	1	0.021	0.01	4.75	0.03	1.02

Practical relevance, potential application and future research direction

The best model among the three was selected based on two criteria: Practicality and efficiency. The forward selection model was the least efficient with an apparent error rate of 0.30. This model requires 11 variables which include all the kernel properties, protein fractions and all the dough rheology tests. This model is time and labor intensive and costly. Its implementation will require evaluation of some variables that have not been significantly known to be good predictors of either tortilla diameter or rollability. The high apparent error rate means that this model can predict tortilla making

ability of wheat varieties with seventy percent accuracy. Hence, this is not a practically feasible and efficient model.

Backward elimination model has nine variables. Compared to the forward selection model it does not require IPP and dough elasticity (ForceD). However, elimination of these two variables does not significantly decrease the apparent error rate (0.26) compared to forward selection. It misclassified 38 of the wheat varieties whereas forward selection model misclassified 45 of the wheat lines. Similarly this model will be as time intensive and costly as the forward selection model this is because the variables selected require similar time commitment and sample size like the forward selection model. This model has a prediction accuracy of 74% which is not robust enough to be used in determining the tortilla making functionality of wheat varieties.

Stepwise variable selection model has seven variables. These include kernel, flour and dough properties. This is a significant decrease in the number of variables compared to the forward selection model. The stepwise parameters are determined using small sample size and would require a significantly low time to achieve. For instance elimination of protein content and the high molecular weight to low molecular weight glutenin sub-unit ratio means that implementation of this model will eliminate the need for one HPLC column. Overall, this will decrease cost of analysis by a significant percentage. The stepwise model was the most efficient with an apparent error of 0.17. This model can be used to predict the suitability of a wheat line for tortilla production with an accuracy of 83%.

The stepwise model was thus used to classify and cross validate the data and the following is a summary of the results.

Classification summary

There were a total of 54 lines that had superior tortilla making properties and 73 lines that produced poor quality tortillas (Figure 25). Eight of the good varieties were misclassified as poor performing lines whereas thirteen of the poor lines were misclassified. A closer look at the misclassified lines reveals six of the suitable lines that were misclassified as poor performing lines that were from TIA, which have been shown to have great tortilla making ability (Jondiko et al. 2012a). These lines possessed variability in the composition of the HMW-GS alleles of the Glu A1, Glu D1 and Glu D1 (Table 16) which can be attributed to the superior tortilla quality from these lines. For instance, line 86 was misclassified it possessed deletion of sub-unit 18 and 10 at the GluB1 and GluD1 loci this contributed to the decreased dough strengthening associated with presence of 5+10 and consequently the dough from this line was highly extensible and required less work to extent (4.1 N.mm) resulting in production of large diameter tortillas (178 mm) that had low rollability score (2.0) (Table 16). Lines possessing 1/17+18/5+10 (lines 54, 57, 65, 87) produced large diameter tortillas this agrees with findings by Jondiko et al. (2012a) that the absence of sub-unit 2* at GluA1 results in decreased dough elasticity.

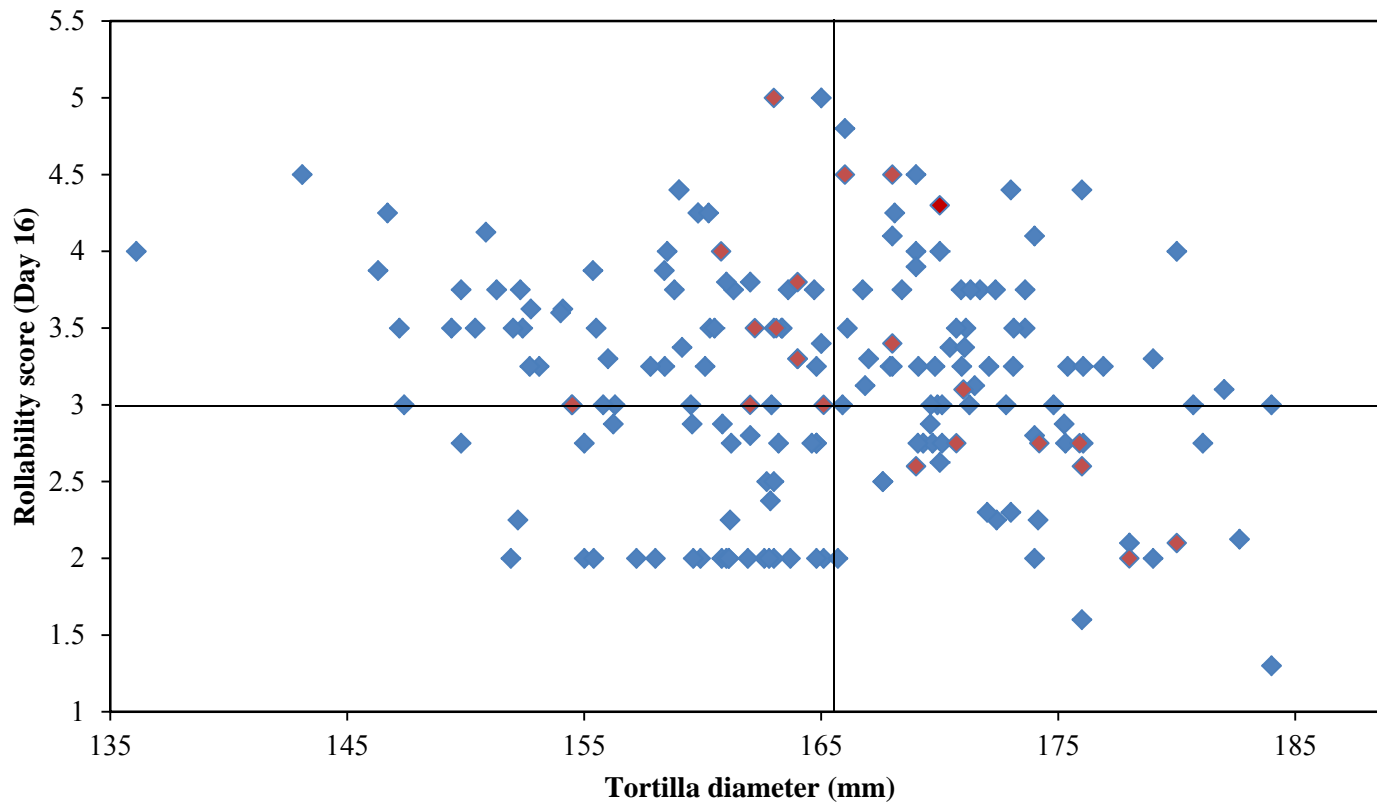


Figure 25. Relationship between tortilla diameter and rollability score (day 16). Quadrant A: good shelf-stability, poor diameter; B: acceptable diameter and shelf-stability; C: poor diameter and shelf-stability D: good diameter, poor shelf-stability.

Table 16
List of lines misclassified by the stepwise prediction model variables

Line	Name	¹ Type	weight	GGRatio	IPP	Area	F_25	CompForce	Diameter	Rollability	² suitability	³ INTO	⁴ Glu A1/B1/D1
153	TX1112-62	GOOD	25.3	0.58	53.8	20.2	9.8	100	165	3.0	1	POOR	ND
21	TX09A001251	GOOD	31.4	0.83	46.2	14.4	7.4	66.1	171	3.0	1	POOR	ND
54	114CS	GOOD	26.6	0.60	44.9	18.7	8.6	109.5	170	4.3	1	POOR	1/17+18/5+10
57	118CS	GOOD	31.2	0.52	47.7	23.3	9.5	95.9	168	4.5	1	POOR	1/17+18/5+10
70	215McG	GOOD	30.3	0.53	43.3	20	9	117.8	166	4.5	1	POOR	2*/7+9/5+10
74	31CH	GOOD	38.1	0.56	48.2	18.4	8.9	95.9	168	3.4	1	POOR	2*/17+18,7/2+12
75	32CH	GOOD	28.6	0.50	46.2	20.7	9.6	81.6	164	3.3	1	POOR	2*/20x+20y/5+10
79	38CH	GOOD	29	0.49	41.2	14.6	9.5	96.6	171	3.1	1	POOR	2*/17/2+12
149	TX1112-58	POOR	23.2	0.69	58.2	19.7	10.1	87.2	162	3.5	0	GOOD	ND
150	TX1112-59	POOR	28.7	0.71	54.7	26.6	10.1	83.2	155	3.0	0	GOOD	ND
157	TX1112-68	POOR	27.1	0.65	46.9	19.1	8.6	46.3	164	3.8	0	GOOD	ND
1	TAM W-101	POOR	33.4	0.74	41.4	19.6	8.9	83.4	161	4.0	0	GOOD	ND
3	TAM 112	POOR	32.4	0.78	46.3	16.3	7.8	99	163	3.5	0	GOOD	ND
35	TX09D1119	POOR	26.2	0.87	37.4	13.9	6.0	65.3	176	2.8	0	GOOD	ND
46	15CS	POOR	27.2	0.52	49.2	24	10.1	107.6	163	5.0	0	GOOD	2*/7+9/5+10
65	29McG	POOR	33	0.75	36.2	10.1	7.0	98.1	176	2.6	0	GOOD	1/17+18/5+10
77	36CH	POOR	29	0.57	46.7	17.7	10.5	98.4	162	3.0	0	GOOD	2*/17+18/5+10
80	39CH	POOR	30.6	0.56	35.5	14.2	9.5	106.4	180	2.1	0	GOOD	1/17+18/5+10
83	313CH	POOR	26.4	0.57	42	17.8	8.2	104.6	174	2.8	0	GOOD	1,2*/17+18/2+12
86	316CH	POOR	25.7	0.51	36.8	4.1	5.9	85.6	178	2.0	0	GOOD	2*/17,7/5
87	318CH	POOR	34.1	0.52	46.7	17.2	7.2	127.3	169	2.6	0	GOOD	1/17+18/5+10

¹Type: Good means number of lines that were actually suitable for tortilla based on raw data, Poor means unsuitable based on actual data

²Suitability score (1 – Diameter > 165mm and Rollability score ≥ 3 and 0 - Diameter < 165mm or Rollability < 3)

³INTO is the classification assigned to a line using the model.

