

EFFECTS OF BARLEY FLOUR AND β -GLUCANS IN CORN TORTILLAS

A Thesis

by

LAURA SILVA

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

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August 2003

Major Subject: Food Science and Technology

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ABSTRACT

Effects of Barley Flour and β -glucans in Corn Tortillas. (August 2003)

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The effects of β -glucan on corn tortilla texture were evaluated. Barley flour (9.7% β -glucan) was substituted at 2.5, 5 and 10% for dry masa flour in corn tortillas. Texture was evaluated after 4 hr and up to 7 d storage at 4°C. Substitution of 2.5-10% barley flour significantly improved tortilla texture.

Combined effects of barley flour (0-2.5%), maltogenic amylase (0-1650MAU) and carboxymethylcellulose (0-0.5%) were evaluated using surface response methodology. Barley flour increased rollability, pliability, energy dissipated and reduced rupture force and final stiffness. Overall, maltogenic amylase decreased rupture force and Young's modulus but decreased rupture distance, rollability and pliability at levels above 825 MAU. CMC improved rollability, pliability, and rupture distance. The best response was found using barley flour and CMC with 825 MAU, where rollability, pliability, rupture distance and energy dissipated increased while rupture force, Young's modulus and final stiffness decreased.

A 70% barley β -glucan concentrate combined with amylase (550 MAU) or CMC (0-0.5%) was evaluated in corn tortillas. Amylase combined with β -glucan did not improve texture. Tortillas with β -glucan and CMC had significantly improved pliability, rollability, final stiffness and energy dissipated.

Texture measurements analysis showed that depending on the stage of storage, objective and subjective methods correlate differently. Subjective and objective measurements of texture were not correlated at 4 hr storage. At the end of storage,

pliability had significant correlations with stress relaxation measurements, but rollability had higher correlation coefficients with extensibility measurements. Pliability had higher R^2 and lower coefficients of variation compared to rollability.

Sensory evaluation was conducted using reheated 14-day-old tortillas of control, 825 MAU with 0.25% CMC, 0.12% β -glucans, 0.18% β -glucan with 0.375% CMC, and 0.24% β -glucan with 0.25% CMC. All tortillas had similar appearance, flexibility, gumminess, flavor and overall quality. Softness and chewiness of treatments with 0.12% β -glucan or 0.24% β -glucan with 0.25% CMC were similar to control. Other tortillas were significantly tougher and chewier.

β -glucan may be the active ingredient in barley flour that modifies firming of corn tortillas during storage. Barley flour is inexpensive and effectively improves texture of corn tortillas.

DEDICATION

This thesis is dedicated to my family: Gorda, Chuy, la Negra Pelú and my dad for all the support, encouragement, and for making me the person I am now.

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First of all, I thank God for all the opportunities I have received throughout my life, this education being one of them, and for all the people I have met on the way and who have contributed not only in my professional career but in my personal development.

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Y wachuwo...

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CHAPTER I

INTRODUCTION

The tortilla is the fastest growing segment of the U.S. baking industry. In 2000, U.S. sales at wholesale prices totaled more than \$4 billion, representing a growth rate of 57% over the past four years (TIA 2003). Shelf stability is limited by texture deterioration, caused by hardening of the tortillas.

Consumers prefer soft and flexible tortillas. However, during tortilla storage there are changes that cause the firming of these products. Corn tortilla textural changes were mainly related to modifications in the starch properties (Miranda-Lopez 1999). In corn tortillas, staling is a rapid hardening of the product caused by a phenomenon called retrogradation. Retrogradation of gelatinized starch involves the recrystallization of both amylopectin and amylose (Quintero-Fuentes 1999). As a result, during storage of baked products they become firmer, less elastic and break upon bending (Quintero-Fuentes 1999).

The use of additives to overcome the loss of freshness in corn tortillas has been reported. Hydrocolloids, such as carboxymethylcellulose (CMC), and other additives like wheat gluten, soy products and amylases have shown improvement in the texture of tortillas over storage (Yau et al. 1994, Serna-Saldivar 1996, Suhendro 1997, Quintero-Fuentes 1999, Bueso-Ucles 2003). The use of barley flour in corn tortillas was reported by Mitre-Dieste (2001), where substitution of 15% yielded tortillas with better extensibility and rollability scores compared to regular corn tortillas, while maintaining many of the sensory characteristics of corn tortillas.

The present study aims to find an optimum formulation using carboxymethylcellulose, maltogenic amylase, barley flour and concentrated β -glucans to improve corn tortilla texture during storage.

The objectives of this study were:

1. Determine the effects of low amounts of barley flour substitution for dry masa flour on corn tortilla texture.
2. Determine the optimum combinations of barley flour, CMC and amylase to reduce staling of corn tortillas.
3. Determine the effect of concentrated β -glucans combined with amylase or CMC on corn tortilla texture quality.

CHAPTER II

LITERATURE REVIEW

Corn and Corn Products

Maize or corn (*Zea mays* L.) is the largest cereal grain crop produced in the world and the leading cereal crop in the United States. The distribution of crop production in the world and in the United States is shown in Fig.1.

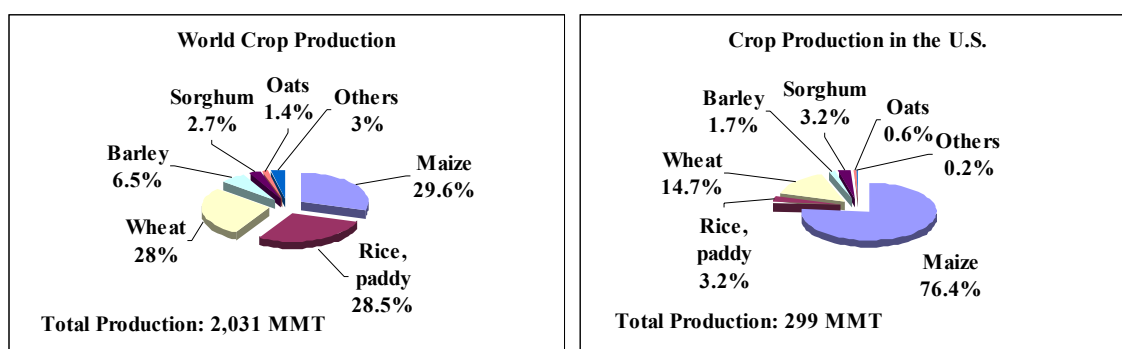


Fig. 1. Crop Production in the World and in the United States (FAO, 2002).

Corn is used in several food products such as tortilla, corn bread, porridges: atole, ogi, kenkey, ugali, ugi, maizena; steamed products: tamales, couscous, dumplings; alcoholic beverages: koda, chichi, Kaffir beer, maize beer; snacks: empanada, chips, tostadas, popped corn. The increase in popularity of Mexican foods throughout the United States has increased the utilization of corn in snack foods and tortillas (Campus-Baypoli et al. 1999). Latin culture has been an increasing influence in America, which is even evident in the food industry. Last year, Americans consumed a total of 7 billion pounds of tortillas, which is the equivalent of 85 billion tortillas (not including tortilla chips) or almost one tortilla per American each day (TIA 2003).

Nixtamalization

The traditional method to process maize into tortillas (nixtamalization) was developed by Latin American Indians and is still practiced in some rural areas of Mexico and Central America. Traditional tortilla preparation entails cooking corn in pots over a fire, stone-grinding the nixtamal by hand and baking hand-molded tortillas on a clay griddle (comal) (Pflugfelder 1986).

Maize is cooked in boiling lime solution for a relatively short time (5-50 min) and steeped overnight. The steep liquor (called *nejayote*) is discarded. The cooked-steeped maize (nixtamal) is washed to remove excess alkali and loose pericarp tissue. Then, it is ground into dough (masa) and flattened into thin disks that are baked (Rooney and Serna-Saldívar 1987). Cooking disrupts the crystalline structure of corn starch. The starch then recrystallizes or anneals during steeping to form a new polymeric structure. Grinding of cooked corn releases starch granules from the endosperm and reduced their crystallinity (Gomez et al. 1992). The reduction of crystallinity in starch, measured by X-Ray powder diffractometry, accounts for 15-25% by nixtamalization and 1-10% by milling operations (Gomez et al. 1989).

Alkaline cooking is another general term for this process (Serna-Saldívar et al. 1990). Alkaline cooking improves flavor, starch gelatinization, and water uptake and partially removes the germ and pericarp of the corn kernels. The maize kernel is only partially cooked. Steeping allows moisture and lime to distribute throughout the cooked grain. Stone grinding disrupts swollen gelatinized starch granules and distributes the hydrated starch and protein around the ungelatinized portions of the corn endosperm, forming masa (Rooney and Serna-Saldívar, 1987). The masa obtained from the nixtamalization process is a network of dispersed and soluble starch polymers, starch granules partially gelatinized in a continuous water phase supported by free starch granules, other pieces of endosperm and lipids (Gomez et al. 1990).

Commercial technology is based on these principles (Fernandez et al. 1999). One of the characteristic stages of the commercial process is the ability of the dough or masa to form a uniform sheet that is cohesive and elastic but with minimum

adhesiveness (Pflugfelder et al. 1988). The development of masa cohesiveness can be attributed to grain disruption and liberation of other components, perhaps proteins and pentosans, and only to a small proportion to a limited solubilization of starch (Campus-Baypoli et al. 1999). Also, the alkaline pH and temperature influences the hydrolysis of hemicelluloses. About 25% of hemicelluloses in corn pericarp are solubilized during nixtamalization (Saulnier et al. 1993), yielding an abundance of pentoses, glucuronic acids and some deoxy sugars. The pericarp contains gums that improve masa cohesiveness and machinability (Saulnier et al. 1993). Therefore, corn pericarp increases water absorption improving corn tortilla texture. Pericarp acts like a natural gum that retains moisture (Guajardo-Flores 1998).

Nixtamalized Corn Flour

Nixtamalized Corn Flour (NCF) is produced by the nixtamalization method used to produce traditional masa. The particles are dried and fed into a sifter, and the appropriate fractions are recombined to provide flours with desired particle size distribution and other properties (Rooney and Serna-Saldívar, 1987).

NCF can be re-hydrated to produce masa, but has different rheological properties than fresh masa; it is less plastic and cohesive and the tortillas stale faster (Gomez et al. 1991). The distribution of moisture in rehydrated masa is not equivalent to that found in fresh masa (Pflugfelder 1986). However, many manufacturers use NCF because it meets standards for certain applications, reduces requirements for labor, energy, floor space, processing time, and equipment for washing and grinding corn, and is very convenient and easy to process into tortillas or snacks (Serna-Saldívar et al. 1990).

Commercial corn masa may be described as particles of corn endosperm, germ, pericarp and tip cap dispersed in a mass of partly gelatinized free starch granules, fragments of cell wall and protein, free lipid and dissolved solids. The free starch fraction molecules or granules accounted for a majority of masa starch and varying amounts of fiber and protein (Pflugfelder 1986).

It is likely that masa free starch, dissolved solids, and free lipids are the primary determinants of the texture, flavor, and keeping quality of masa products (Pflugfelder et al. 1988). Gelatinization of free starch during baking into tortillas and/or frying of masa into chips provides the matrix in which particles are embedded (Pflugfelder et al. 1988).

Corn Tortillas

When masa is molded and baked into tortillas, the starch granules gelatinize and lose their structure and integrity. Some amylopectin leaches out of the granule; along with much of the amylose (Quintero-Fuentes 1999) and birefringence is almost completely lost (Campus-Baypoli et al. 1999). Approximately 35-40% of the total starch crystallinity as determined by X-ray powder diffractometry is lost during the baking process. Baking caused partial to complete starch gelatinization (Gomez et al. 1992) but starch granules are only partially dispersed (Fernandez et al. 1999). Tortilla texture is probably highly dependent on gelatinization of free starch during baking (Pflugfelder et al. 1988).

Amylose gels establish a three-dimensional network right after baking. Starch gels are composed of swollen starch granules filling an interpenetrating amylose gel (Suhendro 1997). Upon cooling, the dispersed amylose associates to form an insoluble amylose network (retrogradation) since tortillas have a reduction in soluble amylose content and these associations cause a rigid structure in the corn tortilla (Fernandez et al. 1999). The ability of starch polymers to form ordered structures upon cooling, via interchain associations, is a critical step for setting of hydrated three-dimensional gel networks and the development of texture for thermally processed products (McGrane et al. 2000).

Staling

Limited shelf life (due to microbial spoilage) and rapid staling are major problems affecting the commercial distribution of corn tortillas (Serna-Saldívar et al. 1990). Staling is defined as the loss of freshness in tortilla, or can be defined in terms of

almost any change, short of microbial spoiling, that occurs during storage which makes baked products less acceptable to consumers (Zobel and Kulp 1996).

Consumers prefer tortillas which are soft and have good flexibility and rollability, while rigid, firm and less rollable tortillas are undesirable (Limanond et al. 2002). The economic losses caused by staling are extremely important. Consumer acceptability of tortillas is highly linked to texture and handling properties with staling being the primary reason for tortillas to be discarded (Limanond et al. 2002). In a market of approximately 450 million tons of corn and wheat tortillas produced annually in the USA, Mexico and Latin America, the return of 4% as unsalable represents a sizable economic burden on both producer and consumer (Limanond et al. 2002).

Staling is an extremely complex phenomenon and is difficult to define. Staling refers to all changes that occur in a baked product after it comes out of the oven. The increase in firmness has probably been used to the largest extent to quantify staling (Suhendro 1997; Quintero-Fuentes 1999; Yeggy 2000; Limanond et al. 2002). Other changes such as loss of flavor, decrease in water absorption capacity, amount of soluble starch and enzyme susceptibility of the starch, increase in starch crystallinity and opacity, and changes in X-ray diffraction patterns have also been used (Quintero-Fuentes 1999).

Staling is a progressive, spontaneous aggregation of the starch components and the corn tortilla textural changes were mainly related to modifications in the starch properties. The gelatinized starch reassociates during staling into an ordered structure, called 'retrograded starch' (Miranda-Lopez 1999, Fernandez et al. 1999). This process starts right after tortillas are made.

During cooling, the amylose fraction retrogrades very fast. The time dependent gelation of amylose occurs in two stages: a relatively fast viscoelastic network formation via entanglement which is not thermoreversible but dilution reversible; followed by a slower, continuous crystallization (folded or extended-chain) process which is thermoreversible (Slade and Levine 1991, Quintero Fuentes 1999); also supported by the fact that reheating gives fresh-like properties in corn tortillas (Yeggy 2000). During

storage amylopectin continues to retrograde, imparting rigidity to the structure (Quintero-Fuentes 1999). Amylose may form double-helical associations of 40-70 glucose units, whereas amylopectin forms shorter double helices than amylose due to restrictions imposed by the branching structure of the molecules and the chain lengths of the branches. Double helices may associate and organize into crystallites; gelation results under appropriate conditions (Klucinec and Thompson 1999).

Recrystallization stiffens the granules, resulting in reinforcement of the amylose gel matrix. Combined amylose-amylopectin co-crystallization may also occur, improving the binding of granules within the amylase gel, resulting in increased rigidity and firmness of the corn tortillas. Recrystallization of amylopectin and amylose-amylopectin co-crystallization in corn tortillas enhanced the solid-like characteristics (Suhendro 1997). Starches in nixtamalized corn flour and masa are partially gelatinized and annealed. These remnants are the nuclei of starch associations and this will facilitate the propagation of crystal structures (associations of amylose and/or amylopectin).

After baking, tortillas initially have dry surfaces, which make them less flexible, with decreased subjective rollability and crinkle scores. After equilibration (about 1 h), tortillas are soft, rollable and flexible (Fernandez et al. 1999). Apparently, moisture from the interior of the tortilla migrated to the exterior. Rapid changes continue to occur; after 4 h of storage, force to deform the tortilla is increased (Fernandez et al. 1999). The rate of staling is higher initially; then the rate decreases but continues during storage (Miranda-Lopez 1999).

During storage, baked products become firmer and break upon bending (Quintero-Fuentes 1999). Rapid changes in starch properties can be evaluated by stabilizing the starch with a methanol dehydration procedure and measuring starch properties such as pasting viscosities and starch solubility (Fernandez et al. 1999, Silva et al. 2002).

Amylose solubility decreases as amylopectin solubility increases in freshly baked tortillas. Increased amylopectin solubility shows that starch is gelatinized and easily dispersed. Amylopectin appears to impart desirable softness and extensibility to tortillas.

By finding a way to delay retrogradation of amylopectin, stability of corn tortillas could be increased (Fernandez de Castro 1998).

Fernandez et al. (1999) found two kinds of gelatinized starch in corn tortillas: the gelatinized and easily dispersed starch; and the gelatinized starch with enhanced inter- and intragranular structures requiring higher temperatures for pasting. The second kind is the starch that has associated fractions that retrograde and thus, causes hardening of corn tortilla during storage.

The rate of retrogradation is highly influenced by the extent of gelatinization. The increased levels of gelatinized, disrupted and solubilized amylose and amylopectin fractions, will give lower rates and extent of retrogradation. This was shown by the results obtained by Seetharaman et al. (2002), where the harsher processing, i.e., higher temperature, longer time and higher moisture environment, resulted in greater dispersion of amylose and amylopectin, and retrogradation of amylose. Experiments show that heating to various temperatures above the range of gelatinization may profoundly affect amylopectin retrogradation, perhaps due to varying extents of residual molecular order in starch materials that are commonly presumed to be fully gelatinized (Fisher and Thompson 1997).

Prevention of Staling

Methods to retard staling include (1) Storage at temperatures below T_g ; (2) Control of effective T_g by moisture content and/or product formulation, relative to storage temperatures, so that conditions for crystallization become less favorable; (3) Modification of starch structure to reduce intermolecular associations and continuity of the gel network (by the addition of hydrolytic enzymes to cleave interconnecting chain segments between crystallites, lipids to complex with individual starch chains, and sugars to decrease the rate of chain aggregation); and (4) Incorporation of hydrocolloids for better moisture retention and a softer texture (McGrane et al. 2000).

Recrystallization of amylopectin plays a major role in the staling mechanism, resulting in increased rigidity and firmness (Fernandez et al. 1999, Miranda-Lopez 1999,

McGrane 2000, Hibi 2001, Limanond et al. 2002). Extent of corn tortilla staling was quantified in terms of degree of crystallinity using data from stress relaxation and Differential Scanning Calorimetry and polymer crystallization theory. DSC indicated that there was more crystal formation (amylopectin retrogradation) during tortilla storage (Quintero-Fuentes 1999, Limanond 2000). Fresh corn tortillas have a lower value of final stiffness and dissipated more energy than stale tortillas. The staling enthalpy in fresh tortilla increases with time indicating an increase in degree of crystallinity. The melting point of fresh tortilla endotherm is significantly less than those of stored, stale tortillas. This means that the amylopectin develops more perfect crystals as staling progresses. (Limanond 2000).

The development of crystallites depends on the probability that a nucleus will develop into a grain and thereafter grows at a steady state, therefore the crystallization takes place more rapidly the further the temperature falls below the melting point of the crystalline phase (Limanond et al. 2002). The crystallization process of corn tortilla is nucleation-limited, and the maximum crystallization temperature was between 10.2 and 12.3, as measured from final stiffness and staling enthalpy data. These temperatures should be avoided to prevent an accelerated rate of nuclei formation (i.e. accelerated staling) (Limanond 2000).

Since recrystallization of amylopectin is mainly of importance in the staling of baked products during storage, an ideal storage condition would be below T_g , but above the melting point of ice in the system. By manipulating crystallization it should be possible to accelerate or inhibit staling (Quintero-Fuentes 1999). For storage at temperatures higher than network T_g , there is sufficient mobility for the formation of crystalline junction zones, resulting in a partially crystalline polymer system (retrograded starch) (Miranda-Lopez 1999). However, the control of temperature is a method not economically feasible. Some anti-staling agents increase the T_g ; thus, the starch recrystallization propagation rate is lower at the storage temperature (Miranda-Lopez 1999)

Additives as Anti-Staling Agents

Enzymes

Staling can be reduced by using amylases that hydrolyze α -1-4 linkages within the amorphous regions of the starch matrix during baking (Zobel and Senti 1959). Addition of α -amylase to bread decreases staling of the bread by changing the structure of starch. The anti-staling effect of the branched-chain products is also believed to be caused by a decrease of or an interference with the crystallization of amylopectin or from interference with the formation of other interactions (Duedahl-Olesen et al. 1999). α -amylase induces dextrinization of starch granules reducing its ability to immobilize water, and free water increases dough mobility (Miranda-Lopez 1999).

During cooking, the amylase diffusion rate and activity reach a maximum. At the same time, the amylolysis activation is counterbalanced by thermal inactivation. The enzymes are deactivated as the surface cooking temperature of 80-90°C is reached and starch hydrolysis stops (Iturbe-Chiñas et al. 1996, Drapron and Godon 1987). However, amylolytic activity can be shorter or longer depending on the thermal kinetics inside the product (Drapron and Godon 1987, Iturbe-Chiñas et al. 1996).

Martin et al. (1991) studied the role of starch hydrolyzing enzymes using the theory of bread firming; they concluded that fungal or bacterial α -amylase produced low molecular weight dextrans and maltose may diffuse away, reducing starch-protein interactions. A bacterial maltogenic amylase has been found to have anti-staling effect (Outtrup and Norman 1984). Si (1997) reported that maltogenic α -amylase (novamyl) reduces the rate of starch retrogradation, while fungal amylase did not affect it (Miranda-Lopez, 1999). It can degrade both amylose and amylopectin at the gelatinization temperature and produces mainly α -maltoses (Quintero-Fuentes 1999).

A limited hydrolysis of amylopectin chains can inhibit retrogradation with the concomitant extension of the shelf life (Iturbe-Chiñas et al. 1996). Fungal α -amylases added at low levels to dry masa flour or in water form making masa may be a valuable tool to reduce starch retrogradation and retard accelerated staling in tortillas, increasing their shelf life (Iturbe-Chiñas et al. 1996).

However, Suhendro (1997) found that tortillas containing bacterial, fungal and malt amylases were softer to the touch but were more brittle as measured by bending technique, and more breakable when rolled on a dowel. Bacterial amylase enhanced softness of the tortillas; but the tortillas were more brittle than those with fungal amylase or malt (Suhendro 1997). Enzymes also affected adversely masa machinability, softer, stickier reduced cohesiveness, so additives that can increase viscosity and create a new network of viscoelastic structure to compensate for the weakened structure affected by enzymes, like CMC, powdered cellulose, and monoglycerides, can be used to reduce tortilla staling (Suhendro 1997, Miranda-Lopez 1999).

Non-Starch Polysaccharides (Hydrocolloids)

Starch gelatinization is influenced by the presence of small-molecular-weight solutes and hydrocolloids. Understanding these effects is important for better process control and for improving the texture and other quality attributes of starch-based products (McGrane et al. 2000).

Among the numerous macromolecules of natural origin some disperse easily and significantly increase viscosity. These food-thickening and gelling agents are known as water-soluble gums or hydrocolloids. These biopolymers can be used for stabilization of suspensions and emulsions, water holding capacity, binding properties, and formation of complexes with proteins (Linden and Lorient 1999). Hydrocolloids are compounds capable to control both the rheology and texture of aqueous systems throughout the stabilization of emulsions, suspensions and foams and are also able to modify starch gelatinization and to extend the overall quality of the product during storage (Rosell et al. 2001).

Hydrocolloids are added into masa to improve texture, eliminate adhesiveness of packaged tortillas, improve freeze-thaw stability and increase yield (Serna-Saldívar et al. 1990). These compounds plasticize the amorphous region, either through water retention or by inhibiting polymer interactions (Miranda-Lopez 1999). Some hydrocolloids have the ability to develop a network through various types of physical and chemical

interactions that result in a viscoelastic structure (Suhendro 1997). However, the addition of excessive hydrocolloids imparts an undesirable gummy and chewy texture in the baked products (Suhendro 1997).

Increased water absorption enhances tortilla softness and retards firming (Fernandez de Castro 1997, Quintero-Fuentes 1999). Tortillas containing hydrocolloids have moisture contents significantly higher than control, which means that the hydrocolloids bind water during mixing and retain the moisture during baking (Yeggy 2000). CMC binds additional water that might lower Tg enhancing the rubbery elastic texture, which requires more force to extend, and extends a longer distance when stress is applied (Yeggy 2000).

Serna-Saldívar et al. (1990) recommends levels from 0.25-0.5% for use in tortilla manufacture. Yeggy (2000) found that CMC at 0.5% had the greatest positive effect on texture of tortillas, among different hydrocolloids tested in corn tortillas. CMC helped tortillas to maintain flexibility for up to 19 days of storage (Suhendro 1997).

CMC is a long chain, linear, water soluble, chemically modified, anionic, cellulose-based polysaccharide with a molecular weight of approximately 700,000 (Yeggy 2000). CMC hydrates and builds viscosity quickly due to repulsions between the charged substituents (Miranda-Lopez 1999). The ability of CMC to develop viscosity and increased structural integrity make it the best choice for industrial use in improving corn tortilla shelf life and acceptability (Yeggy 2000). The hydrocolloid is localized in the continuous suspension phase whose volume decreases as the grains of starch increase. The consequence is a clear increase in gum concentration in this phase, resulting in an extremely spectacular increase in the viscosity of the continuous medium (Linden and Lorient 1999).

CMC decreases starch gelatinization due to competition for water in the dough, and improvement of corn tortilla rollability could be caused by less starch gelatinization and retrogradation (Suhendro 1997, Yau et al. 1994). Hydrocolloids also inhibit recrystallization of gelatinized starch and decrease staling rate of baked goods (Yau et al. 1994). Better quality of tortillas with CMC may be due to the size of the polymer and its

ability to disrupt the continuous phase (Yeggy 2000). Improvement of tortilla rollability could be due to the formation of a new, continuous network that formed as CMC solubilized during preparation creating an improved matrix with increased strength through the association between CMC strands forming junction zones (Suhendro 1997, Yeggy 2000). Quintero-Fuentes (1999) found that CMC retained rollability to tortillas by forming a flexible gel-network, since it did not decrease tortilla water solubility during storage. These results confirmed that CMC improved tortilla texture during storage but it did not retard starch retrogradation. The addition of large molecules, such as CMC, to corn tortillas may lower the glass transition and allow the system to maintain the rubbery state (it has a low glass transition temperature) (Suhendro 1997, Cauvain 1998).

Corn Bran

Nixtamalized commercial corn brans significantly improved the texture of corn tortillas during storage and enhanced the color, flavor and aroma of DMF tortillas. Use of 5% of nixtamalized corn bran was the most acceptable as rated by the sensory panel. This bran can be used as an effective additive to extend shelf stability of tortillas and to improve the flavor of DMF products, since tortillas will stay soft and flexible without microbial contamination for a longer time (Guajardo-Flores 1998).

Nixtamalized bran functions in tortillas in two different manners. First, alkaline pH ionizes starch molecules that form bonds with calcium ions preventing retrogradation. Second, nixtamalized bran form a gum which retains water in the system. These two factors help to conserve tortilla freshness for longer times (Guajardo-Flores 1998). However, the disadvantage of using nixtamalized corn bran is the strong flavor and dark color of the tortillas.

Barley

Barley (*Hordeum vulgare*) is a cereal harvested with the hull intact. The caryopsis is composed of pericarp, seed coat, germ, and endosperm (Hoseney 1994). For

human consumption, it is dehulled and pearled by an abrasive process that removes approximately 35% of the grain weight including glums, pericarp, germ and aleurone layer (Serna-Saldívar 1996). To obtain refined flour, the pearled grain is passed through a series of rollers. Barley flours can hold approximately 2.5 times more water than wheat flour (Klahorst 2000).

Barley and oats are considered major sources of β -glucans (3-9%) (Doxastakis and Kiosseoglou 2000). β -glucans from oat and barley concentrated by dry milling methods have been used to produce enriched flour that can be substituted for a portion of wheat flour in many cereal products including bread, muffins and pasta. However, the lack of gluten of barley restricts its use in leavened bread, since loaf volume decreases after 10% substitution, and breadcrumb color gets darker (Klahorst 2000; Knuckles et al. 1997; Marklinder et al. 1996).

β -glucans are linear homopolysaccharides composed of D-glucopyranosyl residues (GlcP) linked via a mixture of β -(1-3) and β -(1-4) linkages, as shown in Fig.2. The molecular structure of β -glucans from various cereals and plant tissues may differ drastically. Reported molar ratios of (1-3)-linked cellotriosyl to (1-3)-linked cellotetraosyl units, which constitute 85-90% of the β -glucans, vary between 2.8 to 3.3 for barley (Doxastakis and Kiosseoglou 2000). Chemical analysis of isolated aleurone cell walls from barley indicates that they are composed mainly of arabinoxylans (67-71%) and smaller amounts of β -glucans (26%), whereas the endosperm cell walls are built mainly of β -glucans (70%) and to a lesser extent of arabinoxylans (20%) (Doxastakis and Kiosseoglou 2000).

One of the most important physical characteristics of β -glucans, from the point of view of their application in food systems, is their viscosity. They can impart high viscosity to aqueous solutions due to their high molecular weight, conformation and interactive properties and may also modify the texture, water binding properties, and sensory attributes of a food product (Doxastakis and Kiosseoglou 2000).