⁴High molecular weight glutenin sub-unit composition on long arm of loci GluA1, GluB1 and GluD1

Combination of 2*/7+9/5+10 on lines 70 and 46 resulted into dough with high elasticity and high dough compression force (117 and 108 N respectively) resulting in the high tortilla flexibility scores for this lines (Table 16). This could have contributed to the misclassification.

Low dough compression force (< 70N) resulted in misclassification of lines 21, 157, and 35 (Table 16). Based on the stepwise model these lines had a negative suitability score and hence classified as poor.

Diagnosis of suitability using actual data and model classification summary

The lines from TIA population had the highest proportion of varieties that were suitable for tortilla processing (Table 16). This is was because these lines were specifically bred for flat bread production (Jondiko et al. 2012a). As expected most of the TXE/UVT and TAM1112 lines were not suitable for making good quality tortillas (Table A4). These lines (TXE/UVT and TAM1112) produced tortilla with superior rollability scores, however, the doughs were very elastic and resulted into production of small diameter tortillas. This is because the flours formed very strong gluten which is not desirable for tortillas (Waniska et. al., 2004).

Overall 72 lines produced suitable tortillas (Table 17). These lines are located on the B quadrant of the plot between rollability and tortilla diameter (Figure 25). Some lines were misclassified because they had tortilla rollabilty and diameters that were close to the minimu suitability scores (~ 3.0 rollability score and ~ 165 mm diameters) (Figure 25).

Table 17

Actual number of lines and proportions based on all raw data including outliers

¹ Location	² Good	³ Poor	Total
TIA	21 47%	24 53%	45
TXE/UVT	15 37%	26 63%	41
TAM1112	36 36%	65 64%	101
Total	72 39%	115 61%	187

¹ Source of lines where TIA (Lines from College Station, McGregor and Chillicothe), TXE/UVT (Lines from advanced generation planted in Amarillo), TAM1112 (Lines from TAM111x112 planted in Etter).

² Good means number of lines that were actually suitable for tortilla based on raw data

CHAPTER IV

CONCLUSIONS AND FUTURE RECOMMENDATIONS

Prediction of the ability of wheat lines for tortilla production involves a combination of various variables that significantly explain the variability in both tortilla diameter and rollability. This study evaluated the multivariate predictive power of kernel, flour and dough properties in simultaneous determination of both tortilla diameter and rollability after 16 days of storage. The sample size was adequate and representative of a normal wheat population of diverse kernel, flour, dough and tortilla properties.

Stress relaxation force after 25 sec. ($r = - 0.73$) and 100 sec. ($r = - 0.71$) are the best univariate predictors of tortilla diameter. This was consistent with canonical correlation where the first canonical coefficient was negatively correlated with tortilla diameter ($r = - 0.73$). This is a measure of how fast the dough structure relaxes under constant strain. Doughs that exhibit low relaxation force at 25 and 100 sec produced tortilla with large diameters this implies that the flours formed a weak gluten structure that easily spread during processing and did not have enough elasticity to shrink back resulting in large diameter tortillas.

These relaxation parameters explained about 14% of the variability in tortilla rollability with an insignificant correlation coefficient ($p < 0.05$) which means that tortilla diameter and rollability cannot be predicted together using only the stress relaxation variables. However, the principal component analysis identifies potentially significant relationship between tortilla rollability and stress relaxation force after 25 sec.

and 100 sec. This implies that rollability is associated with the strength of dough gluten structure and can be predicted using these parameters.

Dough elasticity explained 46% of the variability in tortilla diameter and was a good predictor of objective tortilla textural properties (Modulus of deformation – $r = -0.40$, force – $r = 0.54$, distance $r = 0.50$ and work to rupture $r = 0.60$).

Canonical correlation analysis revealed that 37% of the variability in tortilla rollability is attributed to dough elasticity. Highly elastic doughs make tortillas that have high rollability scores after 16 days of storage. This correlation is also evident on the principal component plot of PC1 and PC2. Rollability can be explained using compression force, glutenin to gliadin ratio, mixograph peak time and dough extensibility.

The best predictors of tortilla processing suitability of for wheat lines are kernel weight measured using the single kernel characterization system (SKCS), dough extensibility work to extend, dough compression force, stress relaxation force after 25 sec and 100 sec. measured using the texture analyzer (TAXT2i), flour glutenin to gliadin ratio and insoluble polymeric protein content evaluated using a high pressure liquid chromatography system (HPLC). Stepwise variable selection method had a lower apparent error rate compared to forward and backward elimination methods. The resulting model classified 83% of the wheat varieties in to the correct tortilla suitability classification. This is robust and feasible for the wheat breeding program. The model is cost effective, because the variables require small sample sizes: 300 kernels for determination of kernel weight, 100 g of flour for dough extensibility, compression and

stress relaxation tests, 200 mg of flour both glutenin to gliadin ratio and insoluble polymeric protein content determination. This is model will significantly decrease the time consuming, labor intensive and expensive traditional full scale processing from 21 days to about 5 days. The breeding program will also experience increased efficiency for screening of early generation lines. The model efficiency can be improved by increasing the number of lines to 250 using additional lines with different genetic background to improve the model efficiency and verify accuracy. These lines possess HMW-GS 2+12 or 5 + Null on the Glu D1 loci and could provide an increased number of superior varieties for tortilla production.

The model can also be improved by evaluating the potential of dough dynamic oscillatory properties. This is evaluated using dynamic oscillatory tests which determine the viscoelastic properties simultaneously and are known to have minimal destruction of the sample structural properties. This can involve three approaches of oscillatory testing; one is the angular frequency sweeps test that provide viscoelastic characteristics of doughs by monitoring both elastic (G') and viscous (G'') moduli. The second one is the stress sweep tests which determine dough elasticity by analyzing the rheological response of dough during and after application of a constant strain. The third is temperature sweep tests which are useful in determining the thermal stability of the dough structure providing information regarding gelatinization of starch and denaturation of protein. The potential of wheat pentosan content as a predictor of tortilla making ability of wheat varieties should also be investigated this is because the water soluble pentosans of wheat could have a significant role in tortilla shelf stability and

rollability. This study offers the first prediction model that can be used to simultaneously predict both tortilla diameter and rollability. It can be used in developing screening tests for rapid analysis of wheat breeding populations for flat bread processing and also provides a useful tool for grain millers and tortilla processors in evaluating the wheat quality for the growing tortilla market.

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APPENDIX A

Table A1
Classification and Cross-validation Summary using 5 Nearest Neighbors

Line	Type	weight	GGRatio	IPP	Area	F_25	CompForce	Diameter2	Rollability	suitability	_INTO_
100	GOOD	21.0	0.50	49.8	24.0	8.1	111.5	168	4.25	1	GOOD
101	POOR	24.9	0.50	49.5	21.1	10.2	94.5	151	3.75	0	POOR
102	GOOD	23.1	0.50	46.4	16.8	7.5	104.6	169	4	1	GOOD
103	POOR	26.2	0.50	54.0	22.4	11.4	72.0	143	4.5	0	POOR
104	POOR	23.3	0.50	48.4	15.5	8.6	70.1	163	2	0	
105	POOR	24.4	0.66	48.2	18.6	8.4	91.1	160	4.25	0	POOR
106	POOR	24.4	0.66	48.2	18.6	8.4	91.1	160	4.25	0	POOR
107	POOR	23.6	.	41.0	30.3	9.5	49.3	152	3.5	0	
108	POOR	23.3	.	33.6	15.7	9.6	57.0	159	3.75	0	
109	POOR	23.6	.	52.7	29.5	10.5	62.1	147	4.25	0	
110	POOR	24.7	.	56.2	27.9	12.4	105.6	152	3.75	0	
111	POOR	21.4	.	44.3	14.3	7.4	113.5	162	2	0	
112	POOR	21.4	.	44.3	14.3	7.4	113.5	162	.	0	
113	POOR	22.9	.	43.9	15.7	8.5	170.3	161	2	0	
114	POOR	22.9	.	43.9	15.7	8.5	170.3	161	.	0	
115	POOR	23.3	.	30.8	23.4	9.5	130.3	155	2	0	
116	POOR	25.7	.	52.7	25.6	10.5	65.0	152	2	0	
117	POOR	22.1	.	53.4	25.2	10.1	97.2	161	2	0	
118	POOR	22.7	0.71	53.0	21.4	8.1	70.3	164	2	0	
119	POOR	23.7	0.66	49.5	18.1	9.6	94.7	163	2	0	
120	POOR	22.9	0.76	54.2	23.9	7.4	74.7	165	2	0	
121	POOR	26.2	.	.	23.4	7.2	72.1	157	2	0	
122	POOR	26.1	0.69	48.4	18.5	8.7	72.5	160	2	0	
123	POOR	26.5	0.71	53.2	29.0	9.7	80.5	160	2	0	
124	POOR	24.7	0.65	55.7	26.0	10.2	64.8	155	2	0	
125	POOR	23.9	0.63	52.4	17.1	8.7	78.9	161	2	0	
126	POOR	22.9	0.60	72.2	16.4	8.7	82.2	163	2	0	
127	POOR	23.6	0.56	12.8	14.8	9.7	76.3	165	2	0	
128	POOR	22.8	0.67	50.6	18.1	8.1	37.7	171	2.75	0	POOR
129	POOR	22.0	0.64	65.0	29.6	9.0	86.5	161	2	0	
130	POOR	24.9	0.51	51.8	18.8	8.8	99.8	158	2	0	
131	POOR	25.2	0.68	52.4	15.2	9.9	84.6	152	2.25	0	POOR
132	POOR	21.2	0.61	51.4	17.0	9.8	112.8	156	3	0	POOR
133	POOR	25.8	0.63	50.1	21.6	10.2	133.1	160	3	0	POOR
134	POOR	24.2	0.54	52.8	16.6	7.7	96.3	163	2.5	0	POOR
135	POOR	23.6	0.53	58.7	15.7	10.4	92.2	156	3.5	0	POOR
136	POOR	26.5	0.61	51.9	15.1	9.4	109.6	153	3.25	0	POOR
137	POOR	25.9	0.63	55.4	27.2	11.4	87.2	147	3	0	POOR
138	POOR	27.5	0.68	55.6	22.3	8.8	105.2	136	4	0	POOR
139	POOR	22.5	0.58	52.3	15.9	11.0	66.6	150	2.75	0	POOR
140	POOR	25.4	0.53	52.9	15.9	8.6	123.8	160	3.25	0	POOR
141	POOR	22.8	0.68	56.4	23.6	10.5	80.5	159	4	0	POOR
142	GOOD	24.3	0.62	51.8	25.0	7.9	95.3	173	3	1	GOOD
143	POOR	25.4	0.70	51.5	26.2	9.4	48.7	163	2.75	0	POOR
144	POOR	25.9	0.63	50.4	12.7	9.5	60.9	158	3.25	0	POOR
145	GOOD	24.5	0.60	55.0	22.8	8.7	64.2	174	3.5	1	GOOD
146	GOOD	24.8	0.60	52.0	26.1	7.8	61.3	170	3.25	1	GOOD
147	GOOD	27.2	0.67	49.9	24.4	9.8	46.6	163	3.5	1	GOOD
148	POOR	23.1	0.74	51.4	13.9	10.5	70.3	160	3.5	0	POOR
149	POOR	23.2	0.69	58.2	19.7	10.1	87.2	162	3.5	0	GOOD
150	POOR	28.7	0.71	54.7	26.6	10.1	83.2	155	3	0	GOOD
151	GOOD	24.7	0.66	54.8	25.2	8.3	36.0	172	2.25	1	GOOD
152	POOR	26.2	0.63	12.2	26.3	8.5	84.9	169	2.75	0	POOR
153	GOOD	25.3	0.58	53.8	20.2	9.8	100.0	165	3	1	POOR
154	GOOD	26.2	0.58	44.2	18.9	8.3	40.3	175	3.25	1	GOOD
155	GOOD	26.7	0.59	48.4	28.1	9.2	35.8	163	3.5	1	GOOD
156	GOOD	26.0	0.60	49.5	24.0	8.6	40.3	166	3.5	1	GOOD
157	POOR	27.1	0.65	46.9	19.1	8.6	46.3	164	3.75	0	GOOD
158	POOR	24.0	0.63	59.1	23.3	9.9	169.6	147	3.5	0	POOR
159	POOR	25.3	0.81	58.7	17.4	9.9	149.3	150	3.5	0	POOR
160	POOR	26.0	0.76	51.6	23.4	7.3	119.3	170	2.75	0	
161	POOR	25.9	0.68	48.8	13.6	7.9	.	165	2.75	0	
162	GOOD	27.0	0.71	52.4	16.0	7.2	.	170	3	1	
163	POOR	23.5	0.73	34.1	25.4	8.1	93.9	161	3.75	0	POOR

Table A1 Continued

Line	Type	weight	GGRatio	IPP	Area	F_25	CompForce	Diameter2	Rollability	suitability	_INTO_
164	GOOD	24.6	0.81	53.8	26.4	7.5	129.1	165	3.75	1	
165	GOOD	24.4	0.75	57.9	27.1	7.7	60.4	166	3	1	
166	GOOD	26.3	0.70	48.8	22.5	7.2	123.1	169	3.25	1	
167	POOR	26.0	0.80	53.5	24.1	10.1	129.8	149	3.5	0	POOR
168	GOOD	27.5	0.75	52.4	22.0	7.3	87.2	181	3	1	GOOD
169	POOR	25.6	0.75	50.1	20.1	9.3	98.6	161	3.5	0	
170	GOOD	26.5	0.68	48.8	16.6	7.4	93.9	165	2.75	1	
171	POOR	24.2	0.70	50.6	16.7	8.5	133.0	161	2.75	0	
172	POOR	23.1	0.65	48.8	16.7	7.1	139.7	170	2.75	0	
173	GOOD	25.3	0.79	54.9	22.9	8.4	117.6	171	3.75	1	GOOD
174	POOR	27.7	0.65	44.3	16.9	7.0	100.4	168	2.5	0	
175	GOOD	26.0	0.67	52.4	18.3	7.2	114.9	174	3.75	1	GOOD
176	POOR	28.9	0.70	40.4	20.0	7.8	93.7	166	2	0	POOR
177	GOOD	27.6	0.66	43.5	15.8	6.8	116.1	174	2.75	1	GOOD
178	POOR	27.8	0.81	42.3	23.4	9.9	63.3	156	3	0	POOR
179	GOOD	24.8	0.76	53.5	24.0	8.0	82.9	171	3.5	1	GOOD
180	GOOD	26.4	0.75	56.2	26.7	7.4	64.4	170	3	1	GOOD
181	GOOD	27.8	0.71	54.8	24.5	7.5	89.8	181	2.75	1	GOOD
182	POOR	28.7	0.71	48.3	14.8	9.4	102.0	150	3.75	0	POOR
183	GOOD	24.6	0.65	51.1	25.4	7.7	58.3	168	3.25	1	GOOD
184	GOOD	27.2	0.67	56.1	26.6	7.8	66.2	168	3.25	1	GOOD
185	GOOD	26.9	0.61	48.9	26.3	7.4	92.8	175	.	1	
186	GOOD	25.0	0.55	49.6	20.7	7.8	70.8	171	.	1	
187	GOOD	25.2	0.58	47.6	15.9	7.0	107.4	175	.	1	
188	GOOD	25.7	0.63	53.2	22.6	7.5	90.4	168	3.75	1	GOOD
189	GOOD	25.2	0.55	43.0	16.8	6.2	124.9	172	3.75	1	GOOD
190	GOOD	25.2	0.70	50.6	14.3	6.9	96.1	177	3.25	1	GOOD
191	GOOD	24.3	0.63	49.4	.	8.0	84.1	171	3.5	1	
192	POOR	24.6	0.74	53.4	20.5	9.2	122.3	155	2.75	0	POOR
193	POOR	24.1	.	.	16.8	8.0	94.9	165	0	0	
194	GOOD	25.8	0.62	50.6	.	11.5	123.4	175	2.75	1	
195	GOOD	25.8	0.63	47.7	.	7.4	139.2	176	2.75	1	
196	GOOD	27.8	0.66	51.5	.	7.2	82.7	173	3.25	1	
197	GOOD	28.3	0.71	22.9	.	6.1	107.5	175	3	1	
198	POOR	27.1	0.68	48.1	.	8.1	62.4	163	2.5	0	
199	POOR	24.1	0.74	52.8	.	9.1	86.0	163	3	0	
200	POOR	25.4	0.71	50.7	.	8.7	75.8	165	2.75	0	
1	POOR	33.4	0.74	41.4	19.6	8.9	83.4	161	4	0	GOOD
2	POOR	30.3	0.79	39.5	16.4	7.0	96.6	169	2.75	0	POOR
3	POOR	32.4	0.78	46.3	16.3	7.8	99.0	163	3.5	0	GOOD
4	POOR	30.1	0.84	47.4	18.5	9.5	95.6	160	2.875	0	POOR
5	GOOD	28.6	0.78	45.4	22.3	7.3	98.5	173	3.5	1	GOOD
6	POOR	27.2	0.76	49.4	23.7	9.4	83.2	151	4.125	0	POOR
7	POOR	33.0	0.88	39.5	15.2	7.2	80.2	170	2.625	0	
8	GOOD	30.1	0.85	39.0	14.9	6.3	81.8	175	2.875	1	
9	POOR	30.4	0.83	48.8	20.2	8.0	125.6	158	3.875	0	
10	POOR	30.5	0.91	42.9	18.7	7.1	103.2	174	2.25	0	POOR
11	GOOD	29.5	0.82	46.9	14.4	6.6	67.7	171	3.125	1	GOOD
12	POOR	29.0	0.86	36.2	17.8	8.7	125.9	154	3.625	0	POOR
13	POOR	34.8	0.77	45.4	17.0	7.4	77.4	161	2.25	0	POOR
14	GOOD	28.4	0.84	47.6	16.9	6.5	79.1	172	3.25	1	GOOD
15	POOR	34.2	0.84	40.0	12.2	7.7	128.1	163	2.375	0	POOR
16	POOR	33.9	0.82	48.0	22.6	8.6	83.9	160	4.25	0	
17	POOR	32.8	0.92	51.1	24.5	7.3	81.3	159	3.375	0	POOR
18	POOR	31.1	0.79	46.3	21.6	7.7	121.1	161	2.875	0	
19	GOOD	32.1	0.78	42.5	16.5	6.9	103.3	167	3.125	1	
20	POOR	30.4	0.81	40.9	20.5	7.0	87.4	168	2.5	0	POOR
21	GOOD	31.4	0.83	46.2	14.4	7.4	66.1	171	3	1	POOR
22	GOOD	29.4	0.78	40.8	17.2	7.2	79.9	171	3.75	1	GOOD
23	GOOD	34.6	0.88	45.9	18.9	7.2	62.4	176	3.25	1	GOOD
24	GOOD	30.5	0.75	39.6	18.3	7.2	64.6	170	3.375	1	GOOD
25	GOOD	31.2	0.82	39.5	13.8	6.3	105.1	170	3	1	
26	POOR	30.3	0.81	40.4	22.7	10.2	97.8	153	3.625	0	POOR
27	GOOD	30.8	0.79	46.4	22.1	7.5	86.4	172	3.75	1	GOOD
28	POOR	32.6	0.78	35.5	15.5	7.5	71.2	170	2.875	0	POOR
29	POOR	31.4	0.67	46.8	19.2	8.6	129.5	156	2.875	0	POOR
30	GOOD	32.2	0.89	41.7	17.3	7.1	85.2	171	3.25	1	GOOD
31	POOR	33.9	0.86	47.4	19.9	10.1	80.9	153	3.25	0	POOR