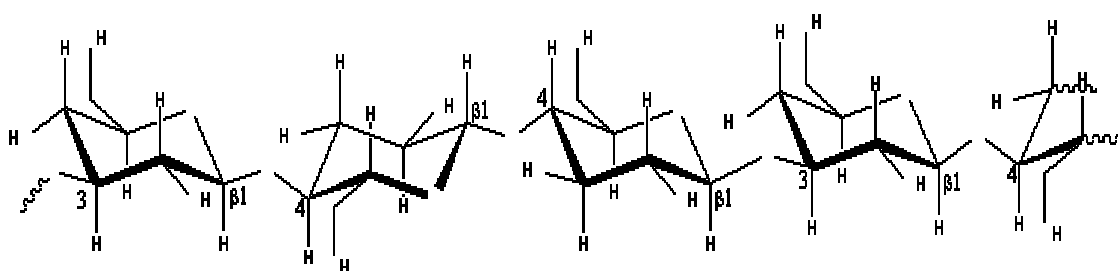


Fig. 2. Structure of β -glucans.

The role of β -glucans in baked products has not been explored to the same degree as that of arabinoxylans. The study of these polymers may be limited because of the absence of efficient processing methods to isolate these materials in large scale and in relatively pure form and because the use of both β -glucan enriched cereals, barley and oats, have not been extensively used in leavened products. However, with the current interest in functional foods, it is likely that the role of β -glucans in such systems will become more prominent not only because of their potential health benefits but also because of their technological functionality (Doxastakis and Kiosseoglou 2000).

Mitre-Dieste (2001) substituted barley flours for DMF at 10-25%, to evaluate if tortilla quality improved, and dietary fiber content increased significantly. β -glucan content of flours correlated positively with water absorption. As barley substitution increased, moisture content of the doughs and tortillas decreased because barley flours have lower water absorption capacity than DMF. Moister doughs processed more easily and tortillas were softer and more extensible. After comparing sensory characteristics with objective and subjective analysis, it was determined that 15% was the optimum barley substitution level. These tortillas showed better scores for extensibility and rollability than corn tortillas, while keeping the sensory characteristics of corn tortillas (Mitre-Dieste 2001). The interaction of amylose and amylopectin, as well as the added β -glucans of the flours helped reduce the staling of the tortillas. The added β -glucans and amylopectin of waxy barley flours increased the water absorption capacity of the composite flour, compared to 100% barley tortillas (Mitre-Dieste 2001).

Addition of β -glucans to bread and other bakery products is increasing due to their hypocholesterolemic activity (Koksel et al. 1998). Soluble β -glucans have been reported to substantially reduce plasma cholesterol and postprandial serum glucose and insulin levels in humans and animals (Doxastakis and Kiosseoglou 2000).

Overall, from a nutritional and a functional viewpoint, cereal foods rich in non-starch polysaccharides fit into the description of 'functional foods' as they provide some of the normal quality attributes of a food, like sustenance and texture, as well as confer specific health benefits (Doxastakis and Kiosseoglou 2000). With the growing demand for nutraceuticals and functional foods, considerable effort will be directed in the near future towards increasing the content and improving the functional properties of these polymers in the popular cereals (Doxastakis and Kiosseoglou 2000).

Texture Evaluation Methods

The subjective techniques commonly used for tortilla evaluation are rollability and pliability. Rollability consists in the use of a dowel to roll the tortilla and score the texture by evaluating cracking and breaking subjectively (Bello et al. 1991). Pliability or squeezability evaluates texture in the same way but the tortilla is held in the palm of the hand and squeezed to be scored.

Suhendro (1997) found that subjective rollability technique was sensitive to textural differences in table tortillas after 1 day of storage. Bueso-Ucles (2003) stated that tortilla rollability remains the basic subjective indicator of tortilla texture, but changes in tortilla texture were detected faster using subjective pliability than rollability, and pliability correlated better with tortilla stiffness.

Stiffness is a general term that refers to the response of food materials to an external stress and stress relaxation is an accurate method to measure this mechanical property as well as energy dissipated (Limanond 2000). By this method, an instantaneous strain is given and the stress required to maintain the deformation is observed as a function of time. An ideal elastic material would show no relaxation while the ideal viscous material would relax instantaneously. Viscoelastic materials would

relax gradually with the end point depending on the molecular structure of the material being tested (Steffe 1996). The change on stiffness as a function of time and temperature may be directly proportional to the degree of crystallization in the starch fraction and the stress relaxation technique successfully detected the textural differences between corn tortilla samples at various storage times and temperatures in terms of stiffness (Limanond 2000). Suhendro (1997) also recommends the stress relaxation technique for fundamental research, for it provides information about viscosity and elasticity of corn tortillas that are not provided by other techniques. The 7-element Maxwell model showed to be sensitive to the textural changes of corn tortillas from fresh up to staling where stiffness and energy dissipated for tortillas were chosen as best texture property predictors and showed good correlation with the subjective rollability scores (Guo 1998).

Another objective measurement of tortilla texture is the extensibility test, in which the tortilla is pulled apart until it breaks. The force and distance to rupture is measured. According to Bueso-Ucles (2003), tortilla rupture force may be a misleading indicator of tortilla hardness, since higher forces do not necessarily mean tortillas are harder or more brittle. Tortilla final stiffness obtained by stress relaxation is a significantly better indicator of tortilla hardness while rupture distance explained changes in tortilla extensibility better.

An effective additive or combination of additives should increase pliability, rollability, rupture distance and energy dissipated, while decreasing rupture force, Young's modulus, and final stiffness.

CHAPTER II

MATERIALS AND METHODS

Raw Materials

The materials used for the production of tortillas are listed in Table I.

TABLE I
Raw Materials Description

Raw material	Description
Dry masa flour (DMF)	Tortilla #4 with no additives. Minsa, Red Oak, IA
Potassium sorbate	ADM Arkady, Olathe, KS
Fumaric acid powder	Balchem Co. Slate Hill, NY
Carboxymethylcellulose (CMC)	Blanose®7HF Molecular mass: 4.35×10^5 g/mol, degree of substitution: 0.65-0.90, pH: 6.5-8.5, sodium fraction: 7-8.9% and average viscosity: 2500 mPa.s at a 1% concentration. Hercules Incorporated, Aqualon Division, Wilmington, DE
Amylase	Innovative Cereal Systems, Wilsonville, OR
β -glucan	Barley β -glucan (70%) from Cargill Health Foods and Technologies, Wayzata, MN

Tortilla Production

The tortillas were produced in the Cereal Quality Lab laboratory pilot plant at Texas A&M University, using the corn tortilla production technology. The flours were mixed for 5 min with fumaric acid (0.38% Baker's percentage) and potassium sorbate (0.38% Baker's percentage) with paddle and then hydrated with distilled water (120%) and kneaded with a hook for 1.5 min at slow speed in a 20 qt mixer (Model A-200, Hobart, Troy, OH) and then at second speed for 0.5 min. Doughs were placed in low-

density polyethylene bags and allowed to rest at room temperature for 5 min, to equilibrate moisture before processing. Doughs were sheeted and cut into 15 cm diameter, flat, 30 ± 1.1 g tortilla disks with a tortilla sheeter (model CH4-STM, Superior Food Machinery, Inc., Pico Rivera, CA). Tortilla disks were baked in a three-tier, gas fired oven (Model C-0440, Superior Food Machinery, Inc., Pico Rivera, CA) for 65 s. the temperature profile of the oven was 320-280-260°C. After the tortillas left the oven, they were cooled to room temperature in another conveyor for 2.5 min. Finally, the tortillas were packaged in plastic bags and stored for 4 hours at room temperature to do the initial measurements. Tortillas were stored in freezer at 4°C.

Masa Evaluation

Masa was evaluated for water absorption subjectively. 100 g dough samples were prepared in a commercial mixer (Model K45, Kitchen Aid, St. Joseph, MI). Treatments that included dusted barley flour from shorts were compared to the control masa. Different levels of water were tried. Dough had to be cohesive and not sticky for processability (Mitre-Dieste 2001). In addition, good processability is defined by machinability, where doughs could be easily sheeted and baked in the pilot plant equipment with a minimum of cripples.

Tortilla Evaluation

Initial measurements included tortilla moisture, pH, diameter, thickness, yield, weight and color.

Moisture (%). Percentage of moisture was determined by the AACC method 44-15A (one stage), for masa and tortillas. It was determined in duplicate with two replicates. Tortillas were ground in a coffee grinder (Model E160 Type CM03, Proctor Silex, Washington, NC). Samples were kept in individual polyethylene bags prior to analysis to avoid moisture loss. Two to three grams of masa or ground tortillas were transferred with a spatula to tared aluminum dishes. Weight was recorded and dishes transferred to an oven set at 130°C for 1 h after temperature equilibration. The dishes

were removed and placed in a dessicator, allowing them to reach room temperature and then weight was recorded. Loss in weight was determined as moisture and calculated using the following equation:

$$\% \text{ Moisture} = \text{moisture loss (g)} / \text{original sample weight (g)} \times 100$$

pH. Ten grams of ground sample were stirred in 100 g of distilled water at 25°C for 10 s. The pH was determined with a potentiometer (Beckman Instruments Inc., Fullerton, CA) calibrated against buffer solutions (pH 4 and 7).

Diameter quotient. Tortillas were measured at largest and shortest diameters. Quotient was calculated by dividing the largest by the shortest diameter. A diameter quotient of 1.0 indicated round tortillas. Increased quotient for tortillas indicated an oval shape. Three tortillas were measured from at least two replicates.

Thickness (mm). An electronic digital caliper (Chicago Brand, NTX, Inc., Cleveland, OH) was used to individually measure tortillas, in duplicate with four replicates.

Yield. The total weight of tortillas per batch was measured. Yield was calculated as this weight divided by the total batch weight from formulation.

Weight. Tortillas were weighed individually in a digital scale (Model Galaxy™4000, OHAUS Scale Corp., Florham Park, NJ) in duplicate with four replicates.

Color. A color meter (model CR 310, Minolta Co., Ramsey, NJ) was used to measure color, calibrated with a white tile ($Y=94.3$, $y=0.3321$, $z=0.3157$). Three points of four whole tortillas were measured and compared in 2 replicates. Values determined where L^* (luminosity: 0=black, 100=white), a^* (redness (+), greenness (-)), and b^* (yellowness (+), blueness (-)). The overall color difference (OCD) was determined by comparing the tortillas to a standard (control) with the following equation:

$$\text{OCD} = \sqrt{[(L_{\text{std}}-L)^2 + (a_{\text{std}}-a)^2 + (b_{\text{std}}-b)^2]}$$

Texture Evaluation

Texture was evaluated initially 4 h after production and then in tortillas stored for 1 d, 4 d, 7 d and 14 d at 4°C after equilibration at room temperature.

Subjective Texture Measurements

Rollability

A tortilla was wrapped and rolled around a 1 cm wood dowel. The rollability scale used was from 1 to 5, where 1= unrollable, breaks easily; 2= cracking and breaking on both sides; 3= cracking and breaking beginning on one surface; 4= signs of cracking but no breaking; 5= no cracking, easily rollable.

Pliability

A tortilla was held in the palm of the left hand, squeezed uniformly and released after 5 sec. Pliability score was rated from 1 to 5, where 1= complete crumbling; 2= almost total crumbling; 3= cracking but no crumbling; 4= signs of cracking but no breaking; 5= completely pliable, no cracking.

Objective Texture Measurements

Stress Relaxation- Final Stiffness and Energy Dissipated

The food products were analyzed using a Texture analyzer (model TA-XT2, Texture Technologies Corp., Scarsdale, NY/ Stable Microsystems, Godalming, Surrey, UK) with a bench top movable system, and the attachment for tension test. The samples of corn and barley tortillas were cut into a 35mm x 70mm rectangle. The sample was gripped with a clamp, with one end attached to the Texture Analyser platform and the other end attached to the Texture Analyser arm. The distance between the two arms was 21.8 mm. The set up to perform force in tension was used and the samples were tested at 3% strain levels (linear viscoelasticity region). The instrument was set in the “measure force in tension” and “hold until time” mode. The force was measured in Newtons, pre-test and post-test of 1 and 5 mm/s, respectively, test speed of 0.1 mm/s, trigger type: auto at 0.05N. A graph of force and time was saved on a file for further data analysis. The

stress relaxation data (force as a function of time) were transformed into relaxation modulus, E , and then fitted to a generalized Maxwell model with seven parameters using the Matlab program developed by Spadaro (1996) and Guo et al. (1999). Data were then transformed into stiffness and energy dissipated using Matlab Software (Matlab 1996).

Extensibility – Rupture Force, Rupture Distance and Young’s Modulus

Texture Analyser was used to evaluate the extensibility of tortillas (force and distance to rupture, Young’s modulus), with a force in tension test. The test was conducted using the “return to start” option, with trigger force of 0.05N. Tortilla strips were cut using a 35 x 70 mm rectangular acrylic template. Samples were placed individually on the platform directly under the test probe, and held by two metal clamps set at a separation of 21.8 mm. One clamp was attached to the electronic movable arm and the other to the static platform. The test consisted of pulling the tortilla strip at a constant speed of 1mm/s, until it ruptured completely. Pre-test and post-test speed was set to 5 mm/s. Rupture force (N) and distance (mm), and Young’s modulus (initial linear slope, N/s) were recorded. Data was analyzed using XTRAD’s software (Mitre-Dieste, 2001).

Statistical Analysis

All experiments were conducted in duplicate and treatments were completely randomized. Data was analyzed with the SAS System for Windows Version 8e (SAS Institute Inc., Cary, NC, USA, 1999-2000). Response surface methodology was analyzed with a response surface regression (RSREG) procedure. Least significant differences (LSD) and analysis of variance (ANOVA) tests were conducted at a 0.05 level of confidence by the general linear model (GLM).

Experimental Design

- 1) Dusted barley flour from shorts was substituted for 2.5, 5 and 10% of dry masa flour in the formulation for tortilla production. Tortillas were evaluated and an optimum level determined for further experiments.

- 2) The optimum level determined in Experiment 1 was combined with CMC and amylase in a central composite design using response surface methodology. Tortillas were evaluated and optimum levels determined for further experiments.
- 3) β -glucan content of optimum levels of flour was calculated. Concentrated β -glucans were substituted according to the levels present in the barley flour and combined with amylase as determined in Experiment 2. Tortillas were evaluated and optimum levels determined.
- 4) Concentrated β -glucans were substituted according to the levels present in barley flour and combined with CMC as determined in Experiment 2. Tortillas were evaluated and optimum levels determined.

CHAPTER IV
BARLEY FLOUR SUBSTITUTED FOR DRY MASA FLOUR ON CORN
TORTILLAS:
RESULTS AND DISCUSSION

The objective of this experiment was to evaluate the effect of barley flour substituted for 2.5-10% of the dry masa flour in corn tortillas. Canadian dusted barley flour from shorts with 10.5% β -glucan, 4% amylose (waxy) and 19.3% dietary fiber was used. Flour from shorts was chosen because it had increased soluble dietary fiber and β -glucan content compared to the 50% extraction flours. Water absorption was adjusted for the barley treatments.

Water Absorption

Doughs with barley flour were compared to NCF masas. Water absorption of 100% dry masa flour for corn tortilla production was 120%. Tortillas made from 100% of dusted barley flour from shorts with 10.5% β -glucan and 4% amylose had a water absorption of 110%, as previously calculated from mixographs (Mitre-Dieste 2001). Cohesive doughs were obtained by optimizing water absorption. As barley flour was substituted for nixtamalized corn flour (NCF), water absorption decreased (Table II).

TABLE II
Formulation with Adjusted Water Absorption Level

Barley flour Substitution	0%^a	2.5 %	5 %	10 %
DMF	100	97.5	95	90
Water	120	115	110	110
β -glucan content ^b	-	0.26	0.53	1.05

^a Control; ^b Calculated value from flour β -glucan content.

Good processability was shown by machinable masas with no yield losses. Changes in water absorption were related to the lower water absorptions from barley flours compared to dry masa flour. Mitre-Dieste (2001) reported that water absorption was positively correlated to β -glucan content. Knuckles et al. (1997) found that using β -glucan enriched barley fractions in bread and pasta, increased water absorption. In the present study, the level of substitution is changed and water absorption was not correlated to the β -glucan content, but to the amount of barley flour used.

Tortilla Moisture, pH Thickness, Diameter Quotient, Weight and Color

Moisture content did not change significantly ($p < 0.05$) during storage (Table III), which confirms that packaging prevented moisture loss. Barley flour significantly reduced moisture content of corn tortillas (Table III). After 1 and 7 days of storage, moisture contents of corn tortillas with barley flour substitution at 2.5 and 5% were not significantly different than control. Only tortillas with 10% of barley substitution had significantly reduced ($p < 0.05$) moisture content. Mitre-Dieste (2001) also found the moisture contents of corn tortillas containing 10% barley flour significantly lower compared to control tortillas.

TABLE III
Effect of Barley Flour Substitution on Moisture Content of Corn Tortillas During Storage at 4°C^a

Barley Flour substitution (%)	Moisture content (%)		
	4 hr	1d	7d
0.0 (Control)	47.1 ^a	46.1 ^a	45.6 ^a
2.5	45.1 ^b	45.1 ^{ab}	45.2 ^a
5.0	44.7 ^b	46.0 ^a	44.1 ^{ab}
10.0	42.2 ^c	44.1 ^b	42.9 ^b
LSD_{sample}($\alpha=0.05$)^b	1.5	1.8	2.1
Mean	44.8	45.3	44.4

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Barley flour did not significantly change tortilla thickness, pH and diameter quotient (Table IV). Mitre-Dieste (2001) reported that tortillas with 10% barley flour substitution had no changes in diameter quotient, spread factor (average diameter/thickness) or pH.

Control tortillas were significantly heavier than tortillas with 2.5 and 5% barley flour (Table III). Barley tortillas had less water added to the formulation, as seen in the initial moisture content (Table III).

Tortilla weight increases while moisture content decreases in tortillas with barley flour. Tortillas with 10% barley flour were not significantly different from the control weight, but had higher weight than 2.5 and 5%; this treatment had significantly lower moisture content and significantly higher weight than the control. Mitre-Dieste (2001) also reported no difference in tortilla weight and significantly lower moisture content in tortillas with 10% barley flour. However, differences in tortilla weight are not consistent with tortilla moisture contents. Since the tortilla weight is determined by the distance between the rollers during sheeting into 30 g flat round shapes, it is more likely that masa weights were not uniform, causing the variation in tortilla weight. Weight is measured once at the beginning of each batch; these results suggest that more frequent measurements are needed.

Overall, the color difference increased when barley was used in the corn tortillas. However, tortillas with 2.5% barley flour substitution did not have significant changes in color (Table III). Barley flour significantly decreases L^* (lightness), b^* (yellowness) and increases a^* (redness), yielding darker, redder tortillas. Lower L^* values, higher a^* values and reduced b^* values were also found in corn tortillas (Mitre-Dieste 2001), in bread (Gordon 2001), and pastas (Knuckles et al. 1997, Marconi et al. 2000), when substituting barley flour in the formulation.

TABLE IV
Measurements of Diameter Quotient, Thickness, Weight, Ph, and Color of Tortillas 4 hr After Production^a

Barley level (%)	Diameter Quotient	Thickness	Weight	pH	<i>L</i>*	<i>a</i>*	<i>b</i>*	Color difference
Control	1.07 ^a	1.62 ^a	23.5 ^a	5.2 ^a	87.8 ^a	12.6 ^c	65.0 ^{ab}	1.7 ^c
2.5	1.04 ^a	1.57 ^a	21.5 ^c	5.1 ^a	84.1 ^b	12.1 ^c	65.9 ^a	4.2 ^c
5.0	1.05 ^a	1.58 ^a	22.5 ^b	5.2 ^a	80.4 ^c	15.0 ^b	61.8 ^{bc}	8.7 ^b
10.0	1.05 ^a	1.57 ^a	22.8 ^{ab}	5.2 ^a	74.2 ^d	17.4 ^a	60.8 ^c	15.5 ^a
LSD($\alpha=0.05$)^b	0.03	0.08	0.84	0.13	3.54	1.53	3.88	4.10

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Texture Evaluation

Subjective Texture Measurements

Subjective texture measurements showed significant differences in fresh and stored tortillas (Table V). Differences among samples were detected after 1 day of storage. Tortillas with barley flour stored for 7 days at 4°C had significantly higher scores for rollability and pliability than corn tortilla control (Figs. 3 and 4). No differences were detected among barley flour substitution levels (Table V).

TABLE V
Effect of Barley Substitution on Pliability Score of Corn Tortillas Stored at 4°C^a

Barley substitution level (%)	Pliability			Rollability		
	4 h	1 d	7 d	4 h	1 d	7 d
Control	5.0 ^a	3.4 ^c	2.8 ^b	5.0 ^a	4.3 ^b	3.3 ^b
2.5	5.0 ^a	4.3 ^a	3.8 ^a	5.0 ^a	4.8 ^a	4.3 ^a
5.0	4.9 ^a	4.0 ^{ab}	3.8 ^a	4.9 ^a	4.8 ^a	4.4 ^a
10.0	4.9 ^a	3.6 ^{bc}	3.5 ^a	4.8 ^a	4.8 ^a	4.4 ^a
LSD($\alpha=0.05$)^b	0.13	0.41	0.30	0.21	0.18	0.41
Mean	4.96	3.82	3.44	4.91	4.68	4.11
LSD_{time}($\alpha=0.05$)	0.19			0.17		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Objective Texture Evaluation

Rupture force, rupture distance and Young's modulus significantly changed during storage (Appendix A.1). Staling rate is higher initially; then it decreases but continues during storage (Miranda-Lopez 1999). Samples with barley flour showed differences even at the initial measurements (fresh tortillas, 4 hr after production). Rupture force was always significantly higher for the control than for the barley treatments (Appendix A.1, Fig. 5).

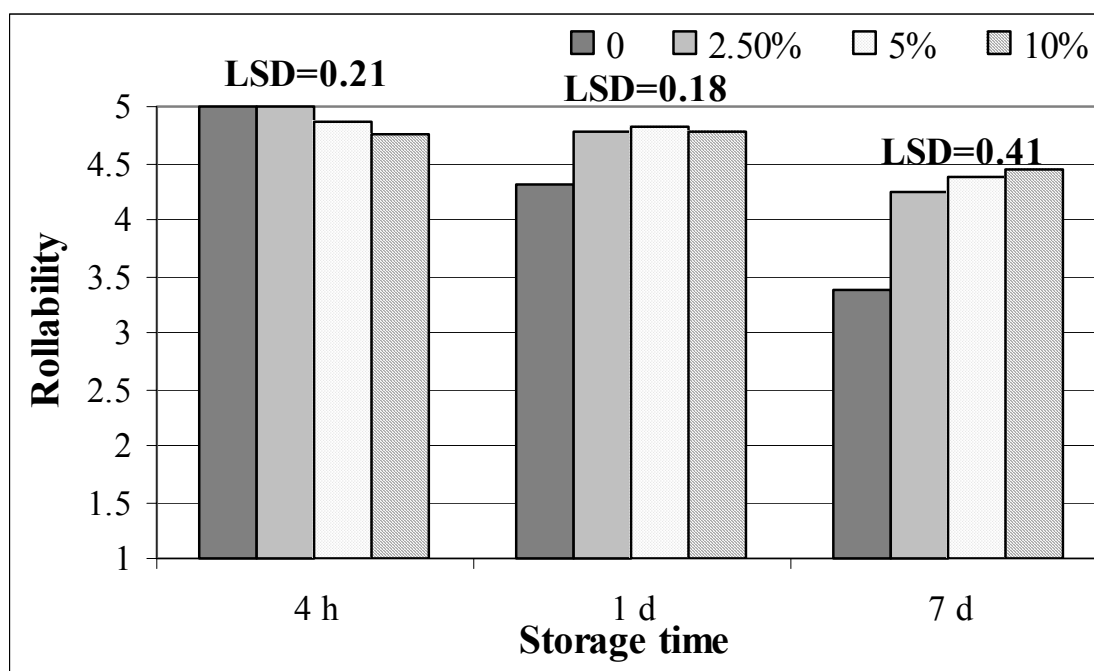


Fig. 3. Effect of Barley Flour Substitution on Rollability of Corn Tortillas Stored at 4°C.

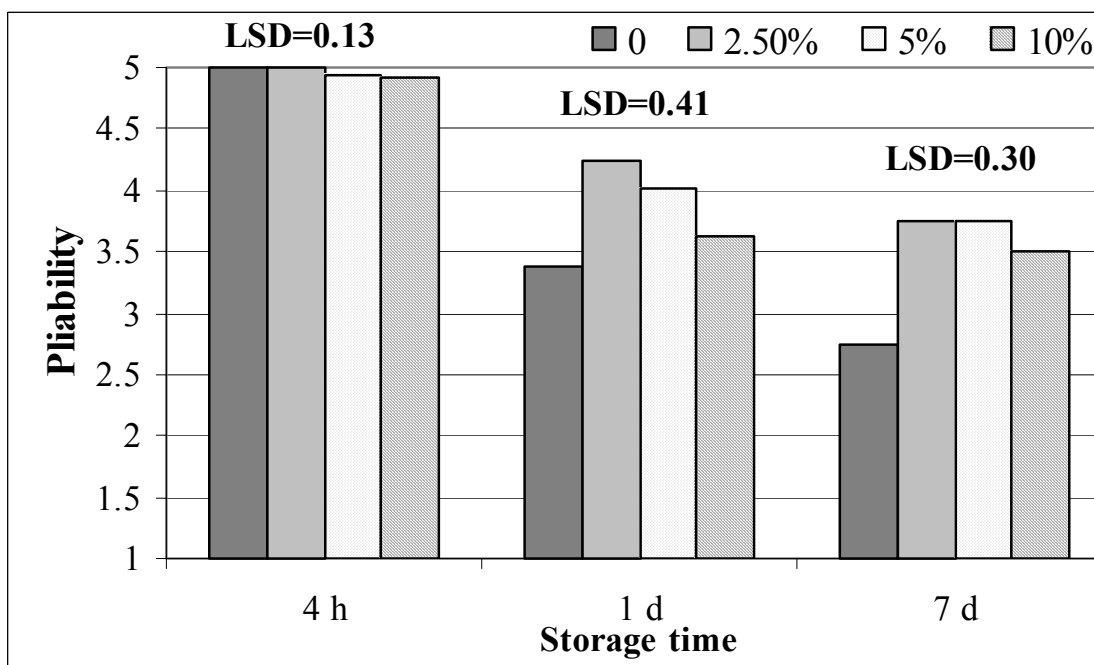


Fig. 4. Effect of Barley Flour Substitution on Pliability of Corn Tortillas Stored at 4°C.

Tortillas containing 5% barley flour had the highest rupture distance for the initial and first day measurements (Appendix A.1, Fig. 6). No difference among treatments was seen at 7 days, where tortilla is already stale. Barley flour or its components (i.e. β -glucans) did not contribute to the elasticity of the sample, but it provided a softer texture, as shown by a decreased rupture force.

The Young's modulus or modulus of elasticity is a parameter that measures the slope of rupture force vs. time. This value relates the force required to deform the sample with the deformation extent (Steffe 1996). Usually, larger values of Young's modulus are associated to a more brittle material. Young's modulus was significantly higher for the control than for barley treatments at all times (Appendix A.1, Fig. 7). However, tortillas with different barley levels were not significantly different from each other.

Mitre-Dieste (2001) reported that tortillas made from 100% barley flour tortillas from shorts had the lowest rupture force when compared to other barley flours and to a 100% dry masa flour tortillas after storage at 4°C for 9 days. Also, Gordon (2001) reported that breads containing 10-25% barley flour were actually softer/ less firm than control at 2 h. The use of 100% barley flour to produce muffins also showed increased penetrometer distance when compared to a wheat control (Newman et al. 1990), which is translated into a softer product when using barley flour.

Selection of Optimum Level of Barley Flour

Tortillas with 2.5% barley flour significantly improved tortilla quality without any negative effects. Thickness, pH and diameter quotient of corn tortillas were not affected by barley flour substitution. Tortillas with 2.5% barley flour substitution did not have significant changes in color and at 7 days of storage moisture content was not significantly different from control. The substitution of barley flour for dry masa flour (DMF) gave tortillas with significantly better texture than control after 7 days of storage at 4°C as it increased rollability and pliability while reducing rupture force and Young's modulus. Texture of tortillas with different barley flour levels were not significantly

different from each other. Therefore, the level of 2.5% of barley flour substitution was chosen as the optimum level for the next experiments.