A1 Continued

Line	Type	weight	GGRatio	IPP	Area	F_25	CompForce	Diameter2	Rollability	suitability	_INTO_
32	POOR	31.4	0.99	51.7	23.8	9.5	80.5	155	3.875	0	POOR
33	POOR	31.0	0.84	43.4	20.9	9.4	115.4	158	3.25	0	POOR
34	POOR	33.4	0.84	44.7	16.9	9.1	143.2	154	3	0	POOR
35	POOR	26.2	0.87	37.4	13.9	6.0	65.3	176	2.75	0	GOOD
38	POOR	26.1	0.83	47.1	23.1	8.6	80.4	163	3.5	0	POOR
39	GOOD	26.5	0.75	41.8	13.7	7.9	86.4	167	3.75	1	GOOD
40	POOR	29.4	0.89	49.2	16.0	9.6	89.6	146	3.875	0	POOR
41	POOR	29.0	0.86	53.6	20.3	8.7	67.1	165	3.25	0	POOR
42	GOOD	30.5	0.84	40.9	14.2	6.8	61.9	171	3.375	1	GOOD
43	GOOD	29.1	0.81	35.8	10.8	5.3	44.4	183	2.125	1	GOOD
44	GOOD	33.5	0.57	45.4	19.6	8.4	80.2	174	4.1	1	GOOD
45	GOOD	24.1	0.48	44.7	20.7	8.5	88.8	169	3.9	1	GOOD
46	POOR	27.2	0.52	49.2	24.0	10.1	107.6	163	5	0	GOOD
47	GOOD	29.2	0.51	44.3	23.1	9.2	90.5	165	5	1	GOOD
48	GOOD	27.7	0.50	37.6	15.2	8.4	112.7	168	4.1	1	GOOD
49	GOOD	27.8	0.52	38.7	16.3	8.1	92.8	169	4.5	1	GOOD
50	GOOD	29.6	0.60	36.4	16.4	7.8	107.9	176	4.4	1	GOOD
51	POOR	27.9	0.56	45.1	31.1	10.5	123.8	159	4.4	0	POOR
52	GOOD	25.2	0.48	35.8	10.8	6.9	73.8	182	3.1	1	GOOD
53	GOOD	29.3	0.52	42.7	17.2	7.8	102.0	173	4.4	1	GOOD
54	GOOD	26.6	0.60	44.9	18.7	8.6	109.5	170	4.3	1	POOR
55	GOOD	29.5	0.52	45.7	23.0	9.3	120.5	166	4.8	1	GOOD
56	GOOD	27.1	0.50	33.5	.	5.8	91.1	184	3	1	
57	GOOD	31.2	0.52	47.7	23.3	9.5	95.9	168	4.5	1	POOR
58	GOOD	25.1	0.59	35.8	14.2	7.0	102.2	179	3.3	1	GOOD
59	POOR	35.5	0.46	43.8	20.8	9.0	134.4	161	3.8	0	POOR
60	POOR	27.9	0.48	44.2	20.2	8.4	92.8	164	3.3	0	POOR
61	POOR	29.4	0.58	47.1	19.8	9.5	104.4	164	3.8	0	POOR
62	POOR	31.1	0.52	40.8	20.7	9.1	106.5	162	3.8	0	POOR
63	POOR	29.4	0.70	29.2	11.8	7.7	101.4	176	1.6	0	POOR
64	POOR	28.9	0.61	35.7	12.9	7.3	92.0	172	2.3	0	POOR
65	POOR	33.0	0.75	36.2	10.1	7.0	98.1	176	2.6	0	GOOD
66	POOR	32.9	0.63	43.1	21.9	9.9	143.8	152	3.5	0	POOR
67	POOR	27.7	0.59	32.4	10.8	6.9	77.2	179	2	0	POOR
68	GOOD	28.8	0.59	35.7	16.0	7.2	101.8	170	4	1	GOOD
69	POOR	29.3	0.58	42.7	17.8	9.7	107.3	159	4.4	0	POOR
70	GOOD	30.3	0.53	43.3	20.0	9.0	117.8	166	4.5	1	POOR
71	GOOD	29.3	0.46	29.8	.	5.7	85.3	180	4	1	
72	GOOD	33.3	0.62	42.7	18.7	9.5	111.7	167	3.3	1	GOOD
73	POOR	28.9	0.71	34.6	13.0	6.7	110.2	178	2.1	0	POOR
74	GOOD	38.1	0.56	48.2	18.4	8.9	95.9	168	3.4	1	POOR
75	GOOD	28.6	0.50	46.2	20.7	9.6	81.6	164	3.3	1	POOR
76	GOOD	28.4	0.55	49.5	19.3	7.6	100.5	165	3.4	1	GOOD
77	POOR	29.0	0.57	46.7	17.7	10.5	98.4	162	3	0	GOOD
78	POOR	28.2	0.55	37.8	.	9.0	110.9	174	2	0	
79	GOOD	29.0	0.49	41.2	14.6	9.5	96.6	171	3.1	1	POOR
80	POOR	30.6	0.56	35.5	14.2	9.5	106.4	180	2.1	0	GOOD
81	POOR	28.7	0.62	48.2	18.9	7.8	145.3	156	3.3	0	POOR
82	POOR	28.2	0.52	34.3	11.8	7.4	80.9	184	1.3	0	POOR
83	POOR	26.4	0.57	42.0	17.8	8.2	104.6	174	2.8	0	GOOD
86	POOR	25.7	0.51	36.8	4.1	5.9	85.6	178	2	0	GOOD
87	POOR	34.1	0.52	46.7	17.2	7.2	127.3	169	2.6	0	GOOD
88	POOR	26.6	0.50	41.3	.	8.9	138.4	173	2.3	0	

Table A2
Kernel properties

Line	Name	Kernel Properties			
		Hardness	Diameter	Weight	protein
1	TAM W-101	72.2	2.7	33.4	14.2
2	TAM 111	77.7	2.6	30.3	13.0
3	TAM 112	81.1	2.7	32.4	13.0
4	TAM 304	89.9	2.6	30.1	12.2
5	TAM 203	86.9	2.5	28.6	14.4
6	TAM 401	74.6	2.4	27.2	14.0
7	TAM 113	78.1	2.7	33.0	12.3
8	TX08A001128	69.4	2.6	30.1	14.2
9	TX08A001249	83.5	2.6	30.4	12.7
10	TX07V7327	79.0	2.6	30.5	12.8
11	TX08V7173	84.0	2.6	29.5	14.1
12	TX08V7313	89.0	2.6	29.0	12.6
13	TX09A001172	78.0	2.7	34.8	12.0
14	TX10A001016	84.6	2.5	28.4	14.2
15	TX06A001132-Resel	74.3	2.7	34.2	12.7
16	TX09A001194	78.8	2.7	33.9	13.1
17	TX09A001197	81.3	2.8	32.8	13.9
18	TX09A001205	80.5	2.6	31.1	13.3
19	TX09A001208	80.1	2.6	32.1	13.1
20	TX09A001235	79.7	2.5	30.4	12.6
21	TX09A001251	78.1	2.7	31.4	12.6
22	TX09A001264	82.8	2.6	29.4	13.9
23	TX09A001343	75.3	2.7	34.6	13.2
24	TX10A001006	84.0	2.7	30.5	14.1
25	TX10A001018	81.7	2.7	31.2	13.3
26	TX08V7140	87.1	2.6	30.3	13.2
27	TX08V7557	79.4	2.6	30.8	12.3
28	TX08V7579	79.5	2.6	32.6	11.9
29	TX08V7675	75.1	2.6	31.4	12.2
30	TX08V7706	76.4	2.7	32.2	12.4
31	TX08V7753	84.8	2.7	33.9	13.0
32	TX07A001418-YRR Resel#2	85.4	2.7	31.4	13.5
33	TX09D1036	73.4	2.6	31.0	12.6
34	TX09D1037	73.5	2.7	33.4	12.0
35	TX09D1119	75.5	2.4	26.2	12.9
38	TX09D1163	85.1	2.5	26.1	12.4
39	TX09D1172	79.0	2.4	26.5	13.0
40	TX09D1193	86.9	2.6	29.4	13.5
41	UVTHP 30	87.1	2.5	29.0	13.0
42	UVTHP 31	80.0	2.6	30.5	12.7
43	UVTHP 32	79.6	2.6	29.1	12.8

Table A2 Continued

Line	Name	Kernel Properties			
		Hardness	Diameter	Weight	protein
100	BD12P001	70.0	2.3	21.0	14.4
101	BD12P002	59.0	2.4	24.9	13.8
102	BD12P003	68.7	2.4	23.1	15.1
103	BD12P004	65.3	2.5	26.2	14.8
104	BD12P005	49.4	2.4	23.3	14.4
105	BD12P007	53.7	2.4	24.4	14.4
106	BD12P007	53.7	2.4	24.4	14.4
107	BD12P008	51.9	2.4	23.6	15.0
108	BD12P009	54.4	2.4	23.3	14.7
109	BD12P010	69.8	2.4	23.6	14.9
110	BD12P011	62.6	2.4	24.7	14.2
111	BD12P012	54.1	2.3	21.4	14.6
112	BD12P012	54.1	2.3	21.4	14.6
113	BD12P013	69.8	2.3	22.9	14.8
114	BD12P013	69.8	2.3	22.9	14.8
115	BD12P014	52.6	2.5	23.3	14.3
116	BD12P015	59.5	2.4	25.7	14.9
117	BD12P016	62.7	2.4	22.1	14.3
118	BD12P017	74.3	2.4	22.7	14.7
119	BD12P018	49.4	2.4	23.7	14.5
120	BD12P019	68.9	2.4	22.9	14.6
121	BD12P020	62.0	2.5	26.2	14.4
122	BD12P021	50.8	2.5	26.1	14.8
123	BD12P022	59.2	2.5	26.5	14.4
124	BD12P023	59.4	2.4	24.7	14.6
125	BD12P024	66.5	2.4	23.9	14.6
126	BD12P025	67.5	2.4	22.9	14.5
127	BD12P026	67.9	2.4	23.6	14.3
128	BD12P027	66.0	2.3	22.8	13.9
129	BD12P028	59.2	2.3	22.0	14.6
130	BD12P029	57.9	2.4	24.9	14.0
131	BD12P030	63.0	2.4	25.2	14.3
132	BD12P031	65.3	2.3	21.2	14.3
133	BD12P032	54.7	2.4	25.8	14.4
134	BD12P033	62.4	2.4	24.2	14.6
135	BD12P034	62.7	2.4	23.6	14.1
136	BD12P035	54.1	2.5	26.5	15.0
137	BD12P036	53.8	2.5	25.9	14.7
138	BD12P037	56.2	2.5	27.5	14.4
139	BD12P038	66.9	2.4	22.5	14.3
140	BD12P039	67.9	2.4	25.4	13.8
141	BD12P053	53.7	2.3	22.8	14.3
142	BD12P054	60.2	2.3	24.3	14.0
143	BD12P055	61.1	2.5	25.4	13.8
144	BD12P057	58.5	2.4	25.9	13.8
145	BD12P058	63.3	2.4	24.5	14.3
146	BD12P059	60.4	2.4	24.8	14.4
147	BD12P060	57.1	2.5	27.2	14.3
148	BD12P061	63.6	2.3	23.1	14.6
149	BD12P062	56.7	2.4	23.2	14.4
150	BD12P063	54.5	2.6	28.7	13.9
151	BD12P064	57.4	2.4	24.7	14.8

Table A2 Continued

Line	Name	Kernel Properties			
		Hardness	Diameter	Weight	protein
152	BD12P065	60.0	2.5	26.2	15.0
153	BD12P066	51.8	2.4	25.3	14.8
154	BD12P067	58.7	2.4	26.2	14.8
155	BD12P070	53.8	2.4	26.7	14.3
156	BD12P071	59.6	2.4	26.0	14.2
157	BD12P072	59.0	2.5	27.1	14.3
158	BD12P078	64.4	2.4	24.0	14.3
159	BD12P079	60.7	2.4	25.3	14.4
160	BD12P080	55.3	2.5	26.0	14.3
161	BD12P081	60.4	2.4	25.9	14.4
162	BD12P082	55.4	2.5	27.0	14.5
163	BD12P083	60.9	2.4	23.5	14.2
164	BD12P084	53.3	2.3	24.6	14.5
165	BD12P085	54.5	2.4	24.4	14.6
166	BD12P086	61.4	2.4	26.3	14.8
167	BD12P087	55.1	2.5	26.0	14.2
168	BD12P088	51.7	2.4	27.5	14.6
169	BD12P089	59.6	2.4	25.6	14.4
170	BD12P090	58.1	2.5	26.5	14.4
171	BD12P091	63.5	2.4	24.2	14.5
172	BD12P092	64.7	2.4	23.1	14.7
173	BD12P093	66.3	2.4	25.3	15.2
174	BD12P094	61.0	2.6	27.7	14.8
175	BD12P095	64.3	2.5	26.0	15.2
176	BD12P096	57.0	2.6	28.9	15.0
177	BD12P097	58.3	2.6	27.6	14.9
178	BD12P098	53.6	2.5	27.8	14.9
179	BD12P099	50.5	2.4	24.8	14.8
180	BD12P_10	65.4	2.5	26.4	15.1
181	BD12P101	57.7	2.5	27.8	14.9
182	BD12P102	57.5	2.6	28.7	15.0
183	BD12P104	63.3	2.4	24.6	14.8
184	BD12P105	65.4	2.4	27.2	15.0
185	BD12P106	55.7	2.6	26.9	15.4
186	BD12P107	53.7	2.4	25.0	14.6
187	BD12P108	51.4	2.4	25.2	14.5
188	BD12P109	64.3	2.4	25.7	14.2
189	BD12P110	62.9	2.4	25.2	14.4
190	BD12P112	69.0	2.4	25.2	14.8
191	BD12P113	61.1	2.4	24.3	14.9
192	BD12P114	63.2	2.4	24.6	14.8
193	BD12P115	57.7	2.4	24.1	14.5
194	BD12P120	51.3	2.4	25.8	14.8
195	BD12P121	58.3	2.4	25.8	15.1
196	BD12P122	59.5	2.6	27.8	14.0
197	BD12P125	51.2	2.5	28.3	14.4
198	BD12P126	61.8	2.5	27.1	14.7
199	BD12P127	61.4	2.4	24.1	14.9
200	BD12P128	55.6	2.4	25.4	15.2

Table A3
Dough rheological properties

Line	Name	Dough Extensibility			Stress Relaxation			Compression
		Elasticity	Extensibility	Work	RT	F_25	F_100	CompForce
1	TAM W-101	0.44	60	20	1.89	8.9	6.1	83
2	TAM 111	0.24	93	16	1.69	7.0	4.3	97
3	TAM 112	0.46	47	16	1.79	7.8	5.1	99
4	TAM 304	0.56	44	19	1.85	9.5	6.9	96
5	TAM 203	0.58	70	22	1.75	7.3	5.0	98
6	TAM 401	0.46	75	24	1.81	9.4	6.6	83
7	TAM 113	0.23	88	15	1.77	7.2	4.5	80
8	TX08A001128	0.19	112	15	1.82	6.3	3.9	82
9	TX08A001249	0.41	66	20	1.70	8.0	5.4	126
10	TX07V7327	0.36	74	19	1.88	7.1	4.5	103
11	TX08V7173	0.24	83	14	1.84	6.6	4.1	68
12	TX08V7313	0.37	65	18	1.68	8.7	5.9	126
13	TX09A001172	0.37	60	17	1.74	7.4	4.8	77
14	TX10A001016	0.24	100	17	1.77	6.5	3.8	79
15	TX06A001132-Resel	0.17	99	12	1.66	7.7	4.9	128
16	TX09A001194	0.52	60	23	1.84	8.6	5.9	84
17	TX09A001197	0.49	69	24	1.63	7.3	4.5	81
18	TX09A001205	0.46	63	22	1.98	7.7	5.2	121
19	TX09A001208	0.25	85	17	1.65	6.9	4.1	103
20	TX09A001235	0.33	86	21	1.80	7.0	4.4	87
21	TX09A001251	0.27	69	14	1.82	7.4	4.9	66
22	TX09A001264	0.36	79	17	1.91	7.2	4.8	80
23	TX09A001343	0.52	49	19	1.67	7.2	4.9	62
24	TX10A001006	0.26	99	18	1.70	7.2	4.4	65
25	TX10A001018	0.17	109	14	1.84	6.3	3.7	105
26	TX08V7140	0.70	44	23	2.04	10.2	7.1	98
27	TX08V7557	0.49	61	22	1.66	7.5	5.0	86
28	TX08V7579	0.25	83	15	1.72	7.5	4.8	71
29	TX08V7675	0.43	60	19	1.75	8.6	5.7	130
30	TX08V7706	0.25	101	17	1.58	7.1	4.5	85
31	TX08V7753	0.53	52	20	1.94	10.1	7.3	81
32	TX07A001418-YRR Resel#2	0.75	43	24	1.81	9.5	6.6	80
33	TX09D1036	0.47	60	21	1.81	9.4	6.6	115
34	TX09D1037	0.41	53	17	1.83	9.1	6.2	143
35	TX09D1119	0.22	79	14	1.64	6.0	3.7	65
38	TX09D1163	0.46	68	23	1.89	8.6	5.9	80
39	TX09D1172	0.27	66	14	1.85	7.9	5.1	86
40	TX09D1193	0.50	42	16	1.73	9.6	6.8	90
41	UVTHP 30	0.76	38	20	1.74	8.7	6.3	67
42	UVTHP 31	0.27	70	14	1.67	6.8	4.3	62
43	UVTHP 32	0.16	91	11	1.52	5.3	3.2	44

Table A3 Continued

Line	Name	Dough Extensibility			Stress Relaxation			Compression
		Elasticity	Extensibility	Work	RT	F_25	F_100	CompForce
100	BD12P001	0.33	105	24	1.79	8.1	5.5	111
101	BD12P002	0.49	59	21	1.73	10.2	6.8	94
102	BD12P003	0.30	75	17	1.65	7.5	4.8	105
103	BD12P004	0.54	56	22	1.89	11.4	7.8	72
104	BD12P005	0.28	77	16	1.81	8.6	5.9	70
105	BD12P007	0.39	62	19	1.64	8.4	5.5	91
106	BD12P007	0.39	62	19	1.64	8.4	5.5	91
107	BD12P008	0.52	84	30	1.81	9.5	6.6	49
108	BD12P009	0.60	36	16	1.67	9.6	6.5	57
109	BD12P010	0.52	80	29	1.91	10.5	7.5	62
110	BD12P011	0.48	86	28	2.28	12.4	8.6	106
111	BD12P012	0.26	66	14	1.59	7.4	4.5	114
112	BD12P012	0.26	66	14	1.59	7.4	4.5	114
113	BD12P013	0.48	43	16	1.55	8.5	5.6	170
114	BD12P013	0.48	43	16	1.55	8.5	5.6	170
115	BD12P014	0.68	48	23	1.72	9.5	6.4	130
116	BD12P015	0.55	66	26	1.85	10.5	7.2	65
117	BD12P016	0.50	69	25	2.00	10.1	7.0	97
118	BD12P017	0.39	78	21	2.12	8.1	5.2	70
119	BD12P018	0.55	44	18	1.73	9.6	6.8	95
120	BD12P019	0.35	101	24	1.75	7.4	4.8	75
121	BD12P020	0.37	92	23	1.63	7.2	4.8	72
122	BD12P021	0.39	62	18	1.88	8.7	5.7	72
123	BD12P022	0.94	43	29	1.79	9.7	6.8	81
124	BD12P023	0.53	67	26	1.85	10.2	7.1	65
125	BD12P024	0.58	41	17	1.74	8.7	5.9	79
126	BD12P025	0.52	42	16	1.74	8.7	5.6	82
127	BD12P026	0.66	31	15	1.91	9.7	6.7	76
128	BD12P027	0.83	31	18	1.40	8.1	5.9	38
129	BD12P028	0.51	89	30	1.79	9.0	5.9	86
130	BD12P029	0.47	54	19	1.62	8.8	6.0	100
131	BD12P030	0.65	33	15	1.65	9.9	6.9	85
132	BD12P031	0.47	47	17	1.48	9.8	6.8	113
133	BD12P032	0.47	64	22	1.63	10.2	7.2	133
134	BD12P033	0.47	48	17	1.48	7.7	5.0	96
135	BD12P034	0.68	32	16	1.71	10.4	7.2	92
136	BD12P035	0.94	25	15	1.56	9.4	6.6	110
137	BD12P036	0.52	72	27	1.71	11.4	7.7	87
138	BD12P037	0.92	35	22	1.44	8.8	6.2	105
139	BD12P038	0.74	30	16	1.94	11.0	7.7	67
140	BD12P039	0.47	46	16	1.74	8.6	5.7	124
141	BD12P053	0.94	36	24	2.35	10.5	7.4	81
142	BD12P054	0.45	80	25	1.61	7.9	5.1	95
143	BD12P055	0.62	57	26	1.85	9.4	6.1	49
144	BD12P057	0.78	24	13	2.00	9.5	6.6	61
145	BD12P058	0.46	69	23	2.37	8.7	5.6	64
146	BD12P059	0.49	76	26	2.23	7.8	5.0	61
147	BD12P060	0.80	43	24	2.33	9.8	6.7	47