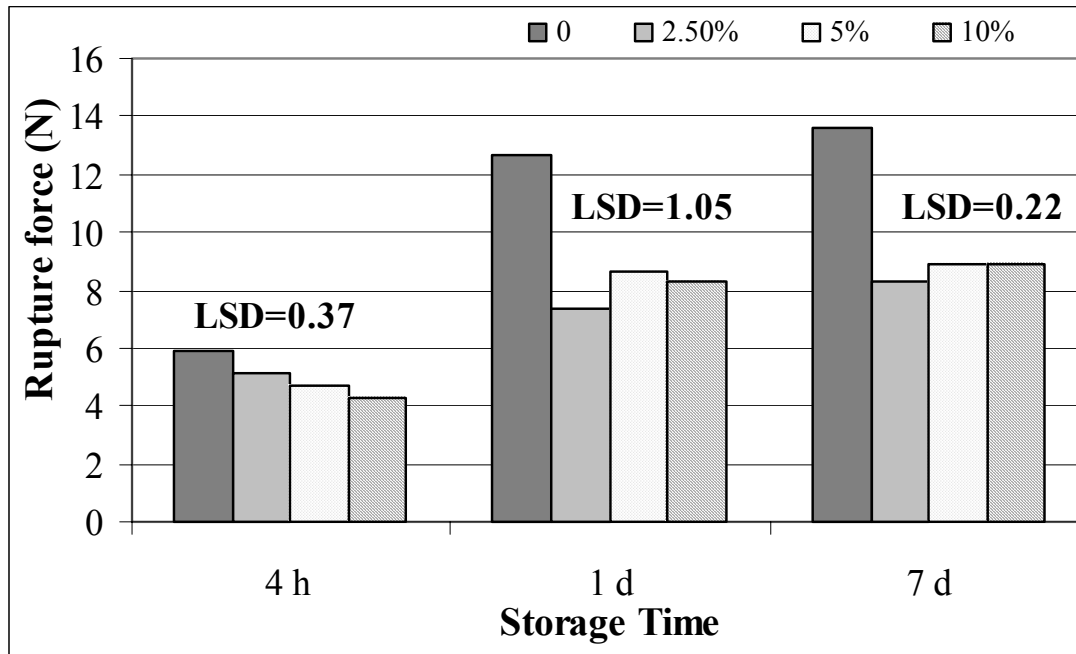


Fig. 5. Effect of Barley Flour Substitution on Rupture Force During Storage of Corn Tortillas at 4°C.

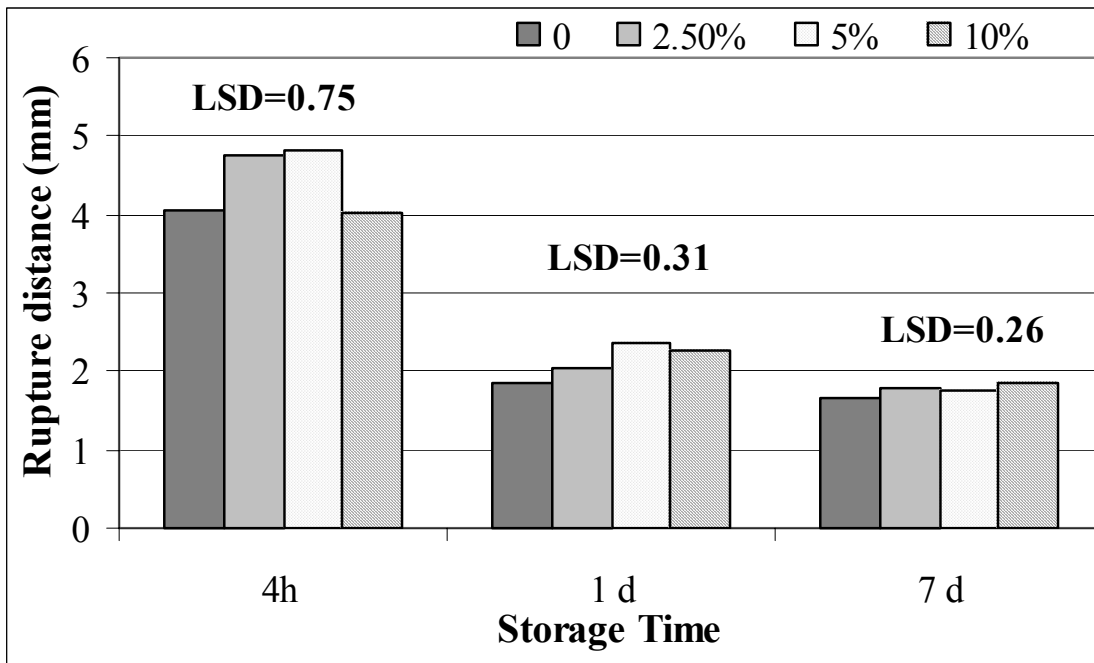


Fig. 6. Effect of Barley Flour Substitution on Rupture Distance During Storage of Corn Tortillas at 4°C.

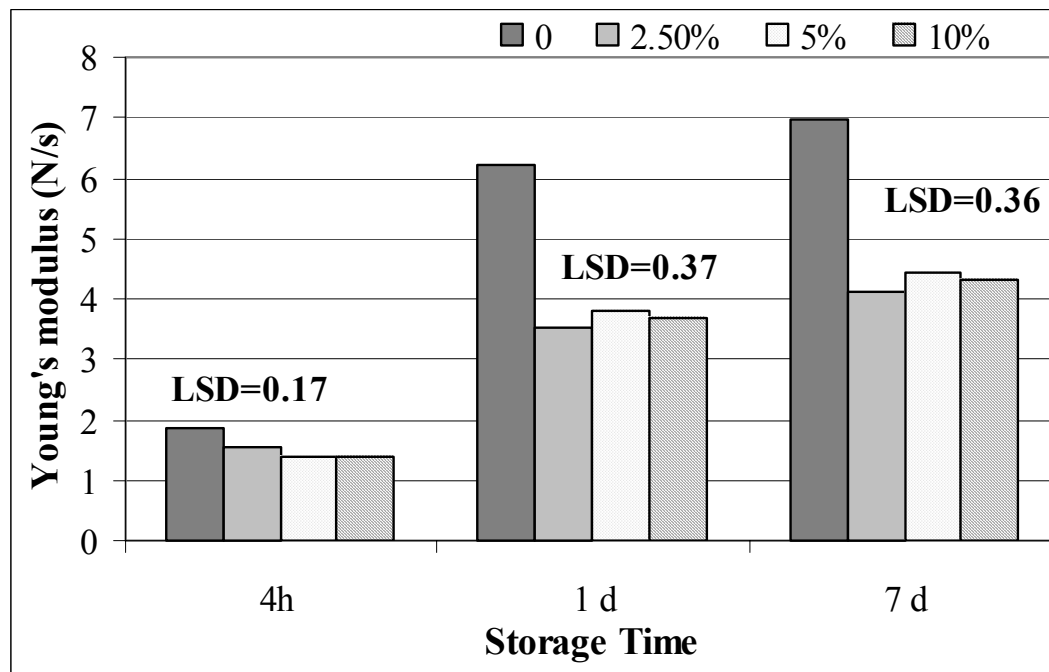


Fig. 7. Effect of Barley Flour Substitution on Young's Modulus During Storage of Corn Tortillas at 4°C.

CHAPTER V
COMBINED EFFECTS OF BARLEY FLOUR, AMYLASE AND CMC USING
RESPONSE SURFACE METHODOLOGY:
RESULTS AND DISCUSSION

The objective of this experiment was to evaluate the effects of combinations of additives in the corn tortilla formulation to optimize staling prevention. The anti-staling additives used were dusted barley flour from shorts, maltogenic amylase and carboxymethylcellulose (CMC).

Barley flour substitution improves tortilla texture quality by decreasing rupture force and Young's modulus while increasing rollability and pliability scores. DMF was substituted with 0-2.5% barley flour (Chapter IV). Canadian dusted barley flour from shorts with 9.6% β -glucan, 37% amylose and 22.3% dietary fiber was used.

Maltogenic amylase was used in levels between 0 to 1650 maltogenic amylase units (MAU). Bueso-Ucles 2003) found amylase combined with CMC reduced staling by preventing intra-granular re-crystallization of amylopectin in tortillas during storage. Carboxymethylcellulose (CMC) has consistently improved tortilla extensibility and subjective rollability in levels from 0 to 0.5% which may be related to its ability to create a flexible amorphous matrix in the continuous phase of tortillas (Bueso-Ucles 2003, Serna-Saldivar 1990, Suhendro 1997).

Response surface methodology in a central composite design was used to evaluate the effects of combinations of these additives. Table VI shows the treatments used in this experiment. Note that a 12th treatment with no additives was the corn tortilla control.

Water absorption was also adjusted to 115%, as discussed in Chapter IV, for all treatments containing barley flour to obtain masa with good machinability.

TABLE VI
Treatment Combinations Used for Evaluating Anti-staling Properties of Barley Flour with CMC and/or Maltogenic Amylase in a Central Composite Design

Barley flour (%)	CMC (%)	Amylase (MAU)
0	0.25	825
2.50	0.25	825
1.25	0	825
1.25	0.50	825
1.25	0.25	0
1.25	0.25	1650
0	0	1650
0	0.50	0
2.50	0	0
2.50	0.50	1650
1.25	0.25	825
0	0	0

Masa and Tortilla Moisture, Tortilla pH, Thickness, and Weight, and Batch Yield

The masa moisture was affected by the additive combinations of barley flour, amylase and CMC (Table VII). Fernandez et al. (1999) reported that masa usually has 58% moisture content. All masas were machinable. Changes in moisture content may be due to the water absorption differences in the different samples with barley flour. As barley flour was added, masa and tortilla moisture decreased. As found in Chapter IV, barley flour was responsible for the moisture decrease.

Tortilla pH was not significantly affected by the additive combinations (Table VII). The low pH is due to the use of an acidulant, fumaric acid, which activates the preservatives in the corn tortilla formulation.

TABLE VII
Effect of Additive Combinations on Masa and Tortilla Moisture and pH^a

Amylase (MAU)	Barley (%)	CMC (%)	Masa moisture (%)	Tortilla moisture (%)	Tortilla pH
0	0	0	59.6 ^{ab}	49.0 ^a	4.6 ^a
825	0	0.25	59.7 ^{ab}	48.9 ^a	4.6 ^a
825	2.5	0.25	58.7 ^d	47.6 ^d	4.6 ^a
825	1.25	0	58.8 ^{cd}	48.0 ^{bcd}	4.6 ^a
825	1.25	0.5	58.6 ^d	47.6 ^d	4.6 ^a
0	1.25	0.25	58.6 ^d	47.5 ^d	4.6 ^a
1650	1.25	0.25	58.9 ^{cd}	48.6 ^{ab}	4.6 ^a
1650	0	0	59.7 ^a	48.7 ^{ab}	4.6 ^a
0	0	0.5	59.8 ^a	49.1 ^a	4.6 ^a
0	2.5	0	58.6 ^d	47.8 ^{cd}	4.6 ^a
1650	2.5	0.5	59.0 ^{cd}	47.6 ^d	4.6 ^a
825	1.25	0.25	59.2 ^{bc}	48.5 ^{abc}	4.6 ^a
LSD ($\alpha=0.05$)^b			0.48	0.77	0.08

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Batch yields were not affected by additives (Table VIII). However, the tortilla weight and thickness was affected by treatment combinations (Table VIII). Tortilla thickness was affected by puffing. Also, the tortilla weight changes may be attributed to this phenomenon, since puffing is caused by steam escaping from the tortilla. Tortillas with CMC and/or barley flour had increased weight with decreased thickness. When puffing takes place, moisture is lost in form of steam and causes thickness to increase and weight to decrease. Tortillas with 1650 MAU did not puff as much as the other treatments because no hydrocolloids were in the formulation. Yau (1994) reported that tortillas with hydrocolloids (0.5 or 1% CMC, hydroxypropylmethylcellulose or xanthan gum) increased puffing of tortillas.

TABLE VIII
Effect of Additive Combinations on Batch Yield, Tortilla Weight and Tortilla Thickness^a

Amylase (MAU)	Barley (%)	CMC (%)	Yield (kg tortilla/kg flour)	Tortilla weight (g)	Tortilla thickness (mm)
0	0	0	0.8 ^a	23.5 ^b	1.62 ^{ab}
825	0	0.25	0.9 ^a	23.5 ^{ab}	1.60 ^{abc}
825	2.5	0.25	0.8 ^a	23.8 ^{ab}	1.54 ^{abc}
825	1.25	0	1.1 ^a	23.5 ^b	1.59 ^{abc}
825	1.25	0.5	1.1 ^a	23.3 ^b	1.65 ^a
0	1.25	0.25	1.0 ^a	24.0 ^{ab}	1.61 ^{ab}
1650	1.25	0.25	1.0 ^a	23.9 ^{ab}	1.58 ^{abc}
1650	0	0	0.9 ^a	23.5 ^{ab}	1.47 ^c
0	0	0.5	1.1 ^a	24.8 ^a	1.51 ^{bc}
0	2.5	0	0.9 ^a	24.3 ^{ab}	1.59 ^{abc}
1650	2.5	0.5	0.9 ^a	23.7 ^{ab}	1.63 ^{ab}
825	1.25	0.25	0.9 ^a	23.9 ^{ab}	1.55 ^{abc}
LSD ($\alpha=0.05$)^b			0.37	1.28	0.13

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Texture Evaluation

Subjective Texture Measurements

Rollability and pliability scores significantly changed at each storage time measured (Appendix B.1 and B.2). No differences were detected by rollability or pliability among treatments 4 hr after production. Fresh tortillas were soft, flexible and rollable after 1h equilibration (Fernandez et al. 1999). After 1 day of storage, subjective measurements detected changes in tortilla structure and differences among treatments.

As tortilla is stored, staling takes place and products become firmer (Quintero-Fuentes 1999). At all storage times, the control (no additives) had significantly undesirable, lower scores for subjective texture measurements ($p < 0.05$).

The use of barley flour did not significantly improve rollability score in stored tortillas (Fig. 8). Amylase improved rollability when used up to 825 MAU and decreased it above this level (Appendix Fig.B.1). However, when levels higher than 825 MAU

were combined with more than 1% barley flour, a synergistic effect gave tortillas with the higher rollability scores.

CMC improved rollability in corn tortillas (Serna-Saldivar 1990, Suhendro 1997, Bueso-Ucles 2003). The combination of the barley flour and CMC did not improve tortilla rollability (Fig. 9). CMC was most effective at increasing rollability in stored tortillas. However, when using 825 MAU, a noticeable synergy was found between amylase, barley and CMC. At levels higher than 825 MAU, barley flour showed a strong effect in rollability (Appendix Fig. B.1).

Barley flour, amylase and CMC improved pliability scores. Barley flour is able to improve pliability scores when combined with amylase or CMC (Fig. 10 and 11, Appendix B2). Levels higher than 0.2% CMC and up to 825 MAU alone significantly increased pliability compared to control after 14 days of storage at 4°C (Bueso-Ucles 2003). In contrast to the rollability results, pliability scores showed a synergy between barley flour and CMC as well as between barley flour and amylase. Pliability is a subjective measurement that detects staling faster than rollability (Bueso-Ucles 2003). The best response was seen in tortillas with barley flour and CMC at 825 MAU (Appendix Fig. B.2). More than 825 MAU decreased pliability.

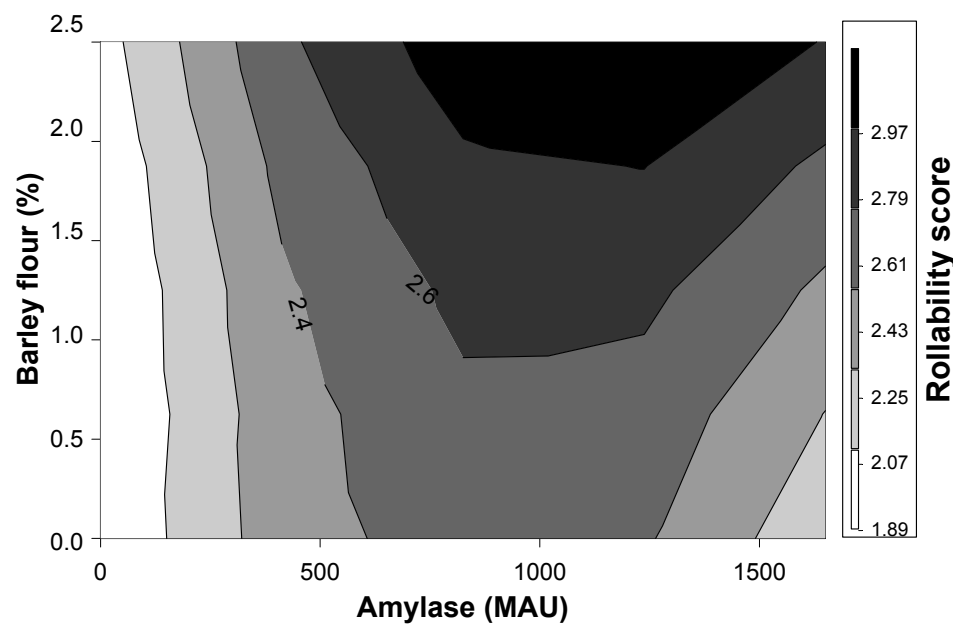


Fig. 8. Effect of Amylase and Barley Flour on Rollability of Corn Tortillas Stored at 4°C for 14 Days. Note: CMC=0; $LSD(\alpha=0.05)=0.60$.

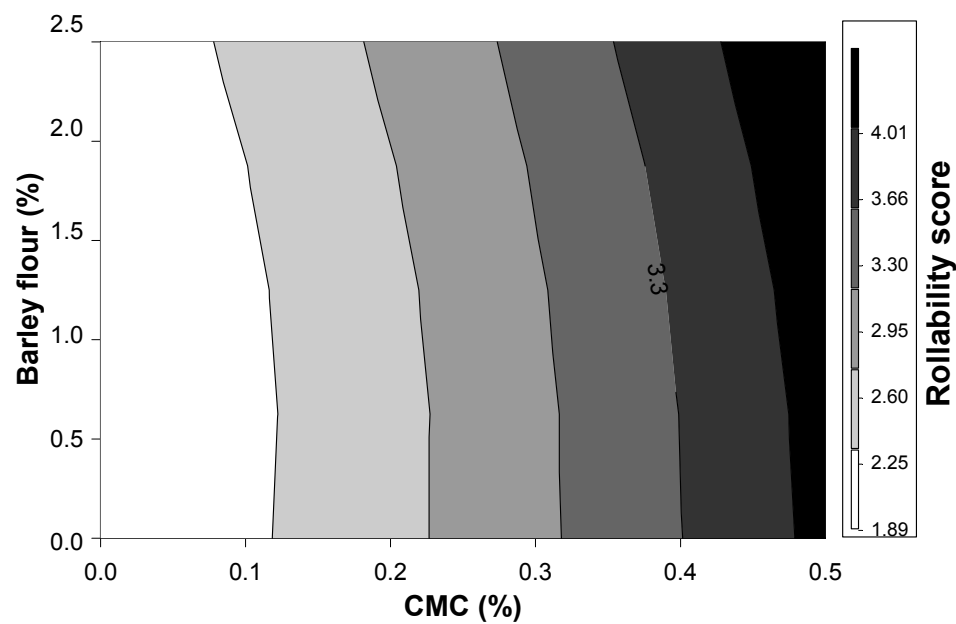


Fig. 9. Effect of CMC and Barley Flour on Rollability of Corn Tortillas Stored at 4°C for 14 Days. Note: Amylase=0; $LSD(\alpha=0.05)=0.60$.

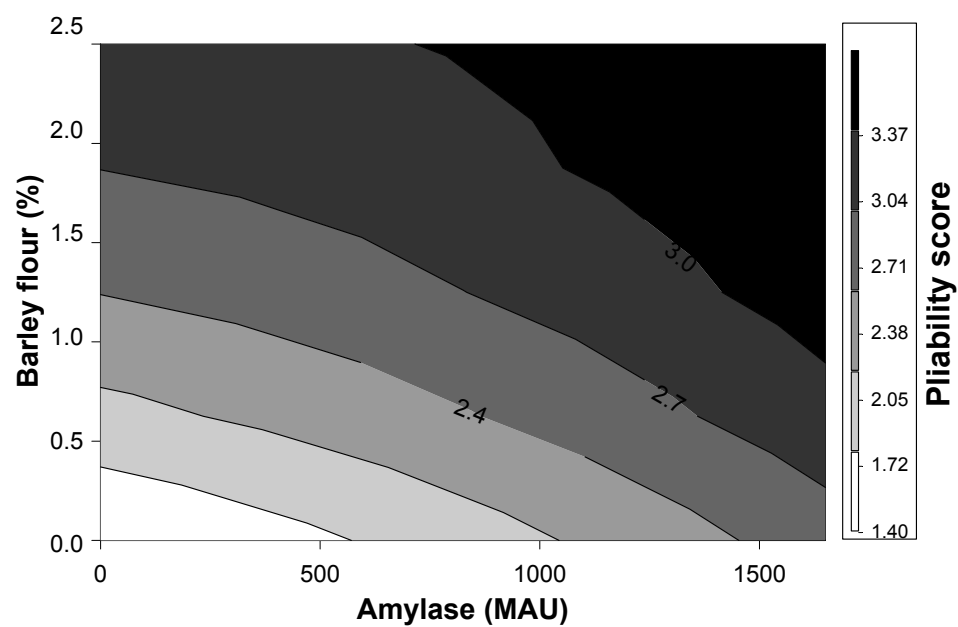


Fig. 10. Effect of Amylase and Barley Flour on Pliability of Corn Tortillas Stored at 4°C for 14 Days. Note: CMC=0; $LSD(\alpha=0.05)=0.52$.

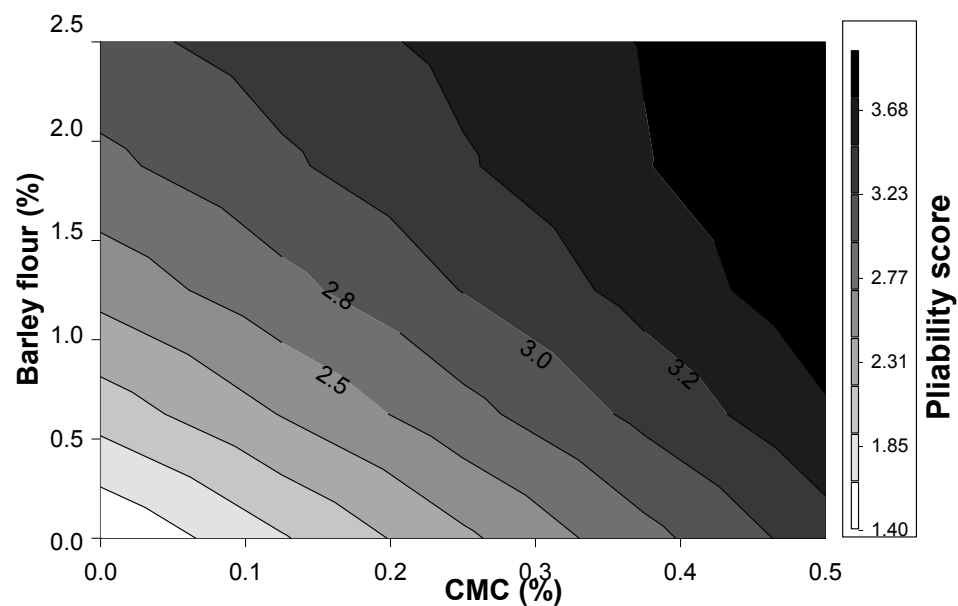


Fig. 11. Effect of CMC and Barley Flour on Pliability of Corn Tortillas Stored at 4°C for 14 Days. Note: Amylase=0; $LSD(\alpha=0.05)=0.52$.

Objective Texture Measurements

During storage, changes in tortillas were seen for all parameters (Appendix B.3, B.4, B.5, B.6, B.7). Fresh tortillas had the lowest rupture force, Young's modulus and final stiffness. Rupture force and Young's modulus increased after one day and did not change after 7 days. However, these values increased from 7 to 14 days of storage, as well as final stiffness. Fresh tortillas had the highest rupture distance and energy dissipated. Rupture distance decreased significantly after 1 day and did not change after 14 days of storage. Energy dissipated was reduced after 7 days and was not further decreased. At 4 hr after production, tortilla additives already showed significant differences in rupture force, rupture distance, Young's modulus, final stiffness and energy dissipated.

Barley flour did not affect rupture force at 14 days of storage while amylase significantly reduced it (Fig.12). Bueso-Ucles (2003) found that amylase significantly reduced rupture force in tortillas stored at 4°C for 14 days. Barley flour had a synergistic reduction in rupture force when combined with amylase above 825 MAU (Appendix Fig.B.3).

CMC did not significantly reduce rupture force in corn tortillas (Fig. 13). This supports the results reported by Bueso-Ucles (2003) where reductions in tortilla rupture force by the use of 0.25-0.5% CMC were not significant. However, the combination of these two additives showed a significantly reduced rupture force for tortillas containing additive over that of the control tortillas (no additives).

After 14 of storage, rupture distance was significantly reduced by the use of amylase (Fig. 14, Appendix Fig. B.4). Barley flour did not change rupture distance while CMC significantly increased the rupture distance (Fig. 15). This confirms previous studies (Bueso-Ucles 2003) where the use of CMC alone significantly improved rupture distance from levels of 0.25 to 0.5%. However, there is a synergistic, positive effect of barley flour combined with CMC. Lower levels than 0.5% CMC combined with barley flour can be used to improve tortilla extensibility.

Young's modulus was always significantly higher for control tortillas than the treatments (Appendix B.5). This parameter measures the force required to rupture the material compared to the strain (deformation) imposed on tortillas. Decreased Young's modulus values are related to large deformations, which are given by viscous materials that will extend before rupture. Barley flour did not affect Young's modulus (Fig. 16 and 17, Appendix B.5). CMC and 825 MAU significantly decreased Young's modulus in tortillas stored for 14 days (Appendix Fig. B.5).

Combinations of amylase and barley flour significantly decreased final stiffness (Fig. 18, Appendix Fig. B.6). CMC alone or combined with barley flour did not significantly reduce final stiffness of stored corn tortillas (Fig. 19).

Tortillas stored 7 days were not different from 14 day old tortillas. No additive significantly increased energy dissipated (Fig. 20 and 21, Appendix Fig. B.7). However, the combination of more than 1% barley flour with more than 825 MAU showed a synergistic effect increasing this value.

Selection of the Optimum Level

Several treatments had significantly improved texture of tortillas. The best treatments (higher rupture distance and pliability or lower rupture force and final stiffness) are shown in Table IX. These treatments were not significantly different among each other. Therefore, treatments with the highest additives levels were discarded to reduce the cost. The combination of 825 MAU, 1.25% barley flour and 0.25% CMC was the best treatment. Bueso-Ucles (2003) showed that a combination of 825 MAU with 0.25% CMC had improved texture properties. However, it was not better than the treatments shown in Table IX. This means that the use of barley significantly improved this combination.

TABLE IX
Optimum Treatments Evaluated for Different Texture Measurements

Amylase (MAU)	Barley (%)	CMC (%)
825	1.25	0.25
825	2.5	0.25
825	1.25	0.5
1650	2.5	0.5

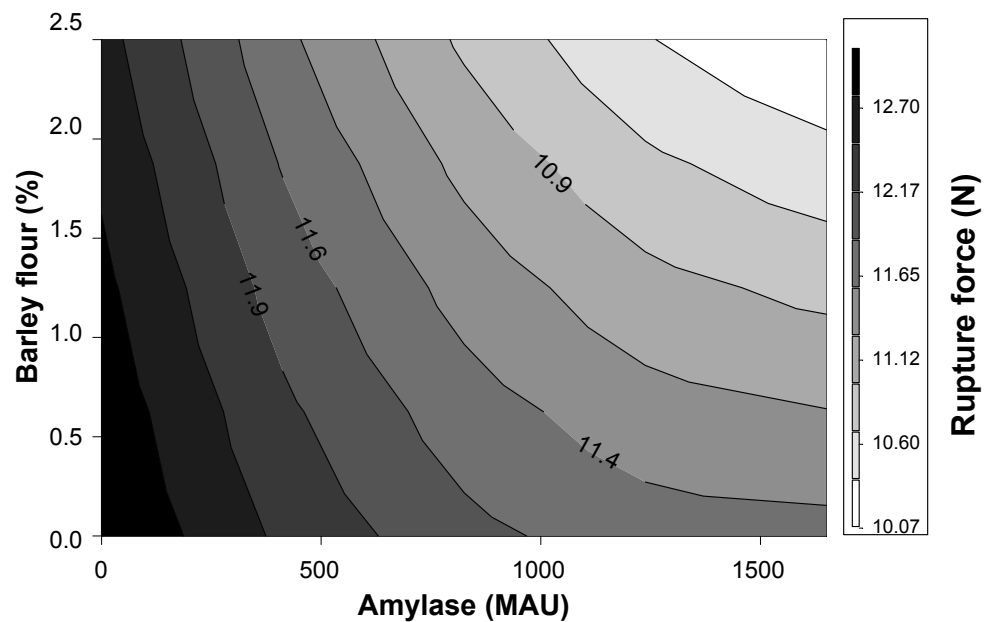


Fig. 12. Effect of Amylase and Barley Flour on Rupture Force (N) of Corn Tortillas Stored at 4°C for 14 Days. Note: CMC=0; LSD($\alpha=0.05$)=0.96.