Table A3 Continued

Line	Name	Dough Extensibility			Stress Relaxation			Compression
		Elasticity	Extensibility	Work	RT	F_25	F_100	Force
148	BD12P061	0.52	36	14	1.89	10.5	7.1	70
149	BD12P062	0.65	43	20	1.76	10.1	7.0	87
150	BD12P063	0.79	48	27	2.57	10.1	7.0	83
151	BD12P064	0.58	59	25	1.78	8.3	5.8	36
152	BD12P065	0.59	62	26	1.60	8.5	5.8	85
153	BD12P066	0.59	45	20	1.97	9.8	6.8	100
154	BD12P067	0.39	65	19	2.28	8.3	5.3	40
155	BD12P070	0.63	60	28	2.72	9.2	6.1	36
156	BD12P071	0.41	80	24	2.56	8.6	5.6	40
157	BD12P072	0.51	51	19	2.69	8.6	5.5	46
158	BD12P078	1.02	33	23	1.94	9.9	6.8	170
159	BD12P079	1.17	25	17	1.41	9.9	7.0	149
160	BD12P080	0.42	75	23	1.40	7.3	4.9	119
161	BD12P081	0.65	29	14	1.52	7.9	5.3	
162	BD12P082	0.50	43	16	1.41	7.2	5.0	
163	BD12P083	0.70	50	25	1.46	8.1	5.5	94
164	BD12P084	0.48	77	26	1.42	7.5	4.9	129
165	BD12P085	0.68	54	27	2.16	7.7	5.5	60
166	BD12P086	0.47	65	22	1.72	7.2	4.7	123
167	BD12P087	0.84	40	24	1.50	10.1	7.1	130
168	BD12P088	0.52	56	22	1.36	7.3	5.0	87
169	BD12P089	0.61	46	20	1.50	9.3	6.3	99
170	BD12P090	0.35	65	17	1.35	7.4	5.0	94
171	BD12P091	0.49	47	17	1.40	8.5	5.9	133
172	BD12P092	0.37	58	17	1.33	7.1	4.6	140
173	BD12P093	0.94	36	23	1.53	8.4	5.8	118
174	BD12P094	0.38	58	17	1.47	7.0	4.3	100
175	BD12P095	0.46	55	18	1.37	7.2	4.7	115
176	BD12P096	0.39	67	20	1.66	7.8	5.1	94
177	BD12P097	0.31	68	16	1.42	6.8	4.4	116
178	BD12P098	0.72	45	23	1.95	9.9	6.7	63
179	BD12P099	0.39	83	24	1.55	8.0	5.3	83
180	BD12P_10	0.49	77	27	1.68	7.4	5.1	64
181	BD12P101	0.45	77	25	1.65	7.5	5.1	90
182	BD12P102	0.86	25	15	1.73	9.4	6.2	102
183	BD12P104	0.48	74	25	1.59	7.7	5.4	58
184	BD12P105	0.74	49	27	1.60	7.8	5.5	66
185	BD12P106	0.50	71	26	1.55	7.4	4.9	93
186	BD12P107	0.58	49	21	1.47	7.8	5.2	71
187	BD12P108	0.60	36	16	1.51	7.0	4.5	107
188	BD12P109	0.45	66	23	1.45	7.5	4.8	90
189	BD12P110	0.32	67	17	1.34	6.2	3.8	125
190	BD12P112	0.67	30	14	1.55	6.9	4.7	96
191	BD12P113	.	.	.	1.64	8.0	5.3	84
192	BD12P114	0.89	34	21	1.62	9.2	6.4	122
193	BD12P115	0.48	45	17	1.62	8.0	5.3	95
194	BD12P120	.	.	.	1.67	11.5	8.2	123
195	BD12P121	.	.	.	1.40	7.4	5.1	139
196	BD12P122	.	.	.	1.48	7.2	4.9	83
197	BD12P125	.	.	.	1.40	6.1	3.9	108
198	BD12P126	.	.	.	1.84	8.1	5.3	62
199	BD12P127	.	.	.	1.82	9.1	6.3	86
200	BD12P128	.	.	.	2.01	8.7	6.0	76

Table A4
Physical properties of tortillas

<i>Entry</i>	<i>NAME</i>	<i>Moisture</i>	<i>Weight</i>	<i>Thickness</i>	<i>Diameter</i>	<i>Sp. Volume</i>	<i>Lightness</i>
		%	g	mm	mm	cm ³ /g	L-value
1	TAM W-101	33.1	41.4	2.4	161	1.2	81.8
2	TAM 111	35.5	39.8	1.5	169	0.84	82.9
3	TAM 112	31.9	40.9	2.8	163	1.43	82.6
4	TAM 304	34.7	40.9	3.0	160	1.47	83.0
5	TAM 203	39.1	39.9	3.0	173	1.76	81.7
6	TAM 401	34.8	42.1	2.8	151	1.17	81.4
7	TAM 113	33.6	39.8	3.1	170	1.75	82.6
8	TX08A001128	33.3	40.6	3.2	175	1.87	83.7
9	TX08A001249	32.6	41.4	2.8	158	1.35	82.0
10	TX07V7327	34.7	41.6	3.0	174	1.72	83.5
11	TX08V7173	37.8	41.1	2.8	171	1.55	82.6
12	TX08V7313	35.0	41.8	2.7	154	1.22	81.9
13	TX09A001172	33.5	38.9	3.0	161	1.59	72.9
14	TX10A001016	32.9	41.3	3.0	172	1.71	82.8
15	TX06A001132-Resel	33.1	39.4	2.9	163	1.52	83.3
16	TX09A001194	32.6	39.1	2.9	160	1.49	78.3
17	TX09A001197	35.1	40.4	2.8	159	1.37	81.6
18	TX09A001205	34.8	41.5	2.9	161	1.43	81.0
19	TX09A001208	33.2	40.9	3.1	167	1.66	83.6
20	TX09A001235	34.0	41.9	2.9	168	1.53	84.2
21	TX09A001251	33.3	40.5	3.0	171	1.71	83.9
22	TX09A001264	33.5	40.7	3.0	171	1.69	82.1
23	TX09A001343	31.9	40.5	2.5	176	1.53	83.7
24	TX10A001006	33.9	39.9	2.7	170	1.53	82.5
25	TX10A001018	35.2	40.7	2.6	170	1.43	83.2
26	TX08V7140	33.8	40.8	2.9	153	1.29	80.8
27	TX08V7557	35.1	39.6	2.8	172	1.63	83.7
28	TX08V7579	33.9	41.4	2.6	170	1.42	83.0
29	TX08V7675	33.3	41.9	2.9	156	1.34	81.4
30	TX08V7706	33.6	40.7	2.9	171	1.64	83.7
31	TX08V7753	33.4	41.3	2.7	153	1.22	82.2
32	TX07A001418-YRR Resel#2	35.6	41.6	3.0	155	1.36	80.8
33	TX09D1036	34.9	43.1	3.0	158	1.38	82.6
34	TX09D1037	34.6	39.5	3.0	154	1.44	83.0
35	TX09D1119	33.6	40.1	3.0	176	1.79	83.2
38	TX09D1127	33.4	40.9	2.8	163	1.45	82.5
39	TX09D1172	33.9	39.9	2.8	167	1.51	80.6
40	TX09D1193	33.5	39.6	2.7	146	1.15	81.9
41	TX06A001263	25.8	40.3	2.9	165	1.55	83.4
42	TX07A001505	34.6	40.9	2.9	171	1.65	82.0
43	TX03A0563-07AZHR247	37.7	40.2	2.8	183	1.82	83.2