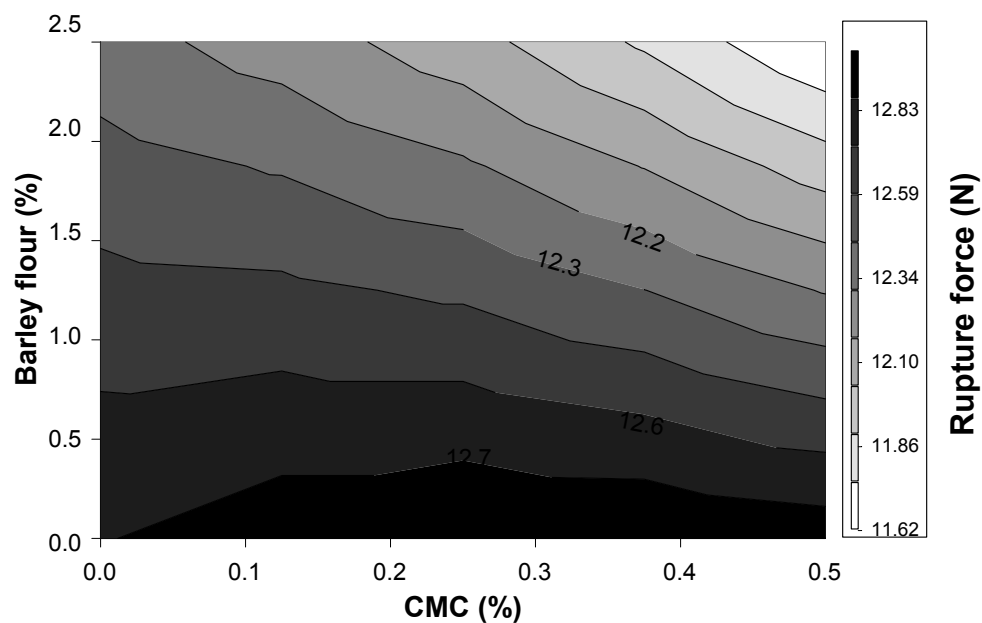


Fig. 13. Effect of CMC and Barley Flour on Rupture Force (N) of Corn Tortillas Stored at 4°C for 14 Days. Note: Amylase=0; $LSD(\alpha=0.05)=0.96$.

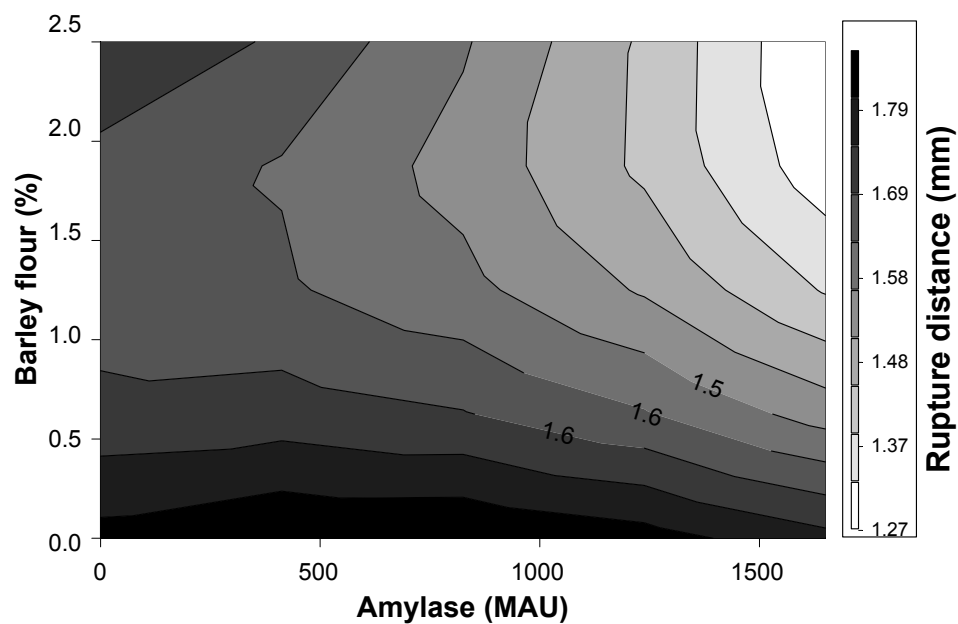


Fig. 14. Effect of Amylase and Barley Flour on Rupture Distance (mm) of Corn Tortillas Stored at 4°C for 14 Days. Note: CMC=0; $LSD(\alpha=0.05)=0.27$.

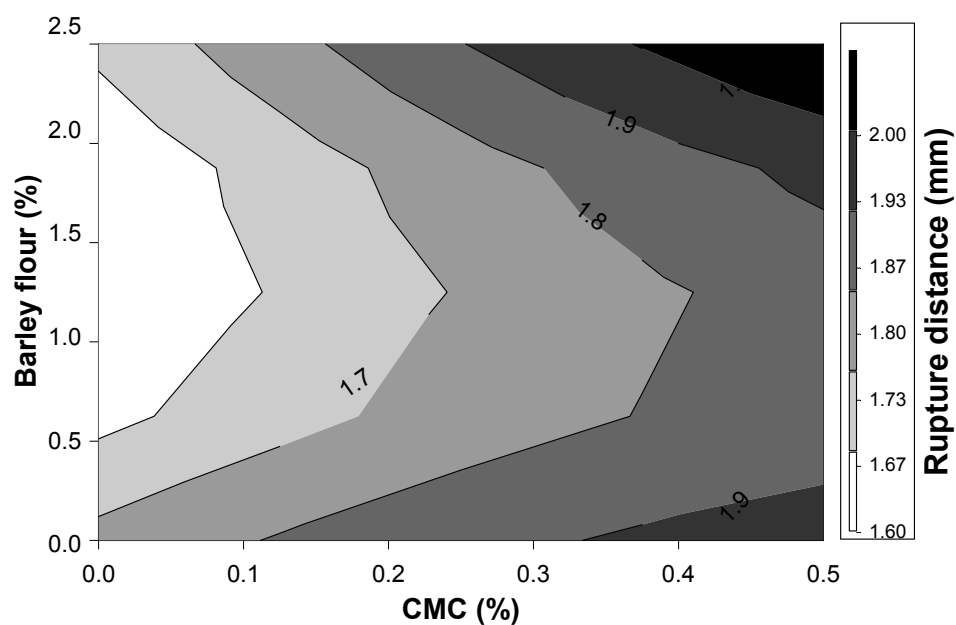


Fig. 15. Effect of CMC and Barley Flour on Rupture Distance (mm) of Corn Tortillas Stored at 4°C for 14 Days. Note: Amylase=0; $LSD(\alpha=0.05)=0.27$.

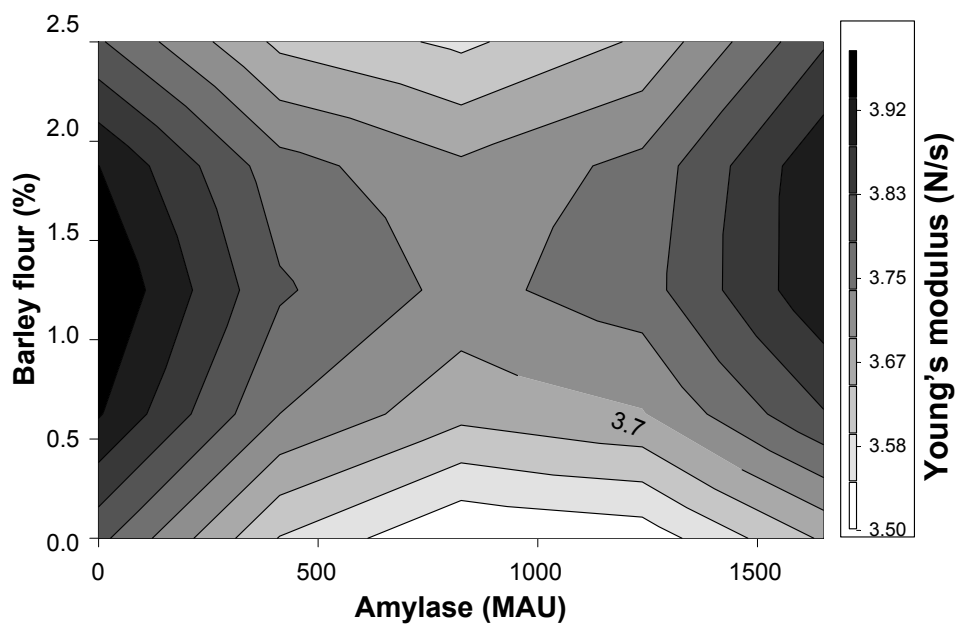


Fig. 16. Effect of Amylase and Barley Flour on Young's Modulus (N/s) of Corn Tortillas Stored at 4°C for 14 Days. Note: CMC=0; $LSD(\alpha=0.05)=0.33$.

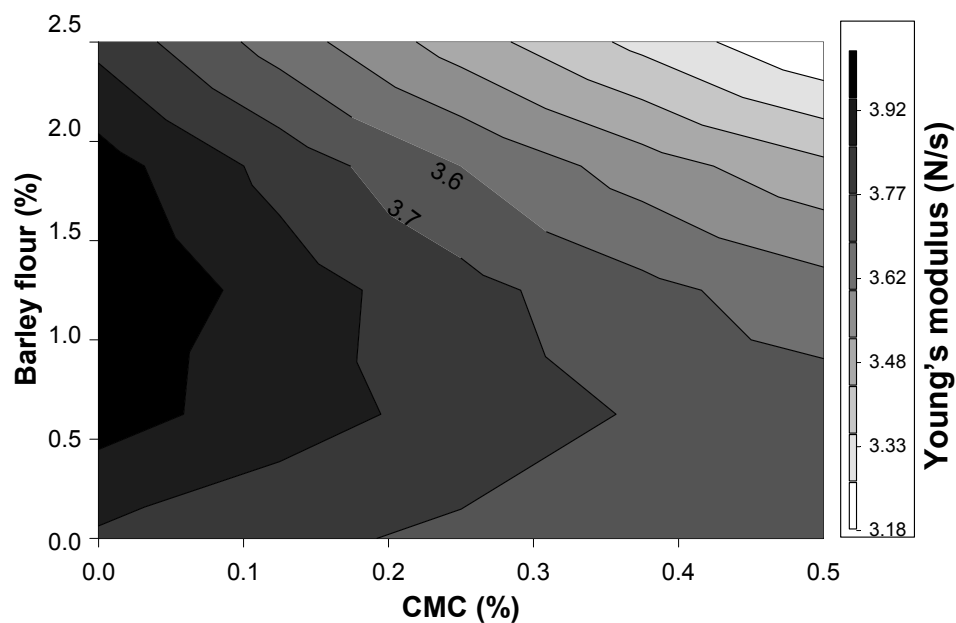


Fig. 17. Effect of CMC and Barley Flour on Young's Modulus (N/s) of Corn Tortillas Stored at 4°C for 14 Days. Note: Amylase=0; LSD($\alpha=0.05$)=0.33.

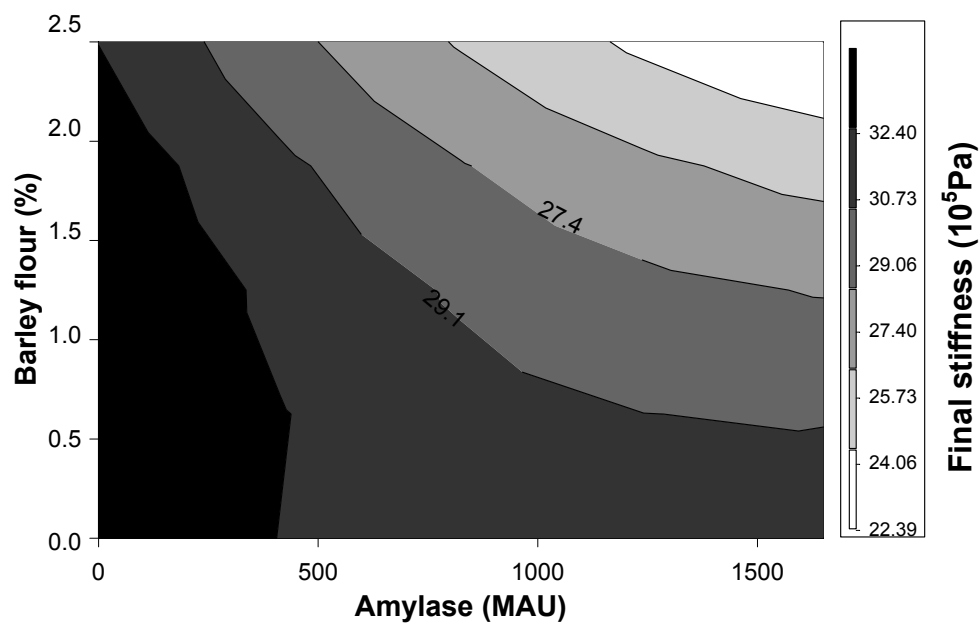


Fig. 18. Effect of Amylase and Barley Flour on Final Stiffness (10⁵ Pa) of Corn Tortillas Stored at 4°C for 14 Days. Note: CMC=0; LSD($\alpha=0.05$)=1.98.

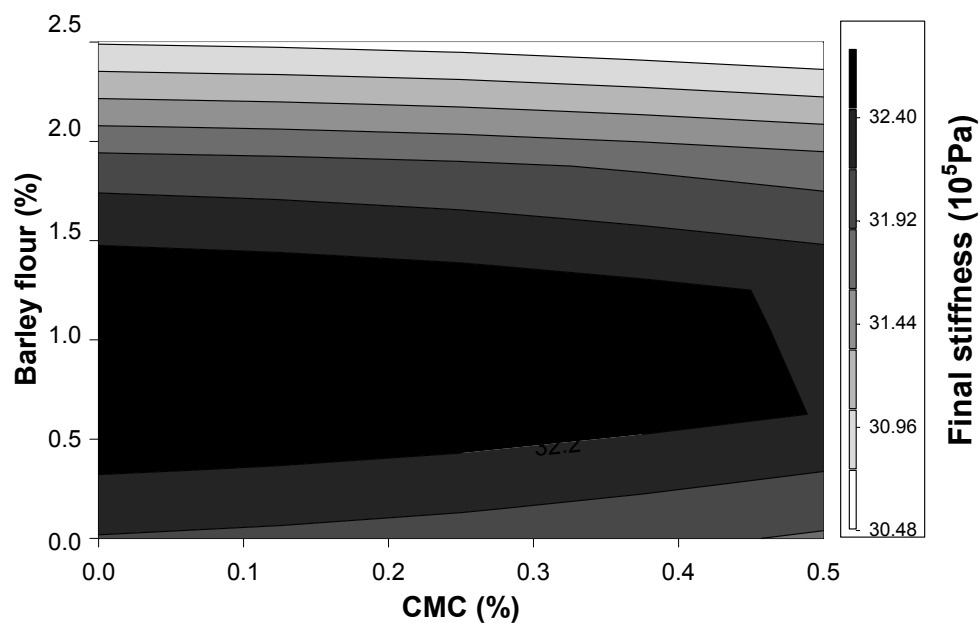


Fig. 19. Effect of CMC and Barley Flour on Final Stiffness (10^5 Pa) of Corn Tortillas Stored at 4°C for 14 Days. Note: Amylase=0; $\text{LSD}(\alpha=0.05)=1.98$.

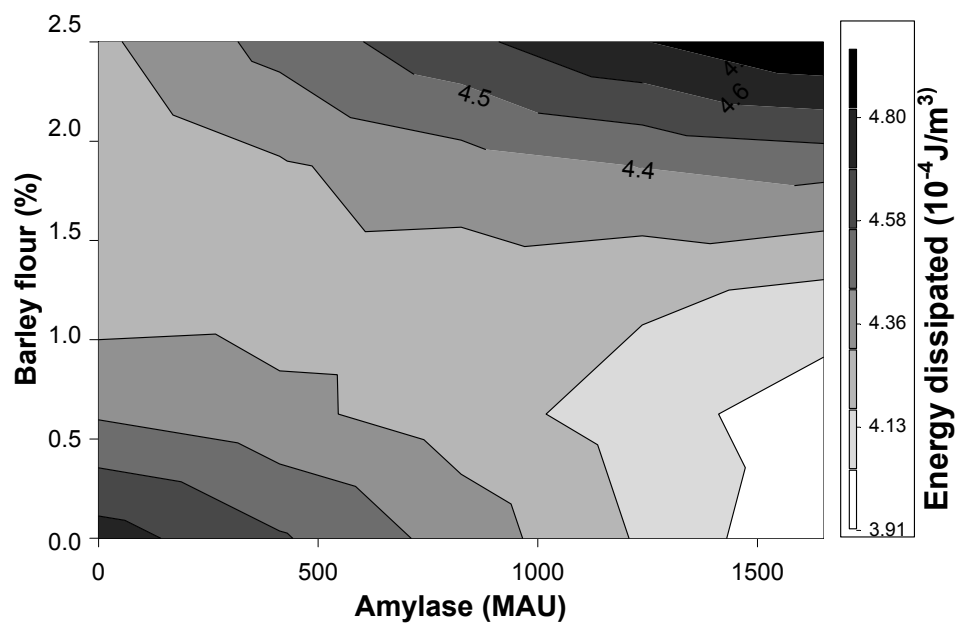


Fig. 20. Effect of Amylase and Barley Flour on Energy Dissipated (10^{-4} J/m³) of Corn Tortillas Stored at 4°C for 14 Days. Note: CMC=0; $\text{LSD}(\alpha=0.05)=0.74$.

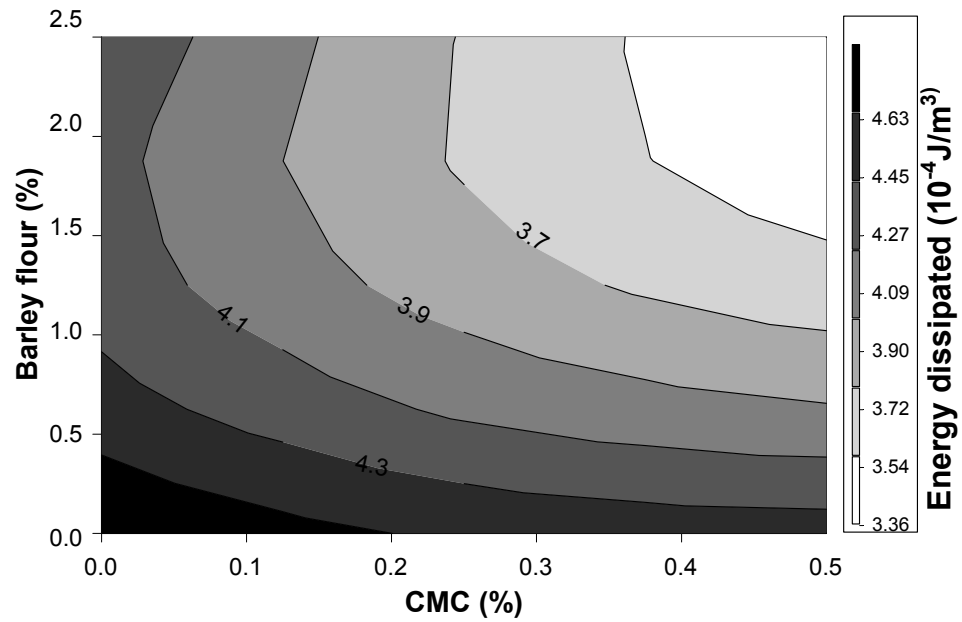


Fig. 21. Effect of CMC and Barley Flour on Energy Dissipated (10^{-4} J/M^3) of Corn Tortillas Stored at 4°C for 14 Days. Note: Amylase=0; $\text{LSD}(\alpha=0.05)=0.74$.

CHAPTER VI
USE OF BARLEY β -GLUCAN CONCENTRATE COMBINED WITH AMYLASE
TO IMPROVE CORN TORTILLA TEXTURE:
RESULTS AND DISCUSSION

The objective of this experiment was to evaluate the potential use of a β -glucan concentrate in combination with amylase to tenderize corn tortillas.

A barley β -glucan concentrate (70% purity) was substituted in corn tortillas at different levels equivalent to the β -glucan content of barley flour. An amylase level of 550 MAU was used with the barley, since this amount reduced rupture force and final stiffness and improved subjective texture measurements. A negative control (no additives) and a positive control (2.5% barley flour) were used. For comparison purposes, 0.5% CMC was also used, as well as a combination of 0.5% CMC with 550 MAU. The treatment combinations are shown in Table X.

TABLE X
Treatment Combinations for Barley β -Glucan Concentrate Evaluation

Barley flour (%)	Barley β -glucan (%)	Maltogenic amylase (MAU)	CMC (%)
0	0	0	0
2.5	0	0	0
0	0.12	0	0
0	0.12	550	0
0	0.24	0	0
0	0.24	550	0
0	0.96	0	0
0	0.96	550	0
0	2.4	0	0
0	2.4	550	0
0	0	0	0.5
0	0	550	0.5

Masa and Tortilla Moisture, Tortilla pH, Thickness, Weight and Batch Yield

Differences in masa and tortilla moisture are shown in Table XI. Masa with 2.4% β -glucan and 0.96-2.4% β -glucan with amylase had significantly lower masa moisture content than control. No treatment was significantly different from control for tortilla moisture or pH ($p < 0.05$). The use of additives did not significantly ($p < 0.05$) affect batch yield, tortilla weight and tortilla thickness compared to the control (Table XII).

TABLE XI
Effect of Additive Combinations on Masa and Tortilla Moisture and pH^a

Treatment	Masa moisture	Tortilla moisture	pH
DMF Control	53.7 ^a	43.4 ^{ab}	4.7 ^{ab}
Barley Flour Control	52.9 ^a	44.6 ^{ab}	4.7 ^{ab}
0.12% β -glucan	52.6 ^{ab}	44.7 ^a	4.6 ^{ab}
0.24% β -glucan	53.4 ^a	41.3 ^{ab}	4.6 ^{ab}
0.96% β -glucan	52.5 ^{abc}	42.1 ^{ab}	4.6 ^b
2.4% β -glucan	51.4 ^{bc}	45.0 ^a	4.7 ^{ab}
0.5% CMC	53.6 ^a	43.1 ^{ab}	4.7 ^a
With 550 MAU amylase			
0.12% β -glucan	53.9 ^a	40.7 ^b	4.6 ^b
0.24% β -glucan	53.9 ^a	44.6 ^{ab}	4.6 ^{ab}
0.96% β -glucan	51.0 ^c	43.7 ^{ab}	4.7 ^{ab}
2.4% β -glucan	51.3 ^{bc}	42.1 ^{ab}	4.6 ^{ab}
0.5% CMC	53.7 ^a	44.2 ^{ab}	4.7 ^a
LSD ($\alpha=0.05$)^b	1.50	3.93	0.15

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE XII
Effect of Additive Combinations on Batch Yield, Tortilla Weight and Thickness^a

Treatment	Batch Yield (kg/kg DMF)	Tortilla weight (g)	Tortilla thickness (mm)
DMF Control	1.1 ^{abc}	23.6 ^{abcd}	1.56 ^{ab}
Barley Flour Control	1.2 ^{ab}	23.5 ^{abcd}	1.59 ^{ab}
0.12% β -glucan	1.1 ^{abc}	24.4 ^{abc}	1.53 ^{ab}
0.24% β -glucan	1.1 ^{abc}	23.7 ^{abcd}	1.50 ^{ab}
0.96% β -glucan	1.3 ^{ab}	23.5 ^{bcd}	1.42 ^{ab}
2.4% β -glucan	1.2 ^{ab}	24.7 ^a	1.60 ^{ab}
0.5% CMC	1.0 ^{bc}	23.1 ^d	1.54 ^{ab}
With 550 MAU amylase			
0.12% β -glucan	0.9 ^c	23.5 ^{bcd}	1.68 ^a
0.24% β -glucan	1.1 ^{bc}	23.5 ^{bcd}	1.63 ^{ab}
0.96% β -glucan	1.3 ^{ab}	24.0 ^{abcd}	1.32 ^b
2.4% β -glucan	1.3 ^a	24.7 ^{ab}	1.60 ^{ab}
0.5% CMC	1.1 ^{abc}	23.4 ^{cd}	1.59 ^{ab}
LSD ($\alpha=0.05$)^b	0.27	1.21	0.33

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Texture Evaluation

Subjective Texture Measurements

Storage time significantly affected rollability and pliability (Appendix C.1 and C.2). Throughout storage, as expected, these values decreased as tortillas stale.

The control tortillas had the lowest rollability and pliability scores after 14 days of storage (Fig. 22 and 23). The use of β -glucan concentrate significantly improved rollability and pliability. However, the use of 550 MAU with β -glucans did not contribute to this improvement.

Tortillas with β -glucan had comparable rollability and pliability scores to the use of CMC alone or combined with amylase. Barley flour control was not significantly different from dry masa flour control as measured by rollability. However, pliability did

show a significant increase. Again, pliability was more efficient in detecting tortilla texture changes subjectively. β -glucan levels from 0.12-2.4% significantly improved rollability and pliability scores.

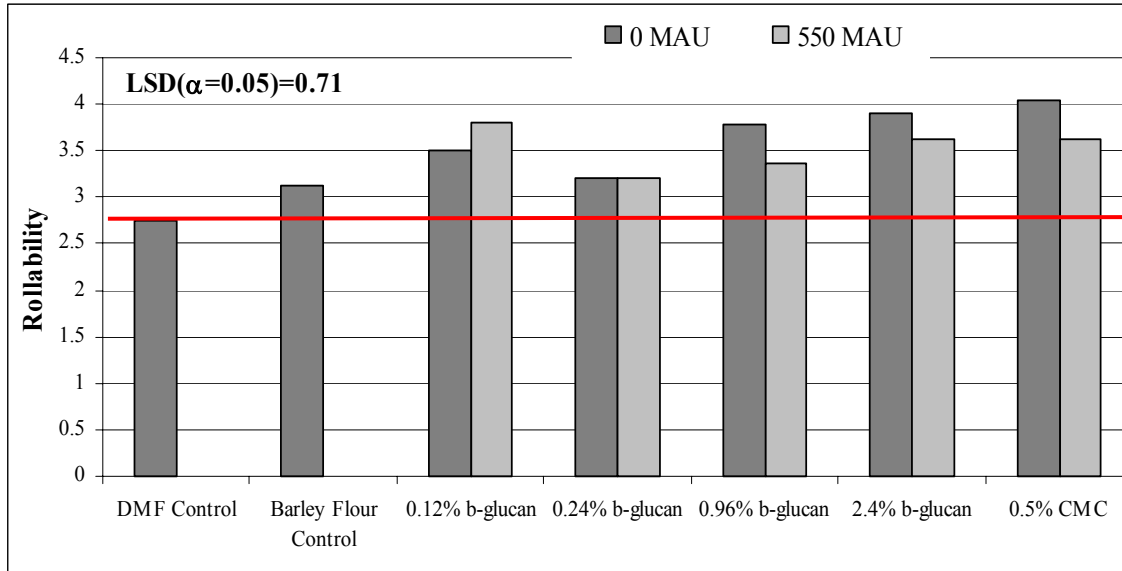


Fig. 22. Effect of β -glucan on Rollability of Corn Tortillas Stored for 14 Days at 4°C.

Objective Texture Measurements

All objective texture measurements were significantly affected by storage time and use of additives (Appendix C.3, C.4, C.5, C.6, C.7). All parameters were significantly different at different storage times except for energy dissipated where no changes were seen after 7 days.

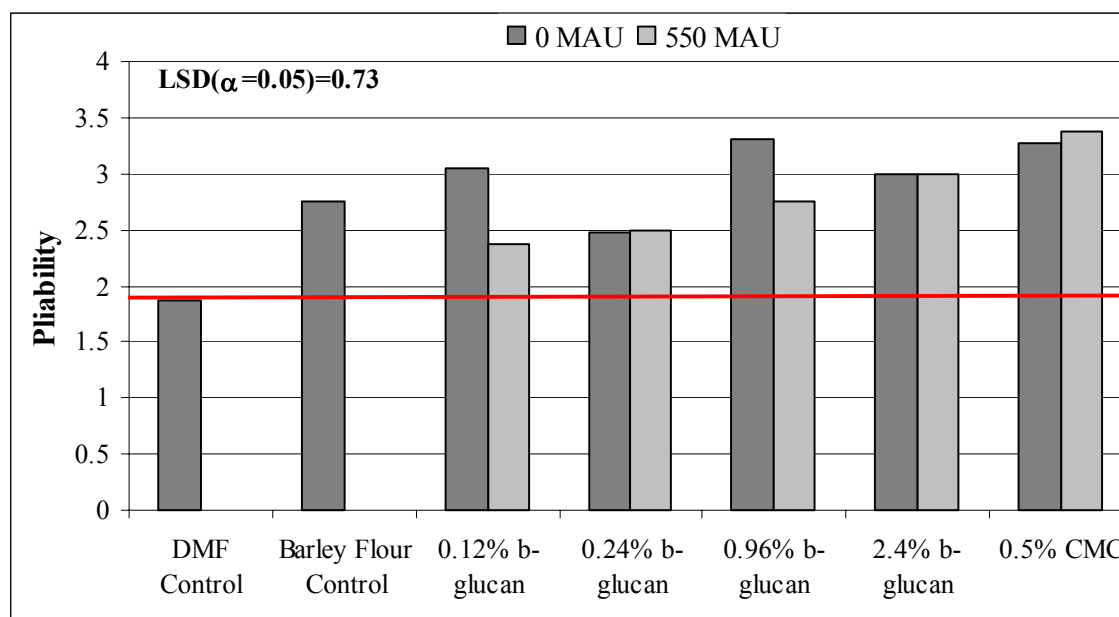


Fig. 23. Effect of β -glucan on Pliability of Corn Tortillas Stored for 14 Days at 4°C.

Amylase combination with β -glucan did not affect the initial rupture force (Appendix C.3). However, between 1 and 7 days, amylase treatments had lower values for rupture force. Amylase did not affect rupture force of tortillas stored for 14 days (Fig. 24). Rupture force significantly increased as level of β -glucan increased from 0.96 to 2.4% compared to control tortillas stored for 14 days at 4°C.