Table A4 Continued

Line	Name	Moisture	Weight	Thickness	Tortilla Diameter	Specific Volume	Tortilla Lightness	Day 16 Flexibility
		%	g	mm	(mm)	(cm ³ /g)	L- value	Rating
100	TAM 111	33.3	45.5	2.74	168.1	1.3	78.0	4.25
101	TAM 112	33.7	40.8	2.48	151.3	1.1	79.6	3.75
102	TX1112-1	34.2	43.9	2.76	169	1.4	81.3	4.00
103	TX1112-2	35.5	42.1	3.76	143.1	1.4	81.1	4.50
104	TX1112-3	33.9	41.2	3.34	162.6	1.7	82.4	2.00
106	TX1112-5	34.4	42.1	2.60	159.8	1.2	80.9	4.25
107	TX1112-6	35.2	40.2	3.35	152.4	1.5	80.8	3.50
108	TX1112-7	34.6	40.1	3.03	158.8	1.5	80.7	3.75
109	TX1112-8	34.2	39.0	3.32	146.7	1.4	82.4	4.25
110	TX1112-9	37.0	42.6	3.00	152.3	1.3	80.9	3.75
112	TX1112-10	31.9	38.7	3.66	161.9	1.9	78.0	.
113	TX1112-11	41.0	46.5	2.98	160.8	1.3	72.6	2.00
128	TX1112-25	31.8	41.8	3.17	170.7	1.7	82.7	2.75
129	TX1112-26	34.4	39.0	3.11	161.1	1.6	84.3	2.00
130	TX1112-27	34.1	42.4	2.94	158	1.4	81.4	2.00
131	TX1112-28	34.0	38.8	2.84	152.2	1.3	81.1	2.25
132	TX1112-29	33.9	44.6	3.13	156.3	1.3	81.5	3.00
133	TX1112-30	32.4	42.2	2.93	159.5	1.4	80.9	3.00
134	TX1112-31	34.7	40.9	3.13	162.7	1.6	82.7	2.50
135	TX1112-32	34.7	38.0	2.90	155.5	1.4	82.1	3.50
136	TX1112-33	34.9	42.2	2.93	153.1	1.3	82.6	3.25
137	TX1112-34	34.7	41.0	3.41	147.4	1.4	81.9	3.00
138	TX1112-35	34.8	40.6	3.36	136.1	1.2	80.7	4.00
139	TX1112-36	36.7	39.4	2.96	149.8	1.3	80.5	2.75
140	TX1112-37	35.6	41.2	2.73	160.1	1.3	82.3	3.25
141	TX1112-49	34.0	45.2	2.92	158.5	1.3	83.9	4.00
142	TX1112-50	34.2	43.1	3.02	172.8	1.6	83.4	3.00
143	TX1112-51	34.7	39.9	3.07	163.2	1.6	84.7	2.75
144	TX1112-53	33.8	37.3	2.83	158.4	1.5	83.0	3.25
145	TX1112-54	34.1	42.1	3.15	173.6	1.8	83.2	3.50
146	TX1112-55	35.0	41.1	2.85	169.8	1.6	84.2	3.25
147	TX1112-56	35.0	41.3	2.71	163.1	1.4	81.9	3.50
148	TX1112-57	33.2	40.5	2.77	160.3	1.4	82.5	3.50
149	TX1112-58	33.7	42.6	2.86	162.2	1.4	80.6	3.50
150	TX1112-59	32.8	39.0	2.84	154.5	1.4	83.5	3.00

Table A4 Continued

Line	Name	Moisture	Weight	Thickness	Tortilla Diameter	Specific Volume	Tortilla Lightness	Day 16 Flexibility
		%	g	mm	(mm)	(cm ³ /g)	L- value	Rating
151	TX1112-60	32.9	42.7	2.93	172.4	1.6	82.9	2.25
152	TX1112-61	31.1	40.2	2.86	169.3	1.6	83.7	2.75
153	TX1112-62	34.8	42.9	2.74	165.1	1.4	83.1	3.00
154	TX1112-63	33.8	44.2	3.20	175.4	1.7	83.5	3.25
155	TX1112-66	32.6	39.6	3.18	163	1.7	82.6	3.50
156	TX1112-67	34.1	41.3	3.24	166.1	1.7	82.1	3.50
157	TX1112-68	33.6	41.5	3.22	163.6	1.6	83.9	3.75
158	TX1112-74	33.8	39.2	2.97	147.2	1.3	81.0	3.50
159	TX1112-75	32.5	40.0	3.02	150.4	1.3	82.0	3.50
160	TX1112-76	33.8	42.5	3.38	169.7	1.8	81.7	2.75
161	TX1112-77	33.3	39.7	3.24	164.8	1.7	81.2	2.75
162	TX1112-78	32.7	42.9	3.46	169.9	1.8	81.7	3.00
163	TX1112-79	32.3	39.1	2.94	161.3	1.5	82.6	3.75
164	TX1112-80	33.5	41.7	3.23	164.7	1.6	80.6	3.75
165	TX1112-81	33.3	37.6	3.24	165.9	1.9	82.0	3.00
166	TX1112-82	38.5	42.4	3.31	169.1	1.8	81.2	3.25
167	TX1112-83	32.6	40.3	3.26	149.4	1.4	76.1	3.50
168	TX1112-84	33.4	41.3	2.94	180.7	1.8	80.8	3.00
169	TX1112-85	33.6	40.8	3.09	160.5	1.5	78.9	3.50
170	TX1112-86	33.3	42.6	3.45	164.8	1.7	80.3	2.75
171	TX1112-87	34.0	40.0	3.35	161.2	1.7	81.1	2.75
172	TX1112-88	34.4	40.0	3.25	170.1	1.8	80.4	2.75
173	TX1112-89	33.5	44.3	2.89	170.9	1.5	83.1	3.75
174	TX1112-90	33.3	38.9	3.16	167.6	1.8	83.6	2.50
175	TX1112-91	32.9	42.9	2.94	173.6	1.6	82.1	3.75
176	TX1112-92	34.7	40.3	3.01	165.7	1.6	84.4	2.00
177	TX1112-93	34.4	41.6	3.23	174.2	1.9	84.5	2.75
178	TX1112-94	34.0	40.9	2.86	155.8	1.3	83.1	3.00
179	TX1112-95	32.4	40.8	3.30	171.1	1.9	84.2	3.50
180	TX1112-96	29.3	38.1	2.73	170.1	1.6	83.2	3.00
181	TAM 111	32.2	41.2	2.82	181.1	1.8	84.4	2.75
182	TAM 112	33.0	40.9	2.93	149.8	1.3	81.1	3.75
183	TX1112-98	34.5	39.4	2.93	167.9	1.6	81.7	3.25
184	TX1112-99	33.1	39.3	2.84	168	1.6	83.8	3.25
188	TX1112-103	33.6	39.8	2.35	168.4	1.3	83.4	3.75
189	TX1112-104	33.2	42.5	2.61	171.7	1.4	83.8	3.75
190	TX1112-106	33.8	40.8	2.61	176.9	1.6	84.6	3.25
191	TX1112-107	19.7	39.2	2.68	170.7	1.6	81.9	3.50
192	TX1112-108	45.9	41.5	2.97	155	1.3	84.9	2.75
193	TX1112-109	32.9	41.7	2.97	164.6	1.5	83.1	0.00
194	TX1112-114	34.1	39.1	2.44	175.3	1.5	82.4	2.75
195	TX1112-115	34.5	42.2	2.60	175.9	1.5	83.5	2.75
196	TX1112-116	32.0	40.4	2.47	173.1	1.4	82.9	3.25
197	TX1112-119	32.1	40.7	2.78	174.8	1.6	81.9	3.00
198	TX1112-120	34.0	41.2	3.11	163	1.6	81.9	2.50
199	TX1112-121	34.2	40.9	2.60	162.9	1.3	82.4	3.00
200	TX1112-122	33.1	41.0	3.00	164.6	1.6	82.4	2.75

Table A5
Texture properties of tortillas after sixteen days of storage

Line	Name	Tortilla Texture Profile after 16 days			
		Modulus	Force	Distance	Work
		(N/mm)	(N)	(mm)	(N.mm)
1	TAM W-101	0.95	10.8	18.4	73
2	TAM 111	0.70	6.6	15.7	37
3	TAM 112	0.88	8.3	16.0	48
4	TAM 304	0.81	7.7	16.7	46
5	TAM 203	0.72	6.6	16.3	39
6	TAM 401	0.89	10.1	18.0	70
7	TAM 113	0.79	6.4	14.8	33
8	TX08A001128	0.69	6.8	16.6	43
9	TX08A001249	0.89	8.5	16.9	54
10	TX07V7327	0.76	5.6	14.1	28
11	TX08V7173	0.83	7.4	16.4	42
12	TX08V7313	1.08	10.6	18.0	66
13	TX09A001172	0.88	7.0	15.0	35
14	TX10A001016	0.84	7.2	16.2	41
15	TX06A001132-Resel	0.78	8.3	16.5	52
16	TX09A001194	0.95	9.1	17.0	56
17	TX09A001197	0.87	9.7	18.5	66
18	TX09A001205	0.95	9.1	16.7	55
19	TX09A001208	0.68	7.4	16.7	44
20	TX09A001235	0.81	7.6	16.4	45
21	TX09A001251	0.78	6.3	14.2	32
22	TX09A001264	0.81	7.3	15.9	40
23	TX09A001343	0.74	6.8	16.7	43
24	TX10A001006	0.96	9.6	18.4	60
25	TX10A001018	0.75	6.2	15.5	34
26	TX08V7140	1.14	12.1	18.8	84
27	TX08V7557	0.76	6.7	15.7	37
28	TX08V7579	0.96	9.4	16.6	57
29	TX08V7675	1.08	8.8	14.6	45
30	TX08V7706	0.88	8.7	17.2	55
31	TX08V7753	1.01	10.7	17.1	69
32	TX07A001418-YRR Resel#2	0.92	9.4	17.3	59
33	TX09D1036	1.08	9.3	15.4	52
34	TX09D1037	1.09	17.5	8.3	59
35	TX09D1119	0.74	5.9	14.8	30
38	TX09D1163	1.02	10.5	17.5	65
39	TX09D1172	0.93	9.5	18.3	59
40	TX09D1193	1.08	11.9	19.6	88
41	UVTHP 30	0.80	7.5	16.5	48
42	UVTHP 31	1.09	9.7	17.5	63
43	UVTHP 32	0.65	5.3	15.2	27