Barley flour control had the lowest rupture force which was the only treatment significantly different from control, confirming results from Chapter IV. This was probably because the β -glucan present in the flour is “diluted” and it did not cause an increase in rupture force. This is also related to the gelatinization temperature of barley and corn starch. Barley starch gelatinization temperature range is 51-60°C, while corn starch gelatinization temperature range is between 62 and 72°C (Hoseney 1994). Retrogradation is related to the extent of gelatinization (Seetharaman 2002), and considering that barley starch has a lower gelatinization temperature, it gelatinizes further and faster than corn, making it less susceptible to retrogradation since amylose

and amylopectin of the starch granule are greatly dispersed and more time is needed for polymer aggregation. Therefore, there is a significant difference at the end of storage in tortillas with barley flour than with the β -glucan concentrate or CMC with or without amylase.

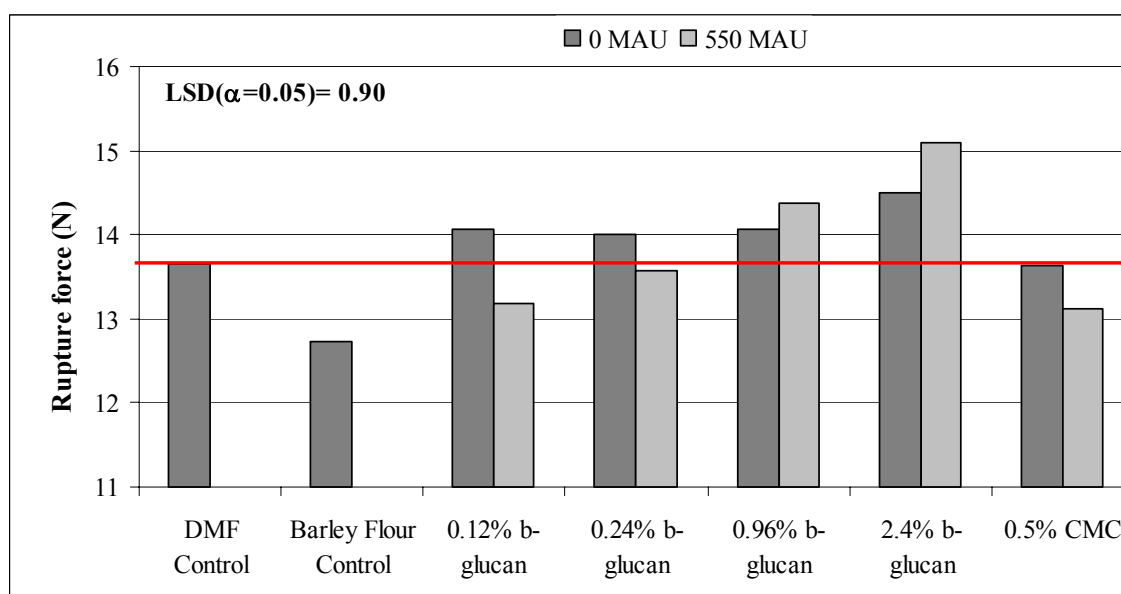


Fig. 24. Effect of β -glucan on Rupture Force (N) of Corn Tortillas Stored for 14 Days at 4°C.

Rupture distance of control tortillas was not significantly different from any of the treatments (Appendix C.4). At the initial measurements, rupture distance was significantly higher than control in tortillas with 0.12% β -glucan or 0.5% CMC with and without amylase. Tortillas containing amylase were no different than control tortillas after 1 day of storage, showing that amylase had weakened the tortilla structure. Additives did not significantly change rupture distance in tortillas after 4 days storage. After 7 days, 0.96% β -glucan was significantly higher than control. However, at 14 days there was no significant difference compared to control (Fig. 25). The use of amylase did not significantly decrease rupture distance. This confirms that the use of this amylase

level was appropriate because higher amounts negatively affected rupture distance in the previous study (Chapter V).

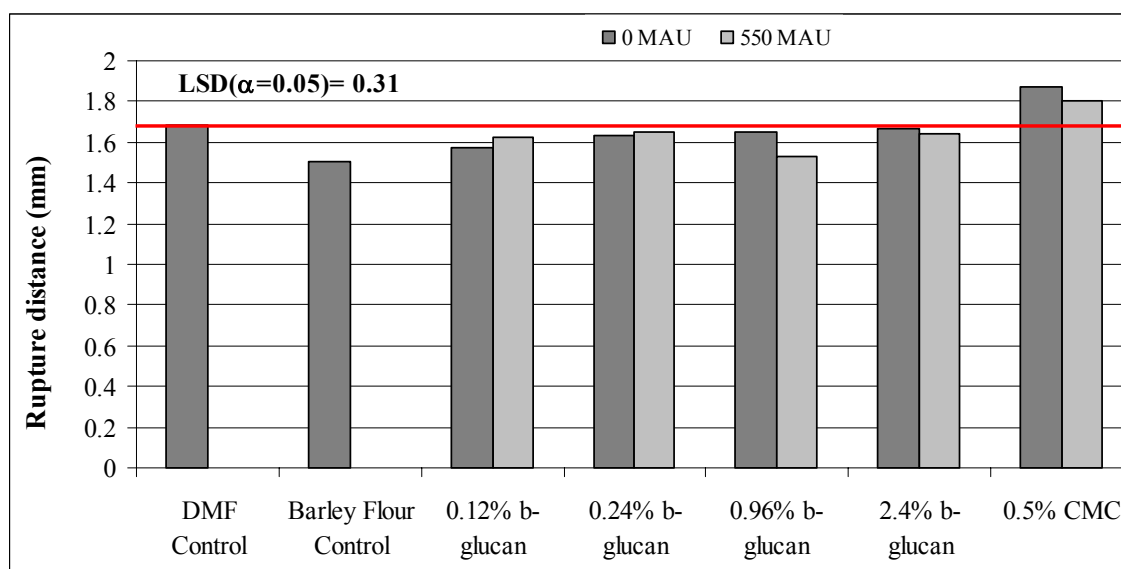


Fig. 25. Effect of β -glucan on Rupture Distance (mm) of Corn Tortillas Stored for 14 Days at 4°C.

At 4hr after productions, Young's modulus of barley flour control, 0.12% β -glucan and 0.5% CMC with or without amylase was significantly lower than the control (Appendix C.5). This trend continued after one day of storage; tortillas with 0.24% β -glucan and amylase were also lower. However, no treatment had significantly lower Young's modulus than control after 4 days of storage.

Young's modulus was significantly higher than control when using β -glucan or CMC in tortillas stored for 14 days at 4°C (Fig. 26). The higher values were obtained for 0.96-2.4% β -glucan in tortillas. The increase in rupture force caused by the use of hydrocolloids may be related to this phenomenon, being the cause of increased Young's modulus.

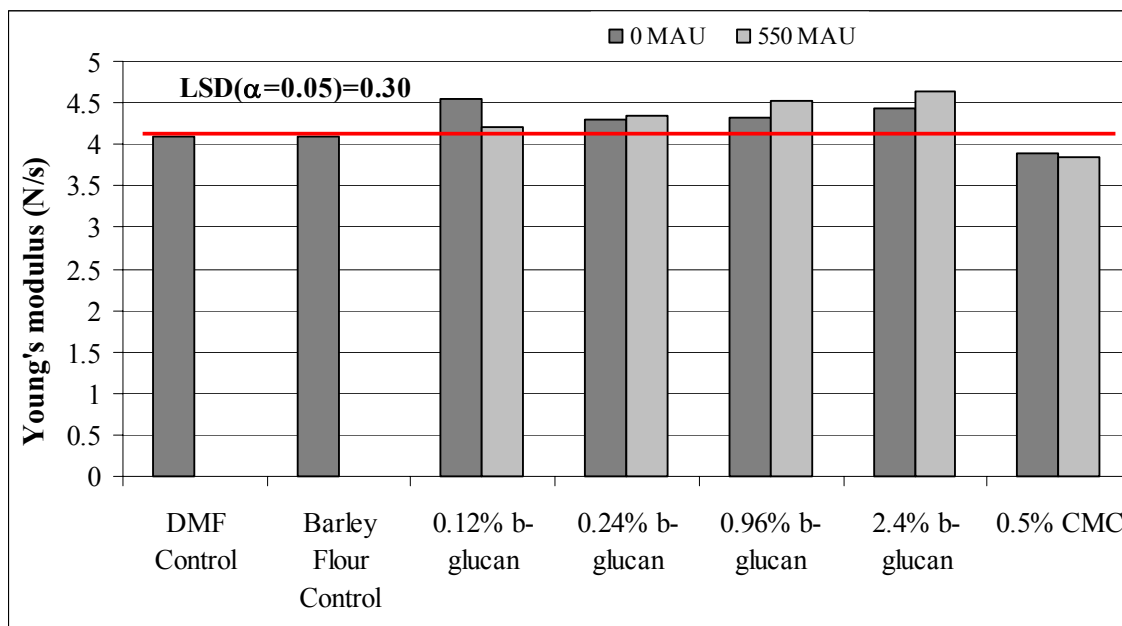


Fig. 26. Effect of β -glucan on Young's Modulus (N/s) of Corn Tortillas Stored for 14 Days at 4°C.

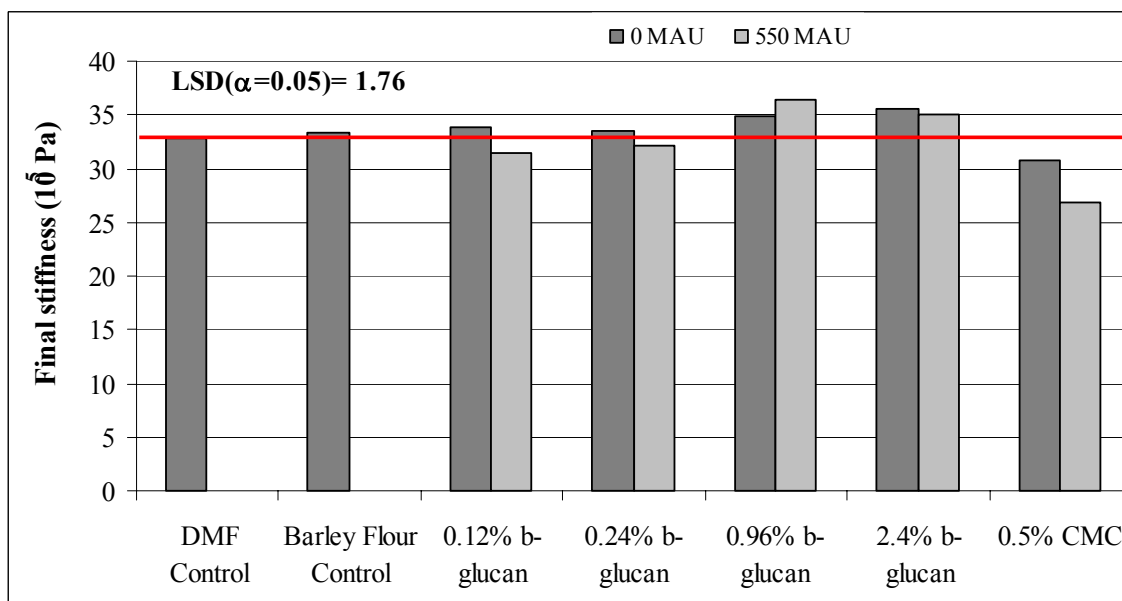


Fig. 27. Effect of β -glucan on Final Stiffness (10^5 Pa) of Corn Tortillas Stored for 14 Days at 4°C.

Final stiffness at 4 hr after production was significantly higher for 0.96-2.4% β -glucan with amylase (Appendix C.6). Amylase had some effect on the high levels of β -glucan at this point (20 minutes after baking). Only 0.5% CMC with or without amylase had a significant decrease in final stiffness after 7 days of storage. After 14 days, only CMC with amylase reduced the final stiffness significantly (Fig. 27). The use of amylase did not significantly affect final stiffness when combined with β -glucans.

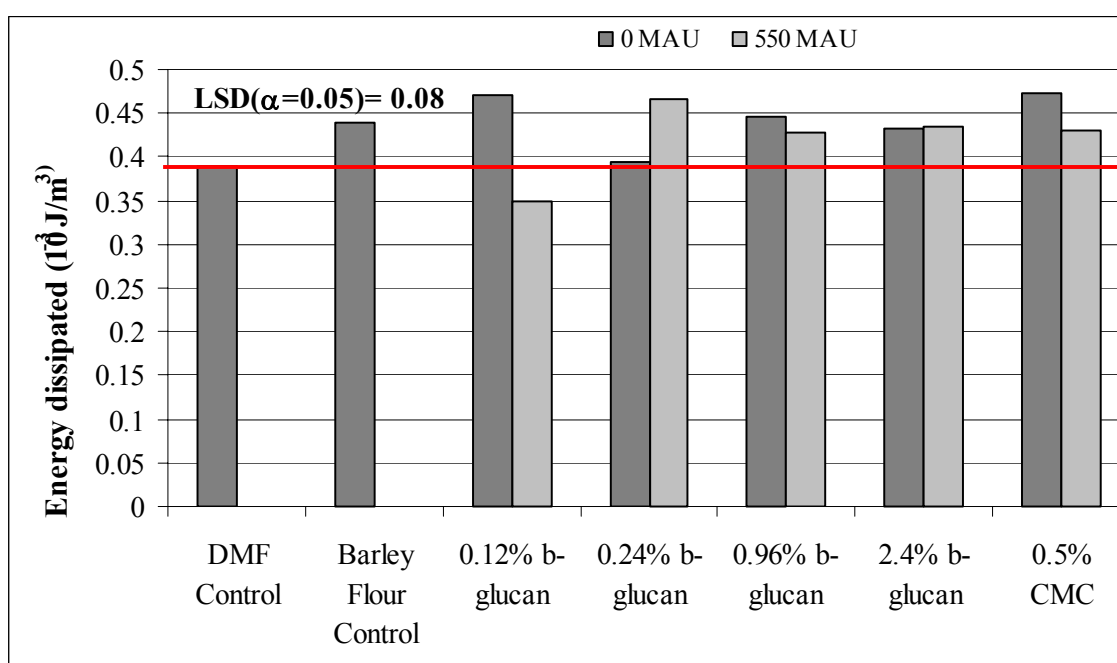


Fig. 28. Effect of β -glucan on Energy Dissipated (10^{-4} J/M^3) of Corn Tortillas Stored for 14 Days at 4°C .

The use of additives significantly affected energy dissipated throughout storage (Appendix C.7). At the initial measurements, only 0.12% β -glucan with 550 MAU was significantly higher than any other treatment. At low β -glucan content, amylase has a higher diffusion rate and increases the reduction of starch polymers, and this effect is even greater since the tortillas are fresh, and polymer aggregation has not occurred.

After 7 days of storage, tortillas with 0.12% β -glucan and 550 MAU had significantly higher value of energy dissipated compared to all the treatments. However, tortillas containing 2.4% β -glucan and 550 MAU amylase were significantly higher than control. Tortillas with 0.12% β -glucan and 0.5% CMC were the only treatments significantly higher in energy dissipated than control after 14 days of storage (Fig. 28). Addition of 0.12% β -glucan avoids aggregation of starch, keeping it more dispersed while levels higher than this may be creating another structure that yields increased rupture force and final stiffness.

Selection of the Optimum Levels

Tortillas containing 0.12% β -glucan were the optimum treatment. No differences were found in comparison to the control for rupture force, rupture distance, Young's modulus and final stiffness. Only the 2.5% barley flour control had significantly lower rupture force than control and other treatments, but it only increased pliability.

Tortillas with 0.5% CMC and 550 MAU had significantly better rollability, pliability, and final stiffness. However, 0.5% CMC and 0.12% β -glucan had significantly higher values of pliability, rollability and energy dissipated. This would suggest that 0.12% β -glucan is as effective as 0.5% CMC in preventing staling of corn tortillas; the lower percentage added suggested that 0.12% β -glucan was the best treatment.

Tortillas with concentrated β -glucans were not different than those with barley flour, except for rupture force. This suggests that β -glucans may be responsible for the improvement shown by barley flour in corn tortillas and the use of a concentrate enhances the effects.

CHAPTER VII
USE OF LOWER AMOUNTS OF β -GLUCANS COMBINED WITH CMC:
RESULTS AND DISCUSSION

The objective of this study was to evaluate the effect of combining β -glucans with CMC to lower the amount of additives in the corn tortilla formulation to improve tortilla texture over storage. Levels of concentrated barley β -glucans ranging between 0 and 0.24% were chosen from results in Chapter VI where a level of 0.12% β -glucan was significantly effective. The treatments used are shown in Table XIII.

TABLE XIII
Treatment Combinations for Barley β -glucan and CMC

Barley β - glucan (%)	CMC (%)
0	0
0	0.25
0	0.5
0.03	0
0.06	0
0.06	0.125
0.12	0
0.12	0.5
0.18	0.375
0.24	0
0.24	0.25
0.24	0.5

Masa and Tortilla Moisture, Tortilla pH, Thickness, Weight and Batch Yield

Masa moisture was significantly lower with β -glucan or CMC. Only 0.12% β -glucan was not different from control masa (Table XIV). Tortilla moisture was only significantly higher for tortillas with 0.12% β -glucan and 0.5% CMC. No differences were found in pH, batch yield, tortilla weight and tortilla thickness (Table XV).

TABLE XIV
Effect of Additive Combinations on Masa and Tortilla Moisture and pH^a

Barley β -glucan (%)	CMC (%)	Masa moisture (%)	Tortilla moisture (%)	pH
0	0	59.6 ^a	47.9 ^{bcde}	4.7 ^a
0	0.25	59.2 ^{bc}	47.5 ^e	4.6 ^a
0	0.5	59.2 ^{bc}	47.9 ^{bcde}	4.6 ^a
0.03	0	59.5 ^{ab}	48.5 ^{ab}	4.7 ^a
0.06	0	59.5 ^{ab}	47.6 ^{de}	4.6 ^a
0.06	0.125	59.5 ^{ab}	47.8 ^{cde}	4.6 ^a
0.12	0	59.5 ^a	48.6 ^{ab}	4.6 ^a
0.12	0.5	59.0 ^c	48.7 ^a	4.7 ^a
0.18	0.375	59.1 ^c	48.0 ^{bcde}	4.7 ^a
0.24	0	59.1 ^c	48.2 ^{abcd}	4.7 ^a
0.24	0.25	59.2 ^{bc}	48.4 ^{abc}	4.6 ^a
0.24	0.5	59.0 ^c	48.1 ^{bcde}	4.7 ^a
LSD (a=0.05)^b		0.37	0.63	0.15

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE XV
Effect of Additive Combinations on Batch Yield, Tortilla Weight and Thickness^a

Barley β -glucan (%)	CMC (%)	Batch yield (kg)	Tortilla weight (g)	Thickness (mm)
0	0	1.1 ^{ab}	23.8 ^a	1.7 ^{ab}
0	0.25	1.3 ^a	23.9 ^a	1.7 ^{ab}
0	0.5	1.1 ^{ab}	23.4 ^a	1.7 ^a
0.03	0	1.4 ^a	24.1 ^a	1.6 ^{ab}
0.06	0	0.9 ^{ab}	23.8 ^a	1.6 ^{ab}
0.06	0.125	1.1 ^{ab}	23.2 ^a	1.5 ^{ab}
0.12	0	1.0 ^{ab}	24.7 ^a	1.7 ^a
0.12	0.5	1.3 ^{ab}	23.9 ^a	1.6 ^{ab}
0.18	0.375	1.1 ^{ab}	23.5 ^a	1.6 ^{ab}
0.24	0	0.8 ^b	23.5 ^a	1.6 ^{ab}
0.24	0.25	1.0 ^{ab}	23.6 ^a	1.7 ^a
0.24	0.5	1.2 ^{ab}	23.7 ^a	1.4 ^b
LSD ($\alpha=0.05$)^b		0.45	1.58	0.30

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Texture Evaluation

Subjective Texture Measurements

Storage time and addition of β -glucan and/or CMC to corn tortilla significantly affected rollability and pliability scores during storage (Appendix D.1 and D.2). Even at the 4 hr after production, control was significantly less rollable than tortillas with additives at any level. After 14 days storage, 0.06-0.12% β -glucan and combinations of β -glucan with CMC had the best rollability scores (Fig. 29). Pliability was significantly higher when using 0.12% β -glucan or combinations of β -glucan with CMC (Fig. 30).

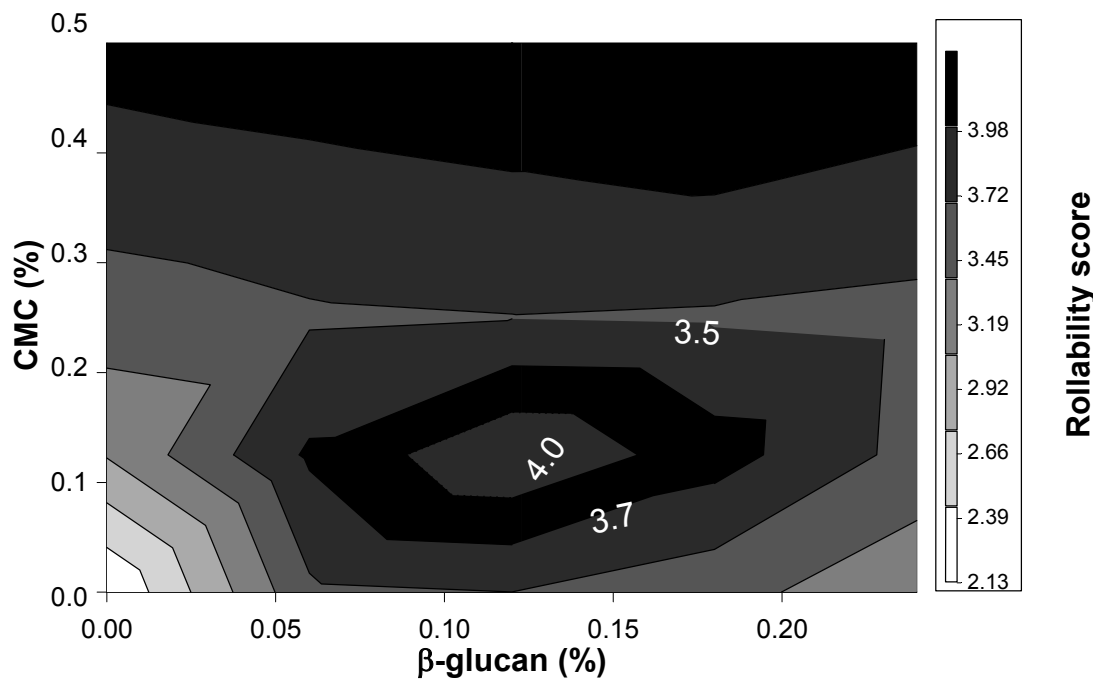


Fig. 29. Effect of β -glucan Content on Rollability of Corn Tortillas Stored 14 Days at 4°C. Note: $LSD(\alpha=0.05)=0.66$.

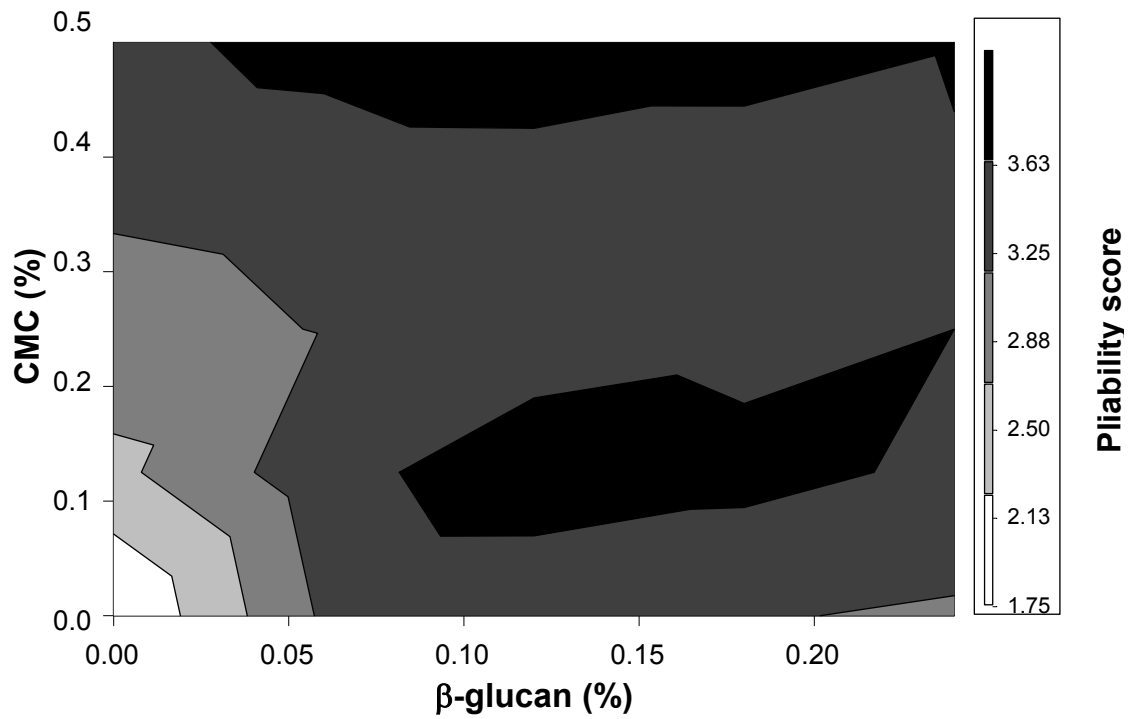


Fig. 30. Effect of β -glucan Content on Pliability of Corn Tortillas Stored 14 Days at 4°C. Note: $LSD(\alpha=0.05)=0.63$.

Objective Texture Measurements

Rupture force was significantly increased with storage time and additives (Appendix D.3). At the initial measurements, 0.5% CMC showed a rupture force significantly higher than other treatments. After one day of storage, no significant differences were detected from the control. At 14 days, no difference was seen in rupture force (Fig. 31). Probably β -glucan or CMC had created a stronger structure that required more force to rupture. However, it has been explained that a higher rupture force does not necessarily mean that tortilla is harder or more brittle (Bueso-Ucles 2003). It is the use of CMC and β -glucan that creates a flexible structure that holds tortilla together and needs a higher force to rupture than control tortillas.

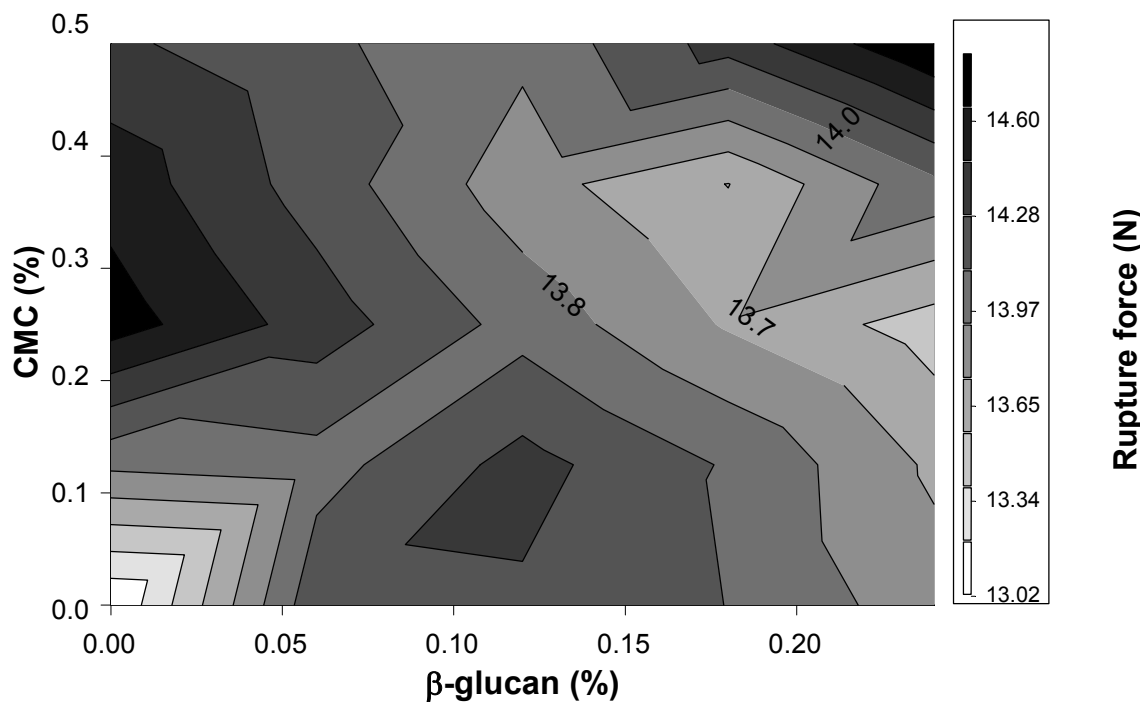


Fig. 31. Effect of β -glucan Content on Rupture Force (N) of Corn Tortillas Stored 14 days at 4°C. Note: $LSD(\alpha=0.05)=1.10$.

Rupture distance significantly decreased with storage and increased with the addition of β -glucan and CMC (Appendix D.4). Four hours after production, control tortillas had a significantly lower rupture force. Combinations of β -glucan and CMC had the highest rupture distance. At 4 days of storage, 0.12% β -glucan with 0.5% CMC, 0.18% β -glucan with 0.375% CMC, and 0.24% β -glucan with 0.5% CMC had significantly higher rupture distance than control. At 7 days, 0.12% and 0.24% β -glucan were not different than the control, having lower rupture distances. At 14 days only 0.24% β -glucan with 0.5% CMC had greater rupture distance (Fig. 32). More extensible tortillas were produced by the addition of these non-starch polysaccharides. However, CMC is providing the extensibility to tortillas; β -glucans do not significantly affect extensibility as seen before (Chapter VI).

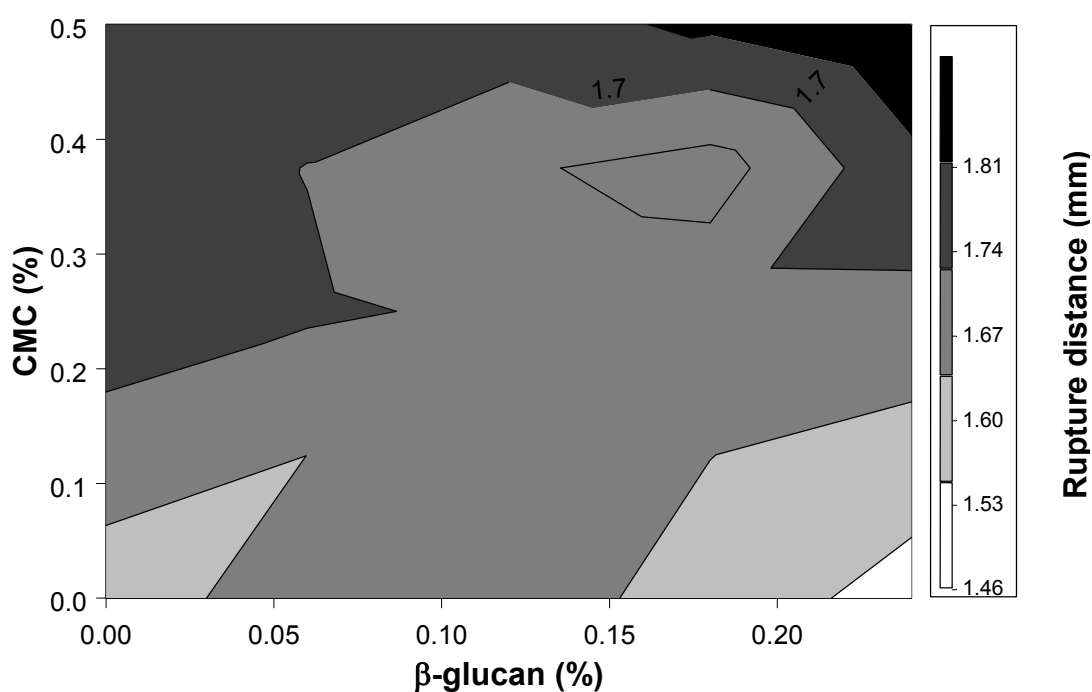


Fig. 32. Effect of β -glucan Content on Rupture Distance (mm) of Corn Tortillas Stored 14 Days at 4°C. Note: $LSD(\alpha=0.05)=0.21$.

Young's modulus significantly increased over time (Appendix D.5) and was not improved significantly by the use of additives. Control tortillas had the lowest value for this parameter (Fig. 33). Tortillas with β -glucan had increased Young's modulus values but combinations with CMC lowered this value, probably because of the higher rupture distances given by CMC.

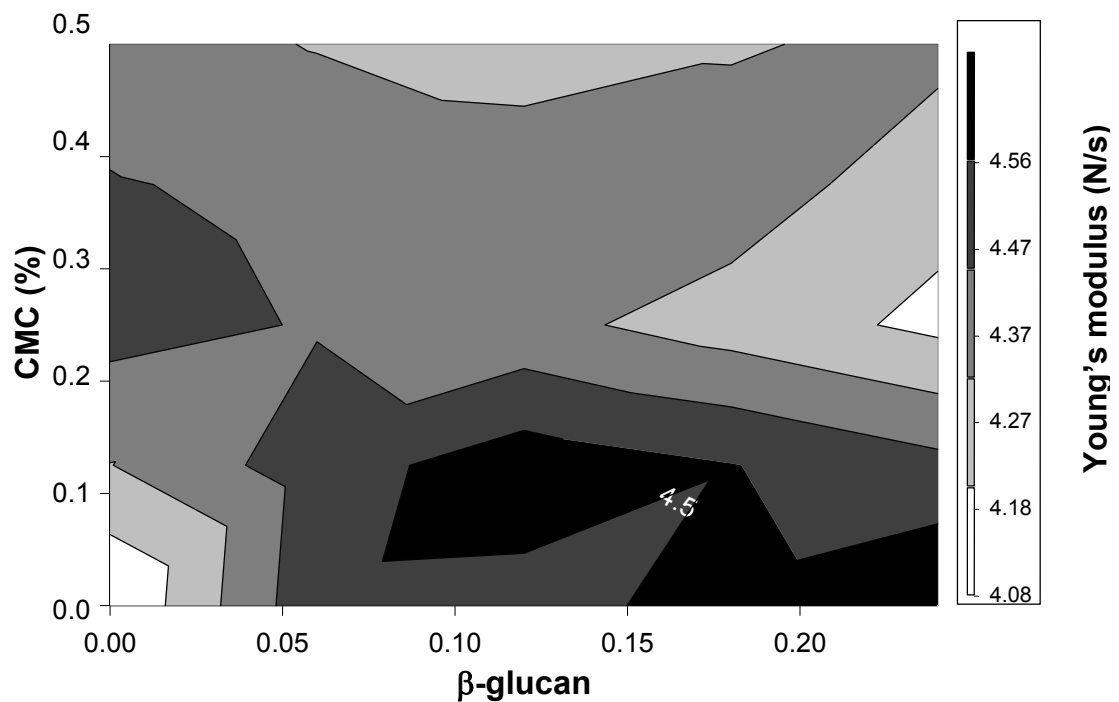


Fig. 33. Effect of β -glucan Content on Young's Modulus (N/S) of Corn Tortillas Stored 14 Days at 4°C. Note: $LSD(\alpha=0.05)=0.26$.

Final stiffness significantly increased with time (Appendix D.6). Initial measurements indicated a significantly higher stiffness in tortillas containing 0.24% β -glucan with 0.25% CMC. However, this treatment had significantly lower final stiffness found after 7 and 14 days of storage. At 14 days of storage, β -glucan level decreased

final stiffness and it had greater effect with increasing levels of CMC (Fig. 34). The lowest final stiffness values found after 14 days of storage was given by treatments with 0.12% β -glucan alone or combined with 0.5% CMC, 0.18% β -glucan combined with 0.375% CMC, and 0.24% β -glucan with 0.25 or 0.5% CMC. A synergistic effect was found by the action of these two additives. Amounts of the additives can be reduced for a better texture quality without jeopardizing other quality parameters.

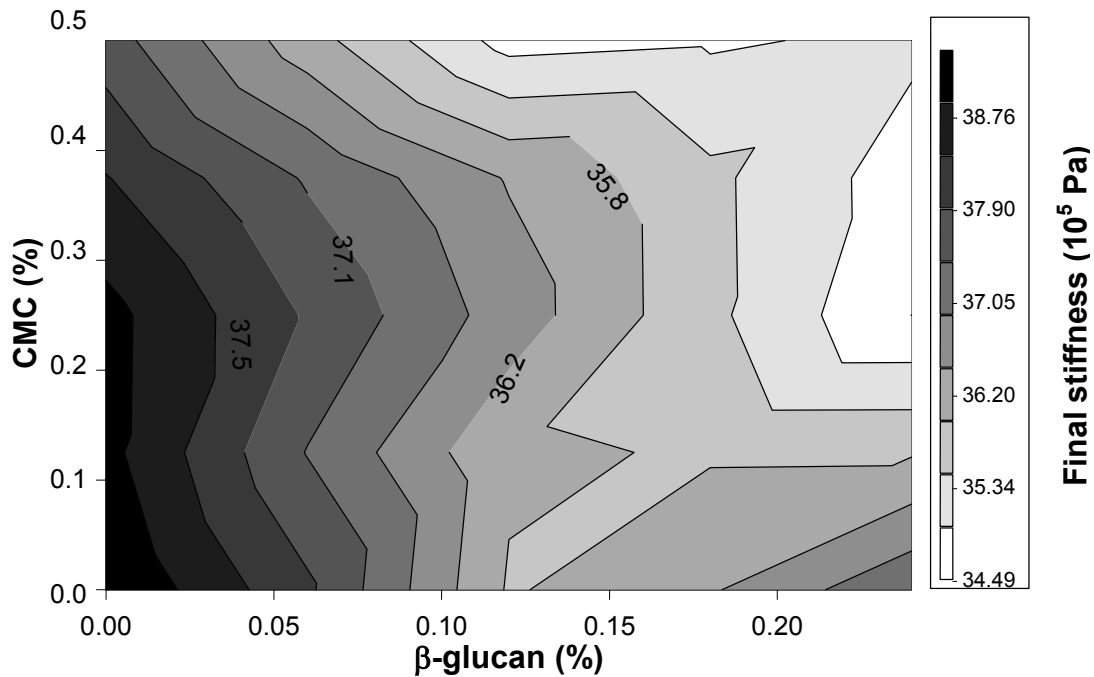


Fig. 34. Effect of β -glucan Content on Final Stiffness (10^5 Pa) of Corn Tortillas Stored 14 Days at 4°C. Note: $LSD(\alpha=0.05)=1.63$.

Energy dissipated significantly decreased after 7 days of storage and decreased further after 14 days (Appendix D.7). Tortilla control had significantly lower values of energy dissipated than 0.12% β -glucan at the initial measurement. After 7 days of storage, only 0.24% β -glucan combined with 0.25% CMC had significantly higher

energy dissipated than the rest of the treatments, including the control. After 14 days of storage, β -glucan level increased the energy dissipated of stored tortillas (Fig. 35). Little effect is seen with increasing levels of CMC. Tortillas with 0.12% β -glucan alone or with 0.5% CMC, 0.18% β -glucan with 0.375% CMC, 0.24% β -glucan alone or with 0.25-0.5% CMC had significantly higher energy dissipated than the control. However, these treatments had no significant difference in this parameter. Therefore, any of these combinations is as effective as the other in keeping the tortilla structure.

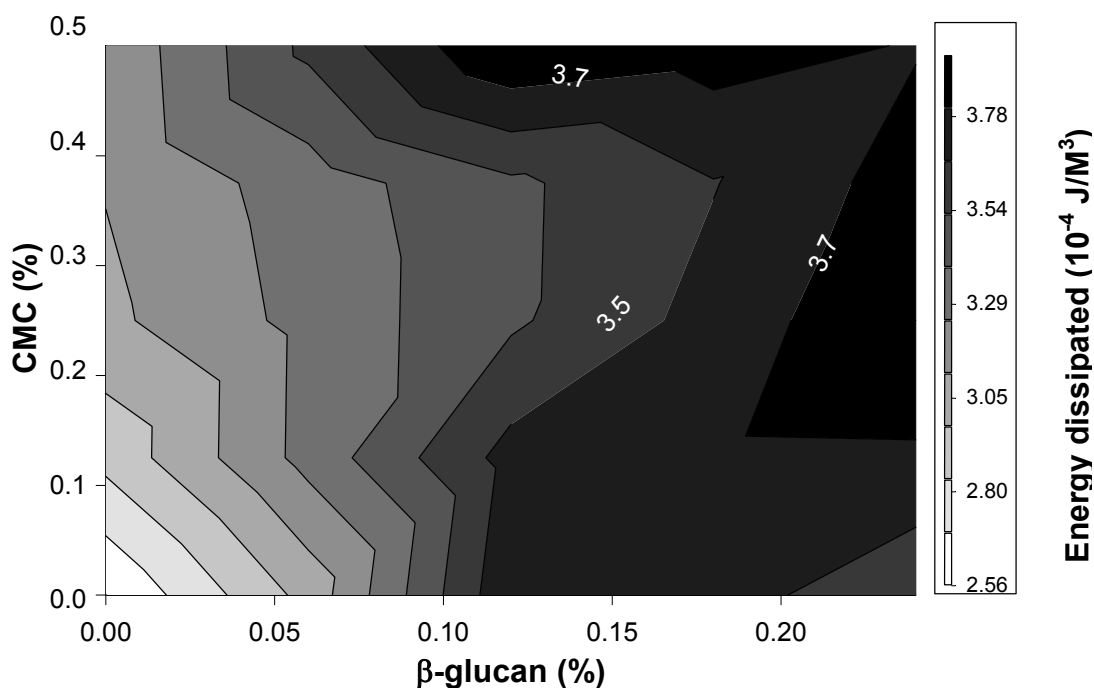


Fig. 35. Effect of β -glucan Content on Energy Dissipated (10^{-4} J/M^3) of Corn Tortillas Stored 14 Days at 4°C . Note: $\text{LSD}(\alpha=0.05)=0.07$.

Selection of Optimum Level

Optimum levels must be chosen at combinations of additives that improve the subjective and objective texture measurements. Treatments did not improve all

parameters measured. However, the best treatments should improve some parameters without affecting adversely others. Rupture force was not improved by the use of additives, but best treatments included those that did not significantly increased rupture force. In terms of rupture distance, only 0.24% β -glucan and 0.5% CMC gave a significantly higher extensibility. In addition, this treatment improved rollability, pliability, and energy dissipated. However, this are the highest levels of additives used, so it was discarded as an “optimum” level. Treatments with significantly better rollability, pliability, final stiffness, energy dissipated were 0.12% β -glucan, 0.18% β -glucan with 0.375% CMC, and 0.24% β -glucan with 0.25%. Other treatments were not significantly different and were higher in levels of additives. For example, treatments with different β -glucan content and 0.5% CMC had significantly better rollability, pliability, final stiffness and energy dissipated than control, but were not different from the treatments mentioned. Thus, the lower levels of additives that gave a significant improvement were chosen.

CHAPTER VIII
SENSORY EVALUATION:
RESULTS AND DISCUSSION

The objective of this experiment was to evaluate the sensory properties of treatments with the best texture measured by subjective and objective methods. Three combinations of β -glucan with CMC were used compared to the dry masa flour control (Table XVI). A positive control included was the optimum level of CMC and amylase determined by Bueso-Ucles (2003).

TABLE XVI
Treatment Combinations for Sensory Evaluation

Barley β- glucan (%)	Maltogenic amylase (MAU)	CMC (%)
0	0	0
0.12	0	0
0.18	0	0.375
0.24	0	0.25
0	825	0.25

Samples were produced in the laboratory with the formulations shown in Table I using fumaric acid and potassium sorbate) as preservatives. Tortillas were packaged and stored for 14 days at 4°C before sensory evaluation to allow staling.

For the evaluation, random numbers were assigned to the treatments. Tortillas were cut in half, warmed up for 10 seconds on each side on a hot plate, and placed on a plate with the number of each treatment.

Evaluation was performed by 20 untrained panelists. Water was provided for the panelists to rinse their mouth between samples. Parameters evaluated were as shown in Table XVII. Panelists were asked to score each sample for each parameter with a hedonic scale (1 to 10), where 10 is the highest or best score given to the tortilla.

TABLE XVII
Parameters Evaluated by Untrained Panelists

Overall appearance:	Consider color, smooth surface, burnt spots, blisters
Flexibility:	How the tortilla feels to the touch. 1=not flexible breaks, 5=can not fold easily, 10=very flexible
Hardness/Softness:	Tortilla is easy to bite. 1=very hard, 5=firm, 10=very soft
Chewiness:	Tortilla is hard to chew, sticks to teeth. 1=tough, 5=chewy, 10=tender
Gumminess:	Tortilla feels like rubber. 1=very gummy, 5=mealy, 10=dissolves in mouth easily
Overall quality:	Rate the tortillas: 1=dislike very much, 5=neither like or dislike, 10=like very much

Taste panel form is shown in Appendix D. For classification purposes, origin of panelist was asked (Mexican, Latin, Asian, American, European, African) as well as age and gender.

Panel Composition

Of the 20 panelists, 55% were female and 45% male. Panel origin was 35% American, 30% Mexican, 20% Asian, 5% Latin, 5% European and 5% African.

Sensory Analysis

Results of the sensory evaluation are in Table XVIII and Figure 36. No statistical differences were detected in overall appearance, flexibility, gumminess, flavor or overall quality. However, differences in softness and chewiness were found. Treatments with 0.12% β -glucan and 0.24% β -glucan with 0.25% CMC had no effect on tortilla quality. Additives were not detected by the panelists and treatments were similar to control. The rest of the treatments were scored significantly lower than the control tortillas, even though they were not significantly different than the other treatments with additives. CMC is known to cause chewiness in corn tortilla (Suhendro 1997). Some of the comments were related to toast spots in tortillas containing CMC, as well as an after taste and tougher texture in the 0.25% CMC and 825 MAU treatment. CMC is a great additive to provide flexibility to the corn tortilla, but it adds chewiness to the product. β -glucans work alone or synergistically with CMC to provide softness to reduce the chewiness.

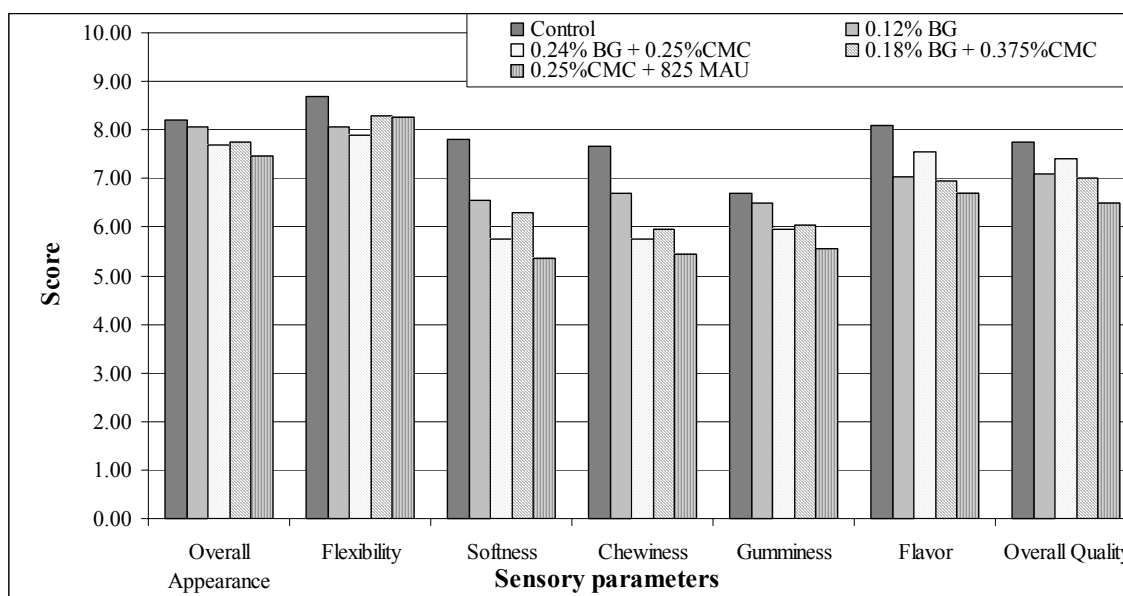


Fig. 36. Sensory Properties of 14 Day Old Reheated Corn Tortillas with Different Additives.

TABLE XVIII
Sensory Evaluation^a of 14 Day Old Reheated Corn Tortillas^b

β-glucan	CMC	Overall Appearance	Flexibility	Softness	Chewiness	Gumminess	Flavor	Overall Quality
0	0	8.2 ^a	8.7 ^a	7.8 ^a	7.7 ^a	6.7 ^a	8.1 ^a	7.8 ^a
0.12	0	8.1 ^a	8.1 ^a	6.6 ^{ab}	6.7 ^{ab}	6.5 ^a	7.1 ^a	7.1 ^a
0.24	0.25	7.7 ^a	7.9 ^a	5.8 ^{ab}	5.8 ^{ab}	6.0 ^a	7.6 ^a	7.4 ^a
0.18	0.375	7.7 ^a	8.3 ^a	6.3 ^b	6.0 ^b	6.1 ^a	7.0 ^a	7.0 ^a
0	0.25 ^c	7.5 ^a	8.3 ^a	5.4 ^b	5.5 ^b	5.6 ^a	6.7 ^a	6.5 ^a
LSD (α=0.05)^b		1.62	1.35	1.63	1.86	2.30	2.08	1.87

^aParameters evaluated with hedonic scale (1-10).

^b Means in the same column followed by the same letter are not significantly different.

^c CMC+825MAU; ^d LSD = Least significant difference for means separation.

TABLE XIX
Texture Evaluation of 14 Day Old Tortillas Equilibrated to Room Temperature^a

β-glucan	CMC	Rollability	Pliability	Rupture force	Rupture distance	Young's modulus	Final stiffness	Energy dissipated
0	0	1.0 ^b	1.8 ^b	13.51 ^b	1.10 ^b	3.34 ^b	38.76 ^a	0.26 ^b
0.12	0	2.3 ^{ab}	2.3 ^{ab}	15.21 ^{ab}	1.40 ^{ab}	4.08 ^{ab}	35.73 ^{bc}	0.36 ^a
0.24	0.25	3.0 ^{ab}	2.8 ^a	15.98 ^a	1.53 ^a	4.36 ^a	34.49 ^c	0.38 ^a
0.18	0.375	3.5 ^a	2.8 ^a	15.10 ^{ab}	1.30 ^{ab}	3.98 ^{ab}	35.44 ^{bc}	0.35 ^a
0	0.25	2.8 ^{ab}	2.0 ^{ab}	14.57 ^{ab}	1.25 ^{ab}	3.70 ^{ab}	37.28 ^{ab}	0.27 ^b
LSD ($\alpha=0.05$)^b		2.23	0.81	0.83	0.31	0.83	2.55	0.07

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

Texture Evaluation

Tortillas subjected to sensory evaluation were also analyzed for subjective and objective texture evaluation (Table XIX). Texture measurements were done in 14 day old tortillas stored at 4°C after equilibration to room temperature, as previously.

Subjective Texture Measurements

Rollability and pliability were significantly increased with treatments compared to control. Rollability was higher with 0.18% β -glucan and 0.375% CMC (Fig. 37). However, pliability was higher for 0.18% β -glucan with 0.375% CMC and for 0.24% β -glucan and 0.25% CMC (Fig. 38).

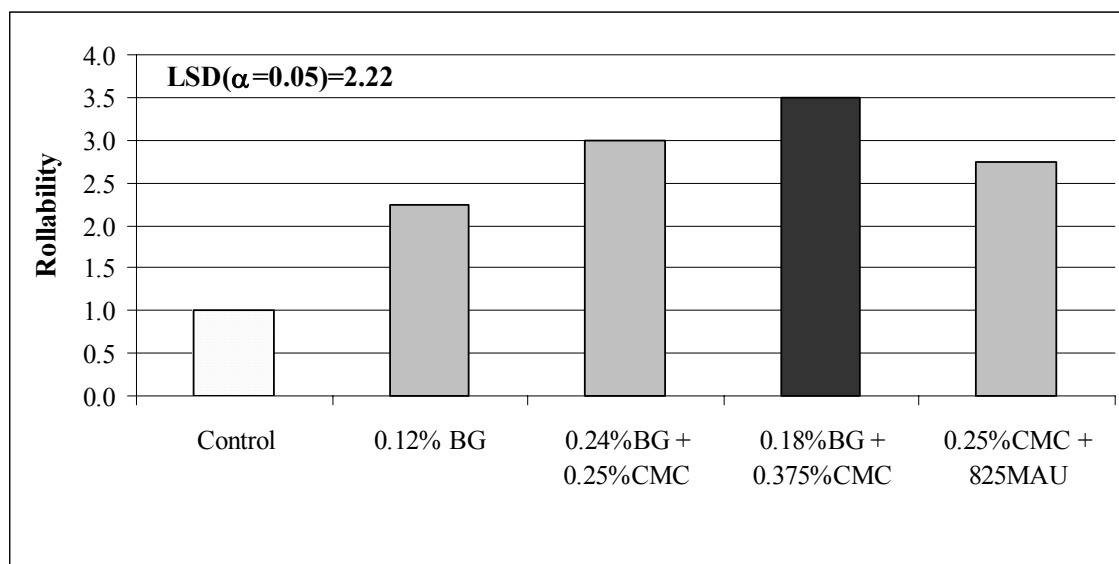


Fig. 37. Rollability Scores of Combinations Used for Sensory Analysis.

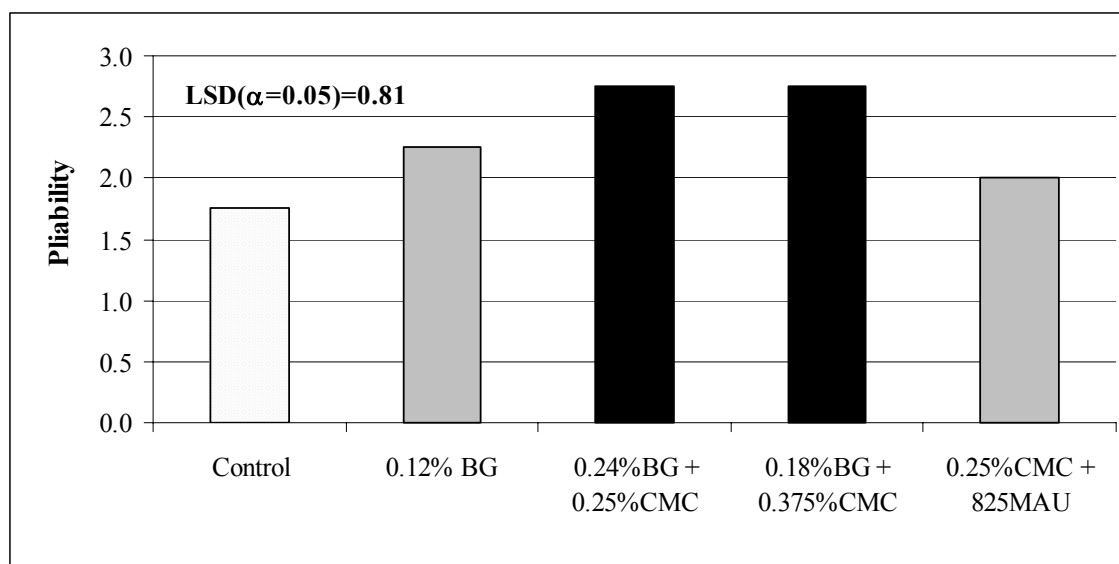


Fig. 38. Pliability Scores of Tortillas with Combinations Used for Sensory Analysis.

Objective Texture Measurements

Rupture force was increased by the use of treatments (Fig. 39), which confirms previous results (Chapter VII). However, rupture distance was significantly increased by the use of 0.24% β -glucan and 0.25% CMC (Fig. 40). Young's modulus, as rupture force, was increased by treatments (Fig. 41).

Final stiffness decreased with treatments, being 0.24% β -glucan and 0.25% CMC the lowest (Fig. 42). Energy dissipated was increased by treatments, except with 825 MAU and 0.25%CMC, which was not different from control tortillas (Fig. 43).

An important fact is that tortillas were reheated, and this resembles the fresh tortilla texture quality (Fernandez de Castro1998, Mitre-Dieste 2001). Panelists did not detect differences in flexibility, even though the rollability and pliability scores reported in Chapter VII were significant. This is because subjective texture measurements were done with the tortilla equilibrated to room temperature after storage at 4°C and not reheated. Consumers keep the tortillas in the refrigerator where temperature is in the range of 4-10°C. Their first contact with the tortilla is either cold or equilibrated to room

temperature; at this time, they notice the staling or hardening of the product. Furthermore, the hardening of the product can cause breaking of the tortilla and high losses in commercial stores. Therefore, the importance of the sensory evaluation resides more on whether the consumer can detect the use of additives in the product, or perceive a negative property in the product.

As far as the sensory evaluation goes, the best treatments were control, 0.12% β -glucans and 0.24% β -glucan with 0.25% CMC with no differences shown among them. Only the tortillas with additives maintained adequate flexibility as measured by rollability and pliability.

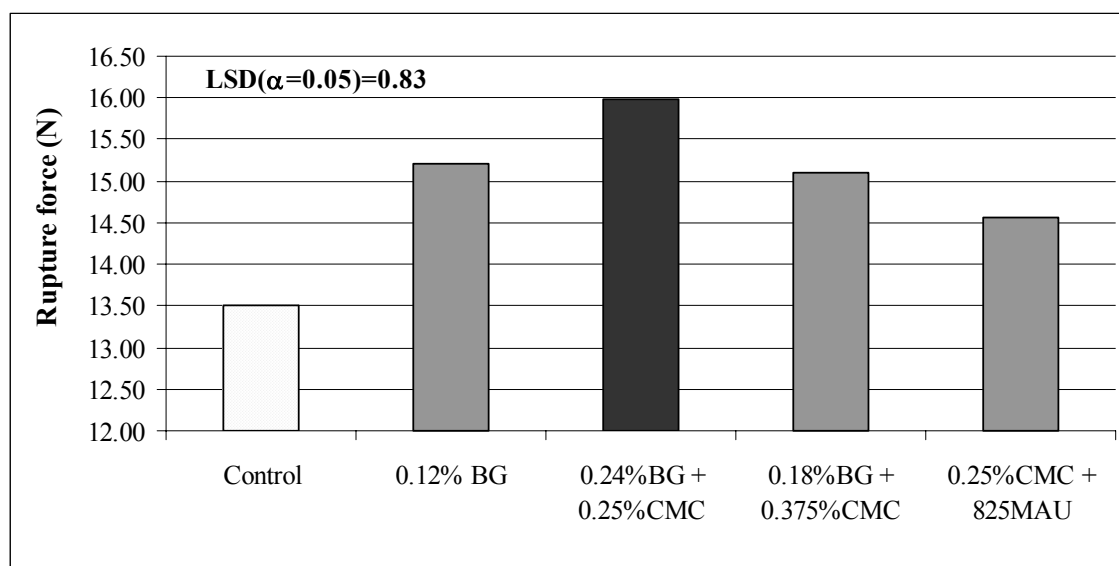


Fig. 39. Rupture Force of Tortillas with Combinations Used for Sensory Analysis.