Table A5 Continued

Line	Name	Tortilla Texture Profile after 16 days			
		Modulus	Force	Distance	Work
		(N/mm)	(N)	(mm)	(N.mm)
100	TAM 111	0.80	8.8	19.2	66
101	TAM 112	1.02	14.6	23.6	140
102	TX1112-1	0.68	8.0	17.7	54
103	TX1112-2	0.84	11.9	21.1	99
106	TX1112-5	0.89	10.5	17.2	72
107	TX1112-6	0.98	12.4	20.2	101
108	TX1112-7	0.96	10.2	17.1	63
109	TX1112-8	0.75	9.1	21.2	83
110	TX1112-9	0.99	13.1	22.0	131
112	TX1112-10	0.93	9.9	18.2	64
113	TX1112-11	1.19	13.9	18.8	98
128	TX1112-25	0.92	7.2	14.1	40
131	TX1112-28	1.07	11.8	17.7	76
132	TX1112-29	1.01	11.7	18.3	85
133	TX1112-30	1.43	14.4	16.9	93
134	TX1112-31	0.88	10.2	19.1	70
135	TX1112-32	1.03	10.8	18.0	70
136	TX1112-33	1.05	14.0	18.8	109
137	TX1112-34	1.08	14.5	21.3	132
138	TX1112-35	0.92	16.9	24.5	203
139	TX1112-36	1.07	13.9	22.4	124
140	TX1112-37	0.88	9.6	18.3	65
141	TX1112-49	0.75	8.2	22.0	73
142	TX1112-50	0.84	10.4	20.3	91
143	TX1112-51	0.83	11.0	20.8	91
144	TX1112-53	1.19	14.8	20.9	121
145	TX1112-54	0.86	9.6	19.7	72
146	TX1112-55	0.90	9.3	17.7	60
147	TX1112-56	0.89	11.1	21.2	94
148	TX1112-57	0.93	9.9	18.4	70
149	TX1112-58	1.00	12.7	21.3	111
150	TX1112-59	1.14	13.8	20.9	114

Table A5 Continued

Line	Name	Tortilla Texture Profile after 16 days			
		Modulus	Force	Distance	Work
		(N/mm)	(N)	(mm)	(N.mm)
151	TX1112-60	0.68	8.4	19.3	65
152	TX1112-61	0.72	6.6	17.7	43
153	TX1112-62	1.03	11.7	18.9	86
154	TX1112-63	0.62	5.8	14.2	32
155	TX1112-66	0.81	10.1	19.5	79
156	TX1112-67	0.83	8.7	15.3	52
157	TX1112-68	0.67	8.9	17.8	61
158	TX1112-74	1.15	14.3	18.4	97
159	TX1112-75	1.38	17.5	21.5	143
162	TX1112-78	0.85	7.4	14.1	43
163	TX1112-79	0.90	10.4	19.9	84
167	TX1112-83	1.05	15.0	22.7	153
168	TX1112-84	0.75	8.1	17.5	60
173	TX1112-89	0.68	8.1	19.5	71
175	TX1112-91	0.80	7.5	17.1	52
176	TX1112-92	0.76	8.0	17.5	57
177	TX1112-93	0.51	5.6	17.1	35
178	TX1112-94	0.98	14.6	24.7	153
179	TX1112-95	0.77	9.6	20.0	73
180	TX1112-96	0.81	10.2	20.7	77
181	TAM 111	0.51	4.5	16.2	28
182	TAM 112	0.96	11.2	19.9	84
183	TX1112-98	1.00	11.4	19.9	92
184	TX1112-99	0.90	10.2	19.1	83
188	TX1112-103	0.98	9.5	17.0	57
189	TX1112-104	0.90	10.4	19.3	65
190	TX1112-106	0.97	7.8	15.0	39
191	TX1112-107	0.99	10.0	16.7	60
192	TX1112-108	1.24	15.5	22.3	140
194	TX1112-114	0.98	8.2	15.3	46
195	TX1112-115	0.76	6.9	14.8	38
196	TX1112-116	0.92	10.9	19.3	81
197	TX1112-119	0.84	9.6	19.4	68
198	TX1112-120	1.00	12.2	20.8	94
199	TX1112-121	0.75	8.9	18.1	61
200	TX1112-122	0.86	12.2	21.7	111

Table A6
Flour protein fraction results for TXE/UVT and TAM1112 lines

Line	Name	Protein Quality			
		Mixo_time	GGRatio	H_L_GS_Ratio	IPP
1	TAM W-101	3.00	0.74	0.37	41.35
2	TAM 111	2.50	0.79	0.30	39.54
3	TAM 112	3.50	0.78	0.29	46.34
4	TAM 304	3.50	0.84	0.35	47.44
5	TAM 203	3.38	0.78	0.31	45.42
6	TAM 401	3.00	0.76	0.40	49.38
7	TAM 113	3.00	0.88	.	39.53
8	TX08A001128	2.00	0.85	.	38.98
9	TX08A001249	3.50	0.83	.	48.77
10	TX07V7327	3.38	0.91	0.31	42.94
11	TX08V7173	2.63	0.82	0.33	46.89
12	TX08V7313	3.00	0.86	0.31	36.20
13	TX09A001172	2.63	0.77	0.32	45.45
14	TX10A001016	2.00	0.84	0.33	47.55
15	TX06A001132-Resel	1.63	0.84	0.33	40.00
16	TX09A001194	3.50	0.82	.	47.98
17	TX09A001197	3.63	0.92	0.32	51.08
18	TX09A001205	3.25	0.79	.	46.29
19	TX09A001208	2.00	0.78	.	42.53
20	TX09A001235	2.50	0.81	0.31	40.94
21	TX09A001251	3.13	0.83	0.34	46.20
22	TX09A001264	2.88	0.78	0.36	40.77
23	TX09A001343	3.38	0.88	0.32	45.93
24	TX10A001006	1.75	0.75	0.33	39.57
25	TX10A001018	1.50	0.82	.	39.48
26	TX08V7140	4.00	0.81	0.34	40.37
27	TX08V7557	3.50	0.79	0.35	46.38
28	TX08V7579	2.00	0.78	0.33	35.49
29	TX08V7675	2.75	0.67	0.39	46.79
30	TX08V7706	2.50	0.89	0.38	41.74
31	TX08V7753	3.88	0.86	0.31	47.41
32	TX07A001418-YRR Resel#2	5.25	0.99	0.34	51.73
33	TX09D1036	4.38	0.84	0.37	43.37
34	TX09D1037	4.25	0.84	0.32	44.65
35	TX09D1119	2.38	0.87	0.32	37.39
38	TX09D1163	4.88	0.83	0.31	47.08
39	TX09D1172	3.50	0.75	0.31	41.81
40	TX09D1193	6.50	0.89	0.31	49.20
41	UVTHP 30	3.00	0.86	0.31	53.64
42	UVTHP 31	5.50	0.84	0.32	40.91
43	UVTHP 32	2.25	0.81	0.31	35.79

Table A6 Continued

Line	Name	Protein Quality			
		Mixo_time	GGRatio	H_L_GS_Ratio	IPP
100	BD12P001	5.30	0.50	0.29	49.8
101	BD12P002	6.40	0.50	0.32	49.5
102	BD12P003	3.20	0.50	0.33	46.4
103	BD12P004	4.00	0.50	0.30	54.0
104	BD12P005	3.45	0.50	0.30	48.4
105	BD12P007	4.40	0.66	0.32	48.2
106	BD12P007	4.40	0.66	0.32	48.2
107	BD12P008	3.00	.	0.29	41.0
108	BD12P009	5.15	.	0.29	33.6
109	BD12P010	3.50	.	0.27	52.7
110	BD12P011	4.15	.	0.27	56.2
111	BD12P012	3.30	.	0.30	44.3
112	BD12P012	3.30	.	0.30	44.3
113	BD12P013	4.30	.	0.25	43.9
114	BD12P013	4.30	.	0.25	43.9
115	BD12P014	4.00	.	0.26	30.8
116	BD12P015	3.45	.	0.27	52.7
117	BD12P016	3.40	.	0.28	53.4
118	BD12P017	4.45	0.71	0.28	53.0
119	BD12P018	3.20	0.66	0.29	49.5
120	BD12P019	3.15	0.76	0.28	54.2
121	BD12P020	4.45	.	0.25	.
122	BD12P021	3.30	0.69	0.00	48.4
123	BD12P022	5.00	0.71	0.29	53.2
124	BD12P023	5.30	0.65	.	55.7
125	BD12P024	3.10	0.63	.	52.4
126	BD12P025	5.00	0.60	0.32	72.2
127	BD12P026	3.15	0.56	0.31	12.8
128	BD12P027	4.45	0.67	0.28	50.6
129	BD12P028	2.30	0.64	0.31	65.0
130	BD12P029	3.40	0.51	0.33	51.8
131	BD12P030	3.20	0.68	0.32	52.4
132	BD12P031	4.00	0.61	0.32	51.4
133	BD12P032	4.15	0.63	0.31	50.1
134	BD12P033	5.40	0.54	0.31	52.8
135	BD12P034	4.15	0.53	0.31	58.7

Table A6 Continued

Line	Name	Protein Quality			
		Mixo_time	GGRatio	H_L_GS_Ratio	IPP
136	BD12P035	3.50	0.61	0.32	51.9
137	BD12P036	5.15	0.63	0.30	55.4
138	BD12P037	5.00	0.68	0.31	55.6
139	BD12P038	7.10	0.58	0.34	52.3
140	BD12P039	7.45	0.53	0.32	52.9
141	BD12P053	4.10	0.68	0.33	56.4
142	BD12P054	3.20	0.62	0.30	51.8
143	BD12P055	4.00	0.70	0.31	51.5
144	BD12P057	3.00	0.63	0.33	50.4
145	BD12P058	4.00	0.60	0.30	55.0
146	BD12P059	3.15	0.60	0.28	52.0
147	BD12P060	3.20	0.67	0.32	49.9
148	BD12P061	3.50	0.74	0.34	51.4
149	BD12P062	2.20	0.69	0.30	58.2
150	BD12P063	3.30	0.71	0.31	54.7