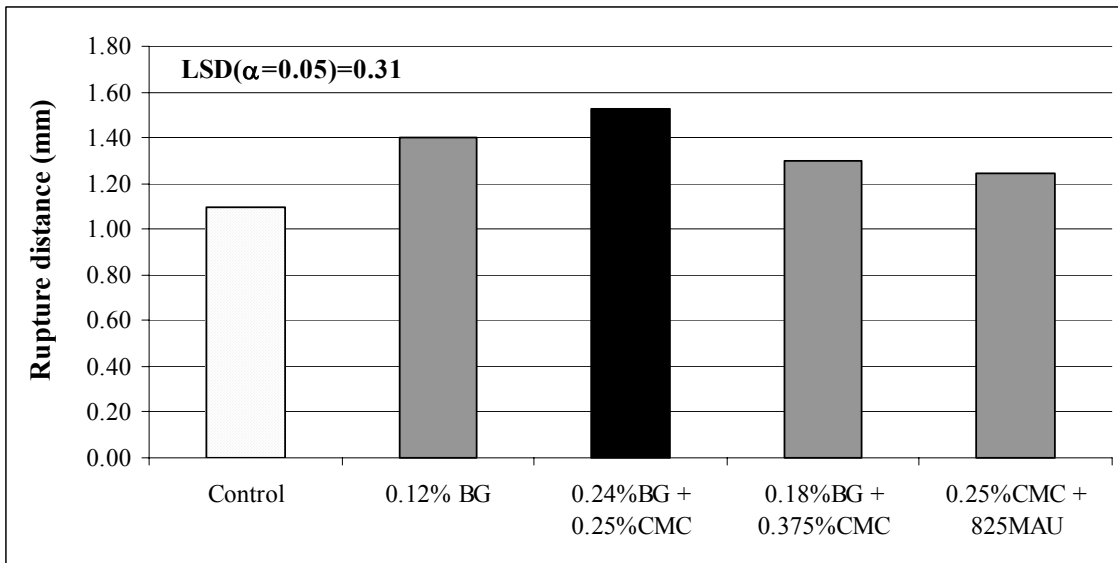


Fig. 40. Rupture Distance of Tortillas with Combinations Used for Sensory Analysis.

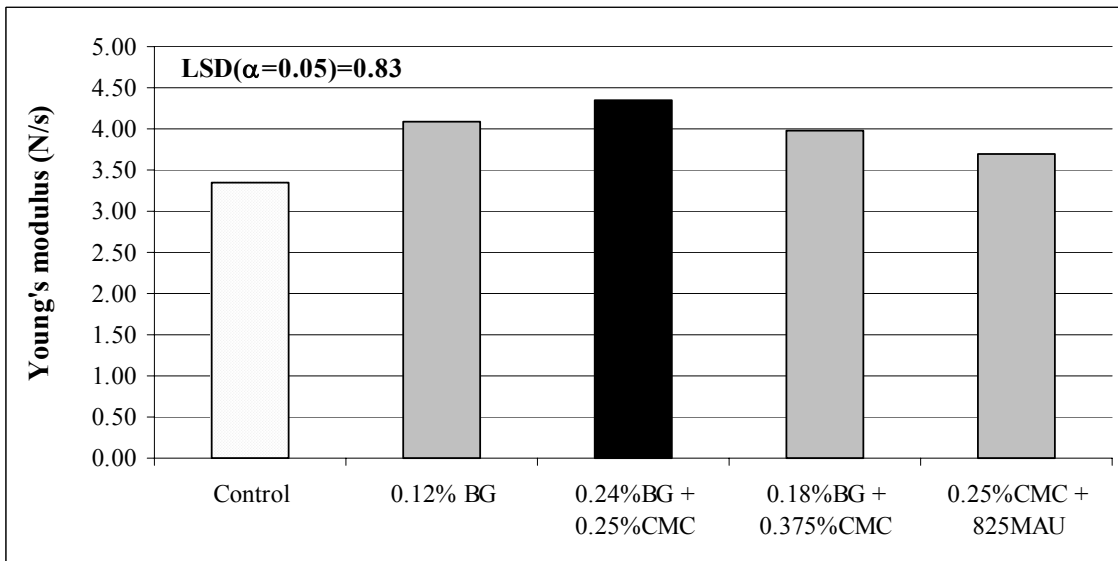


Fig. 41. Young's Modulus of Tortillas with Combinations Used for Sensory Analysis.

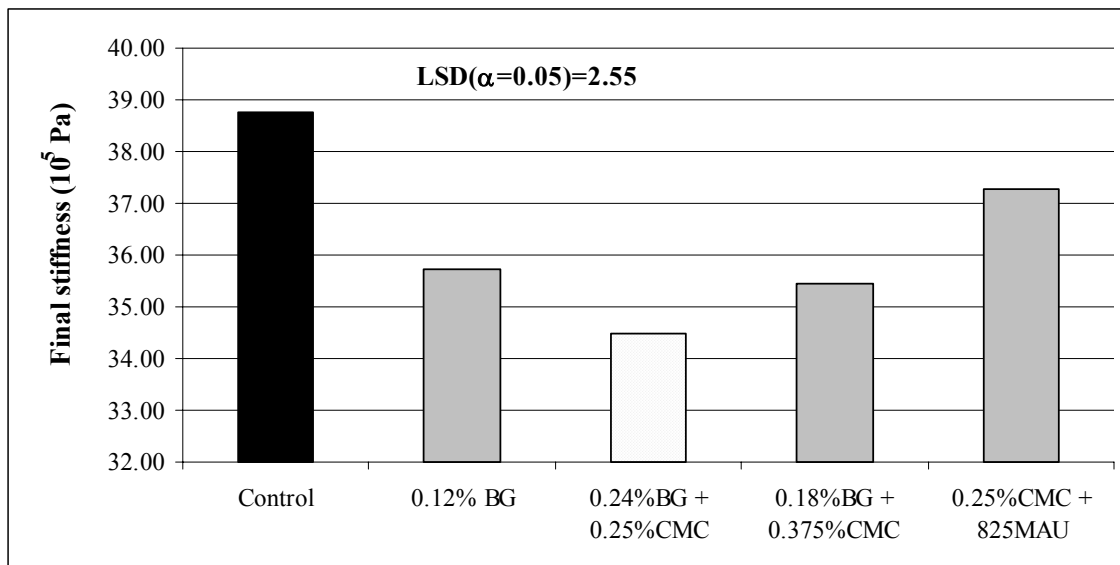


Fig. 42. Final Stiffness of Tortillas with Combinations Used for Sensory Analysis.

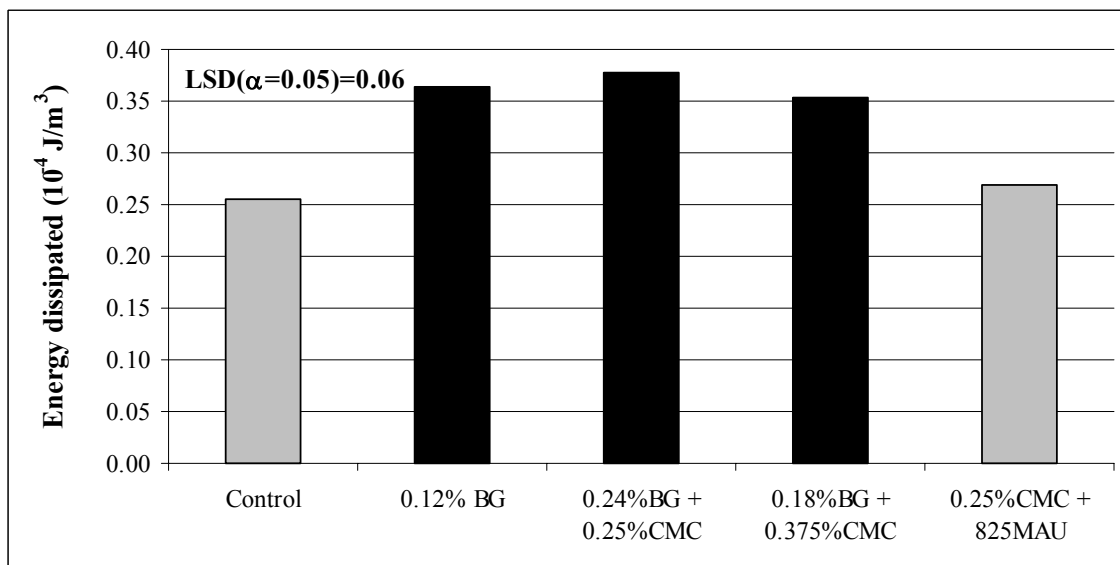


Fig. 43. Energy Dissipated of Tortillas with Combinations Used for Sensory Analysis.

Selection of the Optimum Level

Texture of tortillas was improved by the use of combinations of β -glucan and CMC. The best response was found with 0.24% β -glucan and 0.25% CMC. This treatment had increased rollability, pliability and energy dissipated with decreased final stiffness. Sensory evaluation of this treatment was rated as good as the control. Surprisingly, the positive control (825 MAU and 0.25% CMC) was not better than the treatments with β -glucan and CMC, even though it has been proposed as an effective treatment for staling retardation. The combination of 825 MAU (to interfere with intra-granular amylopectin re-crystallization) and 0.25% CMC (to create a more flexible inter-granular matrix than retrograded amylose) produced less stiff, equally flexible and less chewy tortillas than 0.5% CMC (Bueso-Ucles 2003). In the sensory evaluation performed in this study, the treatments were compared to a control with no additives, and differences were found.

CHAPTER IX
TEXTURE PARAMETERS EVALUATION:
RESULTS AND DISCUSSION

The use of different texture measurements gives us an idea of the changes that corn tortillas undergo during storage and the effects of different additives on texture. Two subjective measurements were performed in this study: rollability and pliability. Advantages of these methods are the short time involved without expensive equipment. However, the values change with different observers.

Objective extensibility and stress relaxation tests were done using a Texture Analyser. Extensibility test pulls the tortilla until it breaks and measures rupture force and rupture distance. From this test, Young's modulus was calculated; it relates the force needed for rupture with the strain (deformation) of the sample. The stress relaxation method is a theoretical approach for calculation of final stiffness and energy dissipated. Extensibility and stress relaxation methods were compared (Table XX). The advantages of the extensibility test are the reduced time for the test and simple data calculations. Stress relaxation takes longer to test and the calculations require more time.

TABLE XX
Comparison of Objective Texture Evaluation Methods

	Extensibility	Final stiffness
Test time	20 s/sample	3 min/sample
Data manipulation		2 min/sample
Value calculation	Instantaneous	30 s/sample

The model p-value from the ANOVA summary defines if the variables used explain the response. In this study, model was determined by all variables and their interactions to obtain the parameter estimates for graph building. The data is fitted to this

model and the R^2 defines how the model explains the behavior of responses according to data provided. An R^2 of 1, is a perfect data fit. The coefficient of variation (CV) defines the variability of the data for a specific response. For research purposes, a good model has a significant p-value (<0.05), coefficients lower than 15% and an R^2 of 0.5-1.

The model R^2 for yield was 0.60 with a coefficient of variation of 19.6% indicating high variability (Appendix F.1). Batch yields were not significantly different among treatments. Tortilla weight and tortilla thickness had good R^2 (0.69 and 0.56 respectively) and low coefficient of variation (2.42 and 3.81%) indicating good repeatability of the measurements.

Objective texture measurements of tortillas 4 hr after production showed significant model correlation at the initial measurement, while pliability and rollability did not (Appendix F.2). However, R^2 was only good for energy dissipated and Young's modulus.

After 14 days of storage, pliability had a higher R^2 (0.83) and lower coefficient of variation (10.1%) than rollability (0.63 and 18.43% respectively) and a significant p-value (Appendix F.3). The model R^2 values were similar for rupture force (0.56) and final stiffness (0.55). However, coefficient of variation was better for final stiffness (6.54%) than for rupture force (8.16%). Young's modulus had a significant model correlation with $R^2=0.43$ and a coefficient of variation of 9.2%, which is also acceptable.

Model R^2 was only 0.27 for energy dissipated and 0.18 for rupture distance with coefficient of variation of 21.33 and 15.44% respectively. In fact, rupture distance only detected differences in the CMC factor, while energy dissipated showed significant differences in terms of barley flour and CMC interaction. This confirms that energy dissipated is a more sensitive measure of texture changes in tortillas than rupture distance.

At 14 days of storage, rupture force, Young's modulus and final stiffness were better indicators of tortilla texture as well as rollability and pliability.

For texture evaluation, subjective measurements have better model correlations, R^2 and coefficients of variation, but an objective measurement is needed to explain

physically what changes are taking place in texture properties. This objective measurement should correlate with the subjective measurement used for texture evaluation. Correlations of the subjective methods (rollability and pliability) and objective methods (extensibility and stress relaxation) used in this study were done to determine which measurements are better suited for texture evaluation in corn tortillas (Table XXI).

All parameters had significant correlations (Table XXI). Pliability and rollability correlated well to each other ($R^2=0.83$), which suggests that there is no need to measure both. Furthermore, pliability correlated better than rollability to all objective measurements. However, the highest correlation between subjective and objective measurements was for final stiffness with pliability ($R^2=0.88$). From the extensibility measurements, Young's modulus had the highest correlation with pliability ($R^2=0.86$). In addition, final stiffness and Young's modulus had the best correlation among objective measurements ($R^2=0.98$). Final stiffness is a theoretical calculated value like Young's modulus. Given these results, and considering that extensibility is a test that takes less time and data manipulation, it is suggested that Young's modulus is the best objective measurement and it is correlated with pliability.

When analyzing data in separate days, different results are seen. In the initial measurements (4 hr after production), only the measurements of extensibility correlated to each other. Rollability and Pliability did not correlate with any objective measurement at this time. Young's modulus correlated with rupture force and rupture distance (Table XXII).

Better correlations were found at 7 days (Table XXIII). Rupture force correlated to all texture measurements, having the highest correlation with rupture distance. Final stiffness correlated with energy dissipated significantly. Rollability and pliability correlated to each other significantly. Rollability correlated better than pliability with final stiffness and energy dissipated. Pliability correlated better to rupture force and Young's modulus. This is because pliability is a large strain procedure as well as extensibility tests, where the tortilla is subjected to rupture, while less strain is involved

in rollability and stress relaxation. The theoretical values of final stiffness and Young's modulus had a significant correlation.

At the end of storage (14 days), rupture force correlated with rupture distance, Young's modulus, final stiffness and rollability (Table XXIV). The highest correlation among objective measurements was found with Young's modulus and final stiffness. At this point, rollability correlated better with extensibility rupture force and Young's modulus and pliability correlated with final stiffness and energy dissipated.

This results show that depending on the stage of storage, objective and subjective methods correlate differently. At the end of storage, rollability had better sensitivity than pliability and better correlated to extensibility. Pliability had good correlations with final stiffness and energy dissipated, but not better than the correlations of rollability with extensibility measurements. It is suggested that depending on the time of texture analysis, the methods should be appropriately correlated for better results.

TABLE XXI
Texture Measurement Correlations of Tortillas at 4 hr, 7 Days and 14 Days

	Rupture Force	Rupture Distance	Young's modulus	Final Stiffness	Energy Dissipated	Rollability	Pliability
Rupture Force	1	-0.87**	-0.96**	-0.96**	-0.80**	-0.68**	-0.83**
Rupture Distance	-0.87**	1	-0.94**	-0.92**	0.80**	0.71**	0.83**
Young's modulus	-0.96**	-0.94**	1	0.98**	-0.83**	-0.73**	-0.86**
Final Stiffness	-0.96**	-0.92**	0.98**	1	-0.84**	0.74**	-0.88**
Energy Dissipated	-0.80**	0.80**	-0.83**	-0.84**	1	0.63**	0.75**
Rollability	-0.68**	0.71**	-0.73**	0.74**	0.63**	1	0.83**
Pliability	-0.83**	0.83**	-0.86**	-0.88**	0.75**	0.83**	1

**Correlation is significant at the 0.01 level (2-tailed); n=864.

TABLE XXII
Texture Measurement Correlations of Tortillas 4 hr after production

	Rupture Force	Rupture Distance	Young's modulus	Final Stiffness	Energy Dissipated	Rollability	Pliability
Rupture Force	1.00	0.12	0.35**	0.03	-0.09	0.04	0.07
Rupture Distance	0.12	1.00	-0.81**	0.06	-0.01	0.08	0.09
Young's modulus	0.35**	-0.81**	1.00	-0.03	-0.05	-0.01	-0.05
Final Stiffness	0.03	0.06	-0.03	1.00	-0.09	0.05	0.06
Energy Dissipated	-0.09	-0.01	-0.05	-0.09	1.00	0.01	0.06
Rollability	0.04	0.08	-0.01	0.05	0.01	1.00	-0.02
Pliability	0.07	0.09	-0.05	0.06	0.06	-0.02	1.00

**Correlation is significant at the 0.01 level (2-tailed); n=288.

TABLE XXIII
Texture Measurement Correlations of Tortillas Stored for 7 Days at 4°C

	Rupture Force	Rupture Distance	Young's modulus	Final Stiffness	Energy Dissipated	Rollability	Pliability
Rupture Force	1.00	0.76**	0.50**	0.52**	-0.34**	-0.20**	-0.20**
Rupture Distance	0.76**	1.00	-0.12	0.19	-0.20	-0.12	-0.09
Young's modulus	0.50**	-0.12	1.00	0.55	-0.30	-0.22**	-0.24**
Final Stiffness	0.52**	0.19	0.55	1.00	-0.44	-0.30**	-0.29**
Energy Dissipated	-0.34**	-0.20	-0.30	-0.44	1.00	0.22**	0.15
Rollability	-0.20**	-0.12	-0.22**	-0.30**	0.22	1.00	0.55**
Pliability	-0.20**	-0.09	-0.24**	-0.29**	0.15	0.55**	1.00

**Correlation is significant at the 0.01 level (2-tailed); n=288.

TABLE XXIV
Texture Measurement Correlations of Tortillas Stored for 14 Days at 4°C

	Rupture Force	Rupture Distance	Young's modulus	Final Stiffness	Energy Dissipated	Rollability	Pliability
Rupture Force	1.00	0.35**	0.60**	0.40**	-0.01	0.22**	0.04
Rupture Distance	0.35**	1.00	-0.47**	-0.21**	0.12	0.02	0.10
Young's modulus	0.60**	-0.47**	1.00	0.50**	-0.10	0.21**	-0.04
Final Stiffness	0.40**	-0.21**	0.50**	1.00	-0.46**	0.07	-0.13**
Energy Dissipated	-0.01	0.12	-0.10	-0.46**	1.00	0.03	0.13**
Rollability	0.22**	0.02	0.21**	0.07	0.03	1.00	0.51**
Pliability	0.04	0.10	-0.04	-0.13**	0.13**	0.51**	1.00

**Correlation is significant at the 0.01 level (2-tailed); n=288.

CHAPTER IX

SUMMARY AND CONCLUSIONS

Barley flour improved tortilla texture, as shown by increased rollability, pliability and energy dissipated, in addition to reduced rupture force, and final stiffness. Tortillas with 2.5% barley flour had significantly improved quality without any negative effects. Higher levels of barley flour were not different in texture but had darker color.

Maltogenic amylase improved the texture of corn tortillas. Tortillas with 825 MAU had increased rollability, pliability, rupture distance and decreased rupture force, Young's modulus and final stiffness but decreased energy dissipated. Pliability was negatively affected at 1650 MAU. When amylases are added to dry masa flour, the substrate most easily hydrolyzed are the starch molecules that had been leached or partially leached from starch granules and enzyme activity would be concentrated on the outside or near the surface of the granule. At these conditions, amylase activity time is long but there is limited substrate. During baking, more substrate is available for a limited time. A short window of amylase activity exists between the temperature at which starch begins to swell and temperature at which amylases are denatured (Martin et al. 1991). The enzymes are deactivated as the surface cooking temperature of 80-90°C is reached and starch hydrolysis stops (Iturbe-Chiñas et al. 1996, Drapron and Godon 1987). Amylases hydrolyzed linkages within the amorphous regions of the starch matrix during baking and reduced the rate of starch retrogradation. (Si 1997, Suhendro 1997). When only amylases are used, the intra-granular phase (amylopectin) crystallizes less over storage producing softer tortillas, as shown by DSC data from Bueso-Ucles 2003. However, it also weakens the amylose matrix by breaking down its molecules (Bueso-Ucles 2003). The anti-staling properties of maltogenic amylase rely on preventing the intra-granular re-crystallization of amylopectin in tortillas during storage (Bueso-Ucles 2003).

CMC improved rollability, pliability, rupture distance in stored tortillas. The anti-staling properties of CMC are probably related to its ability to create a flexible

amorphous matrix in the continuous phase of tortillas that does not interfere with amylopectin retrogradation during storage (Suhendro 1997, Yeggy 2000, Bueso-Ucles 2003).

The combination of amylase and barley flour improved corn tortilla texture. These additives increased rollability, pliability, energy dissipated and reduced rupture force, Young's modulus and final stiffness. The combination of CMC with barley flour improved pliability and rupture distance, but when these additives were used with 825 MAU, pliability, rollability, rupture distance increased while rupture force and Young's modulus decreased.

Tortillas with 0.12% β -glucan had increased rollability, pliability and energy dissipated values, equal to that of tortillas containing 0.5% CMC. The use of amylase combined with β -glucan did not improve tortilla texture quality. This is probably because the level of amylase used was lower than the optimum level.

Combinations of β -glucan and CMC gave tortillas with significantly better rollability, pliability, final stiffness, energy dissipated and rupture distance but increased the rupture force. These hydrocolloids created a very strong structure, making tortillas harder to rupture. However, rupture force may be a misleading indicator of tortilla hardness, since some materials require higher rupture force because they are more extensible, and not because they are harder or more brittle (Bueso-Ucles 2003). This is confirmed also by the fact that subjective measurements are high, but usually these treatments were more "gummy" or "rubbery".

The use of low levels of β -glucan and CMC gave optimum treatments of 0.12% β -glucan, 0.24% β -glucan with 0.25% CMC and 0.18% β -glucan with 0.375% CMC. These treatments were subjected to sensory evaluation of reheated tortillas, including a control with no additives or with 825 MAU and 0.25% CMC). Tortillas with 825 MAU and 0.25% have been reported to be less chewy compared to 0.5% CMC. However, untrained panelists rated tortillas containing 825 MAU with 0.25% CMC and 0.18% β -glucan with 0.375% CMC tougher and chewier, compared to the control. Fewer differences are seen in reheated tortillas, since texture of reheated tortillas are is similar

to that of fresh tortillas (Fernandez de Castro 1998, Mitre-Dieste 2001). The texture measurements performed at room temperature, however, showed differences in all parameters. The first contact consumers have is at room or refrigeration temperatures since tortillas are stored at these conditions. Hardening of tortillas is noticeable when handling the tortillas, while the use of additives is noticed when consumer actually eats warmed up tortillas and can perceive chewiness or toughness. Evaluation of texture measurements (done at room temperature) and sensory evaluation (reheated) suggest that 0.24% β -glucan and 0.25% CMC was the best treatment. The similar results from β -glucan and barley flour suggest that β -glucan may be responsible for the improvement in tortilla texture by barley flour substituted for dry masa flour in corn tortilla formulations. Methods and conditions of β -glucan extraction strongly affect viscosity of this hydrocolloid (Burkus and Temelli, 1998). Therefore, differences between barley flour and β -glucan may be attributed to β -glucan structure, since concentrated β -glucan were not extracted from the particular flour used for this study.

Barley flour or β -glucan have a different mechanism of retarding staling than does CMC. CMC imparts extensibility to corn tortilla as exhibited by increased rupture distance and pliability. In contrast, barley flour or β -glucans impart a softer texture to corn tortillas, as exhibited by increased rollability, pliability energy dissipated and reduced final stiffness. Corn tortilla textural changes were mainly related to modifications in the starch properties. Staling is a progressive, spontaneous aggregation of the starch components. The anti-staling effect of the branched chain products is caused by a decrease of or an interference with the crystallization (retrogradation) of amylopectin or from the interference with the formation of other interactions (Duedahl-Olesen et al. 1999). β -glucans, instead of forming a new flexible structure like CMC, interact in some manner to limit starch re-association which limits firming of corn tortillas.

Evaluation of texture parameters suggested that depending on the stage of storage, objective and subjective methods correlate differently. Overall, pliability was a better subjective measurement than rollability, as found in previous studies (Bueso-Ucles

2003, Fernandez et al. 1999). Final stiffness and Young's modulus had the highest correlation among objective measurements, and both had high correlations with pliability.

However, at 14 days of storage, pliability had good correlations with final stiffness and energy dissipated, but not better than the correlations of rollability with extensibility measurements. Extensibility test is a faster method than stress relaxation, so it is recommended as an objective measurement of corn tortilla texture in combination with rollability. It is suggested that depending on the time of texture analysis, the methods should be appropriately correlated for better results.

Barley is gaining renewed interest as a food component because of its soluble dietary fiber and β -glucan content in particular (Marconi 2000). The use of β -glucan in corn tortillas offers nutritional and functional properties as well. This additive is marketed as a nutraceuticals product. β -glucans are known for reducing serum cholesterol, hypoglycemic and anti-carcinogenic effects (Bhatty 1999). The price of β -glucan is approximately \$2.50/lb, while CMC is about \$0.60/lb. Thus, β -glucans are not competitive with CMC as a functional ingredient. However, the low amounts of β -glucan needed may be comparable in functionality and cost with CMC. Increasing dietary fiber in foods by incorporating β -glucan enriched barley fractions could increase the use of such products in manufactured foods, thus improving the diet of the general population (Knuckles et al. 1997). Researchers have found that levels ranging from 0.1-2 mg/kg boost body immunity (Borek 2001). Tortillas with 2.5% barley flour or 0.24% β -glucan have 75 mg/tortilla. Thus, a 60 kg person needs 120 mg of β -glucan, requirement that is met with less than 2 tortillas.

Further study is suggested to determine the mechanism by which β -glucans retard staling of corn tortillas and to determine if β -glucans with different viscosities affect their functionality in corn tortillas.

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APPENDIX A

TABLE A.1
Effect of Barley Substitution and 4°C Storage on Rupture Force, Rupture Distance and Young's Modulus of Corn Tortillas^a

Barley Flour substitution	Rupture Force (N)			Rupture distance (mm)			Young's Modulus (N/s)		
	4 hr	1 d	7 d	4 hr	1 d	7 d	4 hr	1 d	7 d
Control	5.9 ^a	12.7 ^a	13.6 ^a	4.1 ^b	1.9 ^c	1.7 ^a	1.9 ^a	6.2 ^a	7.0 ^a
2.5	5.1 ^b	7.4 ^c	8.3 ^b	4.8 ^{ab}	2.0 ^{bc}	1.8 ^a	1.6 ^b	3.5 ^b	4.1 ^b
5.0	4.7 ^c	8.7 ^b	8.9 ^b	4.8 ^a	2.4 ^a	1.8 ^a	1.4 ^b	3.8 ^b	4.4 ^b
10.0	4.3 ^c	8.3 ^{bc}	8.9 ^b	4.0 ^b	2.3 ^{ab}	1.9 ^a	1.4 ^b	3.7 ^b	4.3 ^b
LSD^b($\alpha=0.05$)	0.37	1.05	1.22	0.75	0.31	0.26	0.17	0.37	0.36
Mean	5.00	9.25	9.90	4.41	2.13	1.76	1.54	4.31	4.96
LSD_{time}($\alpha=0.05$)		0.64			0.25			0.29	

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

APPENDIX B

TABLE B.1
Effect of Additives and Storage Time on Rollability of Corn Tortillas Stored at 4°C^a

Amylase (MAU)	Barley (%)	CMC (%)	Initial	1 day	4 days	7 days	14 days
0	0	0	5.0 ^a	4.4 ^{cd}	3.6 ^c	3.1 ^f	1.9 ^e
825	0	0.25	5.0 ^a	4.9 ^a	4.3 ^{ab}	4.3 ^{abc}	2.9 ^b
825	2.5	0.25	5.0 ^a	4.7 ^{ab}	3.9 ^{bc}	4.0 ^{cde}	3.3 ^{ab}
825	1.25	0	5.0 ^a	4.4 ^d	4.3 ^{ab}	4.0 ^{cde}	2.7 ^{bcd}
825	1.25	0.5	5.0 ^a	4.8 ^{ab}	4.5 ^a	4.2 ^{abc}	3.6 ^a
0	1.25	0.25	5.0 ^a	4.9 ^a	4.3 ^{ab}	4.1 ^{bcd}	2.8 ^{bc}
1650	1.25	0.25	5.0 ^a	4.6 ^{bc}	4.0 ^{bc}	3.8 ^{de}	2.3 ^{cde}
1650	0	0	5.0 ^a	4.6 ^{bc}	4.3 ^{ab}	3.6 ^c	2.1 ^{de}
0	0	0.5	5.0 ^a	4.9 ^a	4.3 ^{ab}	4.5 ^{ab}	3.8 ^a
0	2.5	0	5.0 ^a	4.9 ^a	3.9 ^{bc}	3.7 ^e	2.0 ^e
1650	2.5	0.5	5.0 ^a	4.9 ^a	4.5 ^a	4.5 ^a	3.0 ^b
825	1.25	0.25	5.0 ^a	4.9 ^a	4.4 ^{ab}	3.8 ^{de}	3.0 ^b
LSD^b($\alpha=0.05$)			0	0.23	0.47	0.40	0.59
Mean			5.0	4.7	4.2	4.0	2.8
LSD_{time}($\alpha=0.05$)					0.14		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE B.2
Effect of Additives and Storage Time on Pliability of Corn Tortillas Stored at 4°C^a

Amylase (MAU)	Barley (%)	CMC (%)	Initial	1 day	4 days	7 days	14 days
0	0	0	5.0 ^a	3.6 ^d	3.5 ^c	3.0 ^b	1.3 ^d
825	0	0.25	5.0 ^a	4.3 ^{bc}	3.9 ^{abc}	2.9 ^b	2.8 ^c
825	2.5	0.25	5.0 ^a	4.4 ^{abc}	4.0 ^{abc}	3.0 ^b	3.1 ^{ab}
825	1.25	0	5.0 ^a	4.1 ^c	3.6 ^{bc}	3.0 ^b	3.0 ^{abc}
825	1.25	0.5	5.0 ^a	4.6 ^a	4.2 ^{ab}	3.9 ^a	3.4 ^a
0	1.25	0.25	5.0 ^a	4.4 ^{abc}	4.2 ^{ab}	3.9 ^a	3.3 ^{ab}
1650	1.25	0.25	5.0 ^a	4.4 ^{abc}	3.9 ^{abc}	3.9 ^a	3.3 ^{ab}
1650	0	0	5.0 ^a	4.5 ^{ab}	3.6 ^{bc}	3.1 ^b	2.5 ^c
0	0	0.5	5.0 ^a	4.6 ^a	4.4 ^a	3.9 ^a	3.1 ^{ab}
0	2.5	0	5.0 ^a	4.3 ^{bc}	3.8 ^{abc}	3.6 ^a	2.9 ^{abc}
1650	2.5	0.5	5.0 ^a	4.3 ^{bc}	4.3 ^{ab}	4.0 ^a	3.2 ^{ab}
825	1.25	0.25	5.0 ^a	4.4 ^{abc}	3.4 ^c	3.1 ^b	2.9 ^{abc}
LSD^b($\alpha=0.05$)			0.013	0.28	0.68	0.41	0.52
Mean			5.0	4.3	3.9	3.4	2.9
LSD_{time}($\alpha=0.05$)					0.15		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE B.3
Effect of Additives and Storage Time on Rupture Force (N) of Corn Tortillas Stored at 4°C^a

Amylase (MAU)	Barley (%)	CMC (%)	Initial	1 day	4 days	7 days	14 days
0	0	0	5.50 ^a	12.64 ^a	13.27 ^a	12.49 ^a	13.26 ^a
825	0	0.25	4.61 ^{cde}	11.95 ^{ab}	11.93 ^{ab}	12.31 ^b	12.36 ^{abc}
825	2.5	0.25	4.64 ^{cd}	10.22 ^c	10.46 ^{cd}	10.20 ^{cd}	11.28 ^{def}
825	1.25	0	4.69 ^{cd}	10.17 ^{abc}	10.38 ^{cd}	11.21 ^{cd}	11.40 ^{cdef}
825	1.25	0.5	5.22 ^{ab}	11.11 ^{abc}	11.36 ^{bc}	11.19 ^b	12.11 ^{bcd}
0	1.25	0.25	4.84 ^{bc}	11.98 ^{abc}	11.61 ^{ab}	11.68 ^b	12.74 ^{ab}
1650	1.25	0.25	4.64 ^{cd}	10.16 ^{abc}	11.15 ^{cd}	11.17 ^{bc}	11.84 ^{bcde}
1650	0	0	3.46 ^g	9.49 ^{bc}	10.33 ^d	10.52 ^d	11.36 ^{def}
0	0	0.5	4.54 ^{cde}	12.32 ^{abc}	12.82 ^a	11.51 ^a	12.67 ^{ab}
0	2.5	0	3.96 ^f	10.94 ^{abc}	10.49 ^{bc}	11.55 ^{cd}	12.16 ^{bcd}
1650	2.5	0.5	4.31 ^{def}	10.81 ^c	11.35 ^c	9.97 ^b	10.70 ^f
825	1.25	0.25	4.16 ^{ef}	10.70 ^{abc}	10.45 ^c	11.28 ^{cd}	11.05 ^{ef}
LSD^b($\alpha=0.05$)			0.47	1.83	1.07	0.79	0.96
Mean			4.51	11.04	11.30	11.26	11.78
LSD_{time}($\alpha=0.05$)					0.39		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE B.4
Effect of Additives and Storage Time on Rupture Distance (mm) of Corn Tortillas Stored at 4°C^a

Amylase (MAU)	Barley (%)	CMC (%)	Initial	1 day	4 days	7 days	14 days
0	0	0	4.50 ^{fg}	1.86 ^{abc}	1.77 ^{bc}	1.77 ^{abc}	1.74 ^{abcd}
825	0	0.25	5.40 ^{bcde}	2.05 ^a	1.94 ^{ab}	2.12 ^a	1.98 ^a
825	2.5	0.25	4.89 ^{ef}	1.85 ^{abc}	1.65 ^{cd}	1.56 ^c	1.79 ^{abcd}
825	1.25	0	4.11 ^g	1.58 ^c	1.48 ^d	1.64 ^{bc}	1.60 ^d
825	1.25	0.5	5.92 ^{abc}	1.84 ^{abc}	1.72 ^{bcd}	1.76 ^{abc}	1.91 ^{ab}
0	1.25	0.25	5.77 ^{bcd}	1.88 ^{ab}	1.62 ^{cd}	1.71 ^b	1.80 ^{abcd}
1650	1.25	0.25	5.52 ^{bcde}	1.62 ^{bc}	1.87 ^{bc}	1.81 ^{abc}	1.62 ^{cd}
1650	0	0	4.53 ^{fg}	1.57 ^c	1.62 ^{cd}	1.52 ^c	1.68 ^{abcd}
0	0	0.5	6.53 ^a	2.06 ^a	2.20 ^a	1.97 ^{ab}	1.87 ^{abc}
0	2.5	0	5.16 ^{def}	1.65 ^{bc}	1.61 ^{cd}	1.85 ^{abc}	1.66 ^{abcd}
1650	2.5	0.5	6.13 ^{ab}	2.10 ^a	2.15 ^a	1.60 ^{bc}	1.76 ^{abcd}
825	1.25	0.25	5.95 ^{abc}	1.81 ^{abc}	1.83 ^{bc}	1.83 ^{abc}	1.62 ^{cd}
LSD^b($\alpha=0.05$)			0.69	0.30	0.26	0.37	0.27
Mean			5.40	1.82	1.79	1.76	1.73
LSD_{time}($\alpha=0.05$)					0.15		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE B.5
Effect of Additives and Storage Time on Young's Modulus (N/s) of Corn Tortillas Stored at 4°C^a

Amylase (MAU)	Barley (%)	CMC (%)	Initial	1 day	4 days	7 days	14 days
0	0	0	0.90 ^a	3.58 ^a	3.87 ^a	3.59 ^a	3.78 ^a
825	0	0.25	0.66 ^d	3.21 ^{bc}	3.22 ^{cd}	3.23 ^{bc}	3.44 ^{bcd}
825	2.5	0.25	0.73 ^c	2.93 ^{ed}	3.04 ^{de}	3.21 ^{bc}	3.32 ^{cd}
825	1.25	0	0.82 ^b	3.22 ^{bc}	3.26 ^{cd}	3.38 ^{ab}	3.69 ^{ab}
825	1.25	0.5	0.68 ^{cd}	3.24 ^{bc}	3.39 ^{bc}	3.21 ^{bc}	3.44 ^{bcd}
0	1.25	0.25	0.66 ^d	3.44 ^{ab}	3.51 ^b	3.43 ^{ab}	3.71 ^{ab}
1650	1.25	0.25	0.66 ^d	3.33 ^{bc}	3.10 ^{de}	3.20 ^{bc}	3.76 ^{ab}
1650	0	0	0.59 ^e	3.14 ^{cd}	3.08 ^{de}	3.33 ^{bc}	3.62 ^{abc}
0	0	0.5	0.55 ^e	3.34 ^{abc}	3.20 ^{cd}	3.33 ^{bc}	3.65 ^{ab}
0	2.5	0	0.59 ^e	3.36 ^{abc}	3.11 ^{de}	3.24 ^{bc}	3.75 ^{ab}
1650	2.5	0.5	0.56 ^e	2.81 ^e	2.82 ^f	3.10 ^c	3.29 ^d
825	1.25	0.25	0.57 ^e	3.18 ^c	2.93 ^{ef}	3.23 ^b	3.50 ^{abcd}
LSD^b($\alpha=0.05$)			0.05	0.25	0.23	0.25	0.33
Mean			0.66	3.23	3.21	3.29	3.54
LSD_{time}($\alpha=0.05$)					0.10		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE B.6
Effect of Additives and Storage Time on Final Stiffness (10^5 Pa) of Corn Tortillas Stored at $4^\circ\text{C}^{\text{a}}$

Amylase (MAU)	Barley (%)	CMC (%)	Initial	7 days	14 days
0	0	0	9.64 ^{cd}	33.09 ^a	32.10 ^{ab}
825	0	0.25	11.64 ^{de}	27.67 ^c	32.42 ^{ab}
825	2.5	0.25	13.94 ^{abc}	25.09 ^e	27.35 ^e
825	1.25	0	13.80 ^{abcd}	26.87 ^{cd}	30.02 ^{cd}
825	1.25	0.5	12.32 ^{abcd}	27.81 ^c	32.06 ^{ab}
0	1.25	0.25	13.22 ^{cd}	30.68 ^b	33.71 ^a
1650	1.25	0.25	10.55 ^{ab}	26.92 ^{cd}	30.79 ^{bc}
1650	0	0	8.75 ^e	27.21 ^{cd}	29.71 ^{cd}
0	0	0.5	9.41 ^e	28.05 ^c	31.41 ^{bc}
0	2.5	0	9.11 ^{bcd}	24.16 ^{ef}	30.47 ^{bc}
1650	2.5	0.5	12.12 ^a	23.32 ^f	27.20 ^e
825	1.25	0.25	11.91 ^{abcd}	25.68 ^{de}	28.26 ^{de}
LSD^b($\alpha=0.05$)			0.33	1.59	1.98
Mean			3.32	27.21	30.11
LSD_{time}($\alpha=0.05$)				0.78	

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE B.7
Effect of Additives and Storage Time on Energy Dissipated (10^{-4} J/m³) of Corn Tortillas Stored at 4°C^a

Amylase (MAU)	Barley (%)	CMC (%)	Initial	7 days	14 days
0	0	0	8.67 ^a	0.40 ^{cd}	0.46 ^a
825	0	0.25	3.82 ^g	0.47 ^{bcd}	0.38 ^{bcd}
825	2.5	0.25	4.57 ^{fg}	0.62 ^a	0.40 ^{abcd}
825	1.25	0	5.56 ^{cde}	0.53 ^{ab}	0.38 ^{cd}
825	1.25	0.5	4.88 ^{ef}	0.47 ^{bcd}	0.36 ^d
0	1.25	0.25	6.54 ^b	0.37 ^d	0.35 ^d
1650	1.25	0.25	4.55 ^{fg}	0.51 ^{abc}	0.38 ^{cd}
1650	0	0	6.08 ^{cb}	0.37 ^d	0.42 ^{abcd}
0	0	0.5	4.28 ^{fg}	0.43 ^{bcd}	0.45 ^{abc}
0	2.5	0	5.90 ^{bcd}	0.48 ^{bcd}	0.44 ^{abc}
1650	2.5	0.5	5.02 ^{def}	0.49 ^{bcd}	0.46 ^a
825	1.25	0.25	4.37 ^{fg}	0.45 ^{bcd}	0.45 ^a
LSD^b($\alpha=0.05$)			0.89	0.12	0.07
Mean			5.30	0.47	0.24
LSD_{time}($\alpha=0.05$)				0.24	

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE B.8
ANOVA Summary for Masa and Tortilla Moisture Content and Ph

Source	Masa moisture (%)		Tortilla moisture (%)		pH	
	F Value	Pr > F	F Value	Pr > F	F Value	Pr > F
Model	1.08	0.4029	0.66	0.7746	1.56	0.2171
Block	4.58	0.0386*	0.02	0.8915	5.78	0.0319*
Amylase	0.72	0.4939	1.42	0.2547	0.63	0.5471
Barley	1.1	0.3425	0.68	0.5133	0.29	0.7563
CMC	0.53	0.5931	0.14	0.8697	5.58	0.0178*
Barley*CMC	0.91	0.4659	0.87	0.4931	0	1
Amylase*Barley*CMC	0.02	0.8934	0.01	0.9294	0	1

*Statistically significant; **Highly significant

TABLE B.9
ANOVA Summary for Yield, Tortilla Weight and Thickness of Corn Tortillas at 4 hr

Source	Yield (kg/kg flour)		Tortilla weight (g)		Thickness (mm)	
	F Value	Pr > F	F Value	Pr > F	F Value	Pr > F
Model	1.65	0.1921	2.41	0.065	1.39	0.2836
Block	11.87	0.0087*	17.42	0.0011*	1.13	0.3078
Amylase	0.01	0.9925	2.03	0.1713	0.51	0.6143
Barley	1	0.3957	0.19	0.828	1.35	0.2935
CMC	1.55	0.2499	0.79	0.4764	0.35	0.7096
Barley*CMC	0.64	0.6402	1.36	0.3017	2.3	0.1134
Amylase*Barley*CMC	0.21	0.6538	0.03	0.8645	1.86	0.1953

*Statistically significant; **Highly significant

TABLE B.10
ANOVA Summary for Rollability and Pliability Score of Corn Tortillas Stored for 14 Days at 4°C

Source	Rollability score		Pliability score	
	F Value	Pr > F	F Value	Pr > F
Model	8.22	<0.001**	23.7	<0.001**
Block	3.47	0.068	32.52	<0.001**
Amylase	10.74	<0.001**	14.54	<0.001**
Barley	0.08	0.9216	39.34	<0.001**
CMC	31.7	<0.001**	30.13	<0.001**
Barley*CMC	1.72	0.15777	13.99	<0.001**
Amylase*Barley*CMC	3.24	0.07	27.9	<0.001**

*Statistically significant; **Highly significant

TABLE B.11
ANOVA Summary for Rupture Force, Rupture Distance and Young's Modulus of Corn Tortillas Measured 4 hr after Production

Source	Rupture force (N)		Rupture distance (mm)		Young's modulus (N/s)	
	F Value	Pr > F	F Value	Pr > F	F Value	Pr > F
Model	5.97	<0.0001**	6.03	<0.0001**	35.2	<0.0001**
Block	1.72	0.1937	0	0.9525	3.16	0.079
Amylase	8.36	0.0005*	0.12	0.8873	24.41	<0.0001**
Barley	2.93	0.0588	0.19	0.8295	5.24	0.0072**
CMC	5.68	0.0049*	3.99	0.0222*	31.24	<0.0001**
Barley*CMC	8.87	<0.0001**	12.46	<0.0001**	62.73	<0.0001**
Amylase*Barley*CMC	0.49	0.4853	13.93	0.0003*	46.53	<0.0001**

*Statistically significant; **Highly significant

TABLE B.12
ANOVA Summary for Rupture Force, Rupture Distance and Young's Modulus of Corn Tortillas Stored for 14 Days at 4°C

Source	Rupture force (N)		Rupture distance (mm)		Young's modulus (N/s)	
	F Value	Pr > F	F Value	Pr > F	F Value	Pr > F
Model	8.82	<0.0001**	1.55	0.1223	5.18	<0.0001**
Block	50.17	<0.0001**	0.33	0.5656	37.73	<0.0001**
Amylase	17.03	<0.0001**	0.97	0.3847	4.91	0.0096*
Barley	5.09	0.0082*	1.8	0.1724	3.16	0.0475*
CMC	0.29	0.7525	3.94	0.0233*	2.94	0.0584
Barley*CMC	2.18	0.0788	1.12	0.3522	0.38	0.82
Amylase*Barley*CMC	2.16	0.145	0.41	0.5218	0.84	0.36

*Statistically significant; **Highly significant

TABLE B.13
ANOVA Summary for Final Stiffness and Energy Dissipated of Corn Tortillas Measured at 4 hr after Production

Source	Final stiffness (10^5 Pa)		Energy dissipated (10^{-4} J/m ³)	
	F Value	Pr > F	F Value	Pr > F
Model	2.96	0.0018*	11.99	<0.0001**
Block	3.78	0.0553	0.82	0.3669
Amylase	2.24	0.113	32.29	<0.0001**
Barley	8.81	0.0003*	2.57	0.0823
CMC	1.24	0.2957	14.22	<0.0001**
Barley*CMC	1.46	0.2209	9.17	<0.0001**
Amylase*Barley*CMC	1.34	0.2495	8.14	0.0055*

*Statistically significant; **Highly significant

TABLE B.14
ANOVA Summary for Final Stiffness and Energy Dissipated of Corn Tortillas Stored for 14 Days at 4°C

Source	Final stiffness (10^5 Pa)		Energy dissipated (10^{-4} J/m ³)	
	F Value	Pr > F	F Value	Pr > F
Model	2.96	0.0018*	11.99	<0.0001**
Block	3.78	0.0553	0.82	0.3669
Amylase	2.24	0.113	32.29	<0.0001**
Barley	8.81	0.0003*	2.57	0.0823
CMC	1.24	0.2957	14.22	<0.0001**
Barley*CMC	1.46	0.2209	9.17	<0.0001**
Amylase*Barley*CMC	1.34	0.2495	8.14	0.0055*

*Statistically significant; **Highly significant

TABLE B.15
Statistical Parameter Estimates from RSREG Procedure

	Rollability	Pliability	Rupture force	Rupture distance	Young's modulus	Final stiffness	Energy dissipated
Intercept	1.915866**	1.396712**	12.69974**	1.755221**	3.761474**	31.90331**	0.46321**
Amylase	0.001353*	0.000497	-0.00162*	0.00011	-0.000562*	-0.00347*	-3.4E-05
CMC	2.414823*	3.491336*	0.83499	0.465683	-0.410862	-0.19939	-0.11935
Barley	-0.05704	0.978267**	-0.14508	-0.21567*	0.252886	1.214334	-0.05601
Amy*Amy	-7.4E-07*	1.22E-07	5.3E-07	-8.6E-08	0.000000292*	1.43E-06	-5.7E-09
Amy*CMC	-0.00222*	-0.00122*	0.001943	0.000193	0.000137	0.006407*	6.43E-05
CMC*CMC	2.514614	-0.07474	-1.35203	-0.37302	0.398755	-0.62635	0.12705
Amy*Bar	0.000138	-0.00017	-0.00024	-8.8E-05	0.000039704	-0.00157*	3.16E-05
CMC*Bar	0.136534	-0.80685*	-0.58843	0.1405	-0.378952	0.027979	-0.04693
Bar*Bar	0.036585	-0.14699*	-0.01052	0.074881	-0.102223	-0.67808*	0.015848

*Statistically significant; **Highly significant

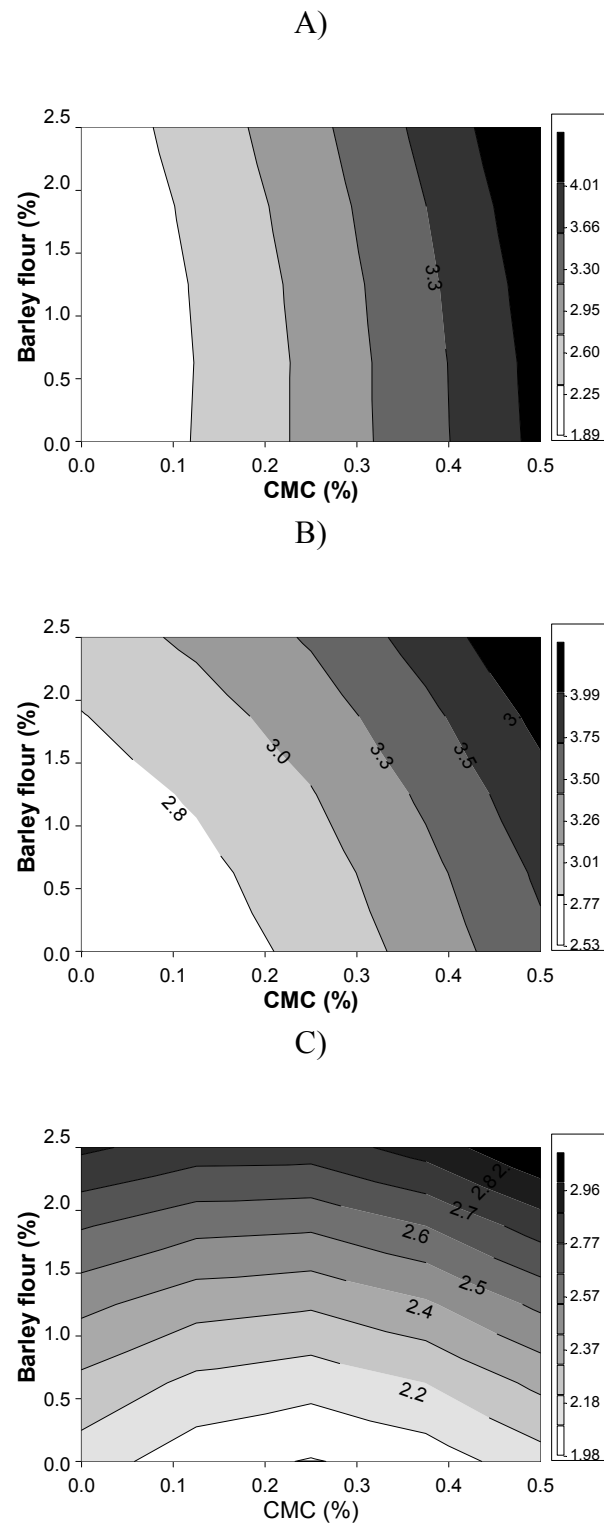


Fig. B. 1. Effect of CMC and Barley Flour on Rollability of Corn Tortillas Stored at 4C for 14 Days. A) 0 MAU, B) 825 MAU and C) 1650 MAU. $LSD(\alpha=0.05)=0.60$.

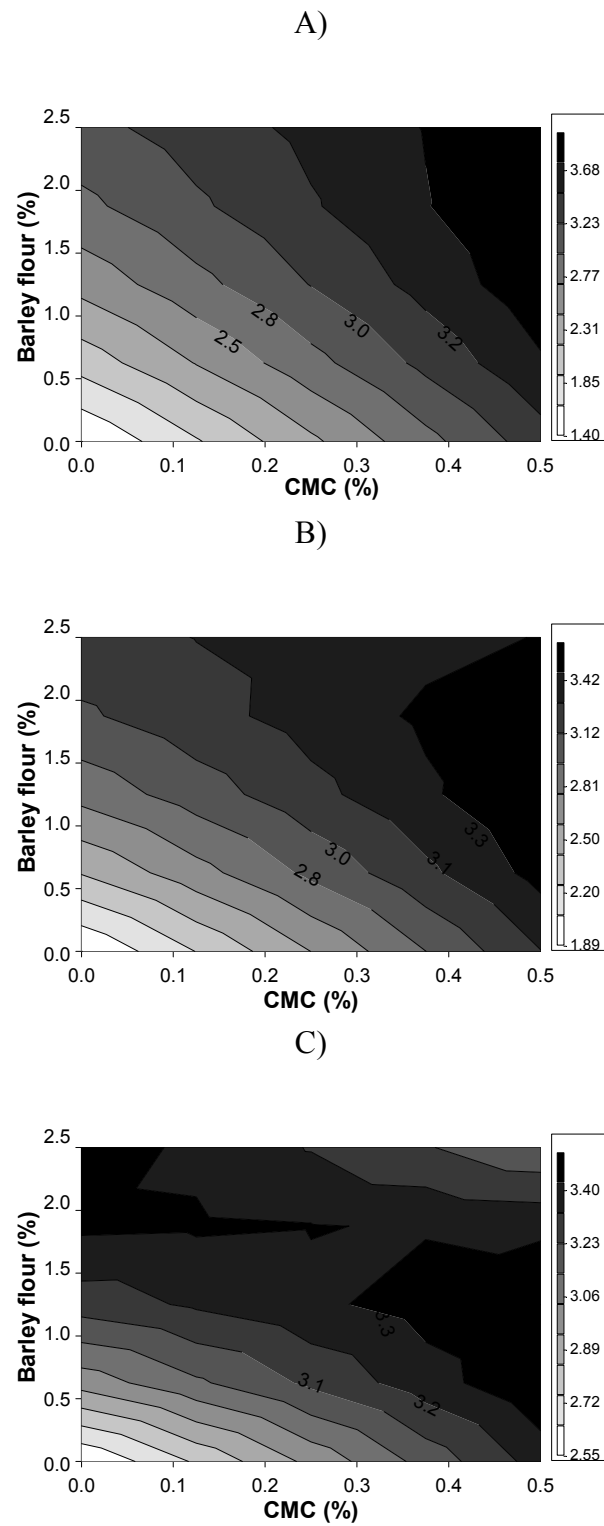


Fig. B. 2. Effect of CMC and Barley Flour on Pliability of Corn Tortillas Stored at 4C for 14 Days. A) 0 MAU, B) 825 MAU and C) 1650 MAU. $LSD(\alpha=0.05)=0.52$.

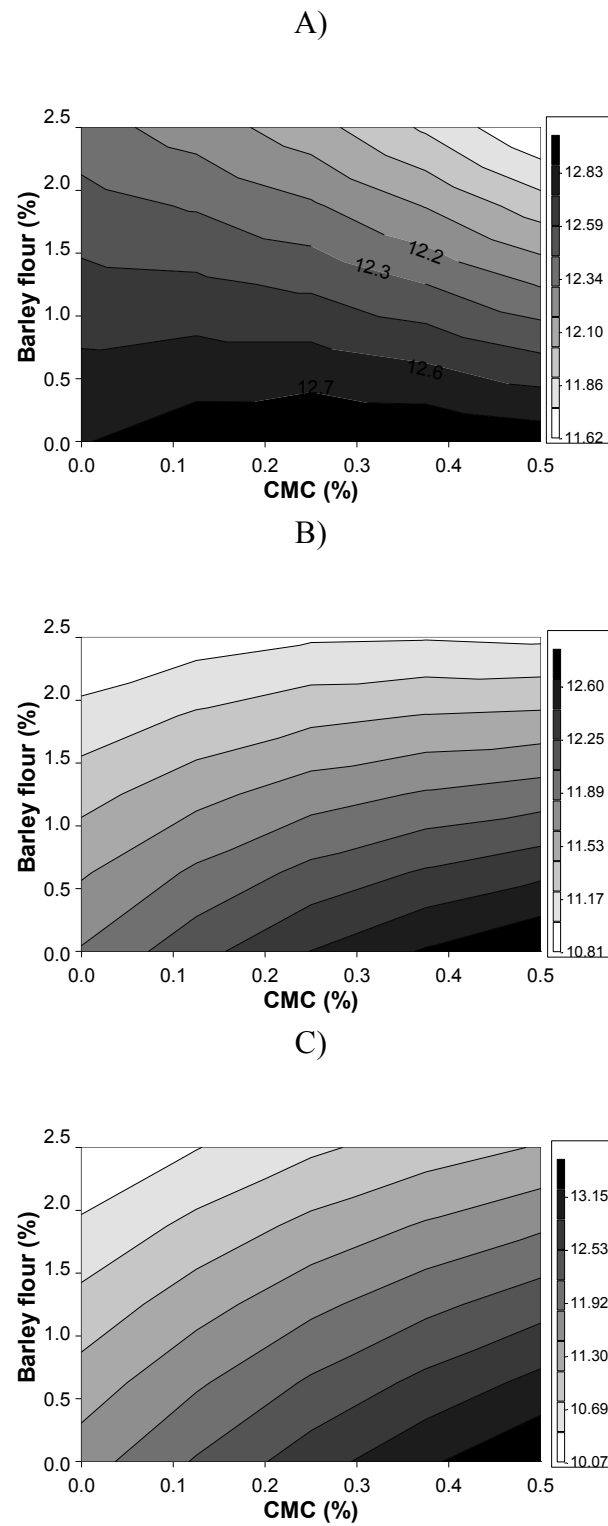


Fig. B. 3. Effect of CMC and Barley Flour on Rupture Force (N) of Corn Tortillas Stored at 4C for 14 Days. A) 0 MAU, B) 825 MAU and C) 1650 MAU. $LSD(\alpha=0.05)=0.96$.

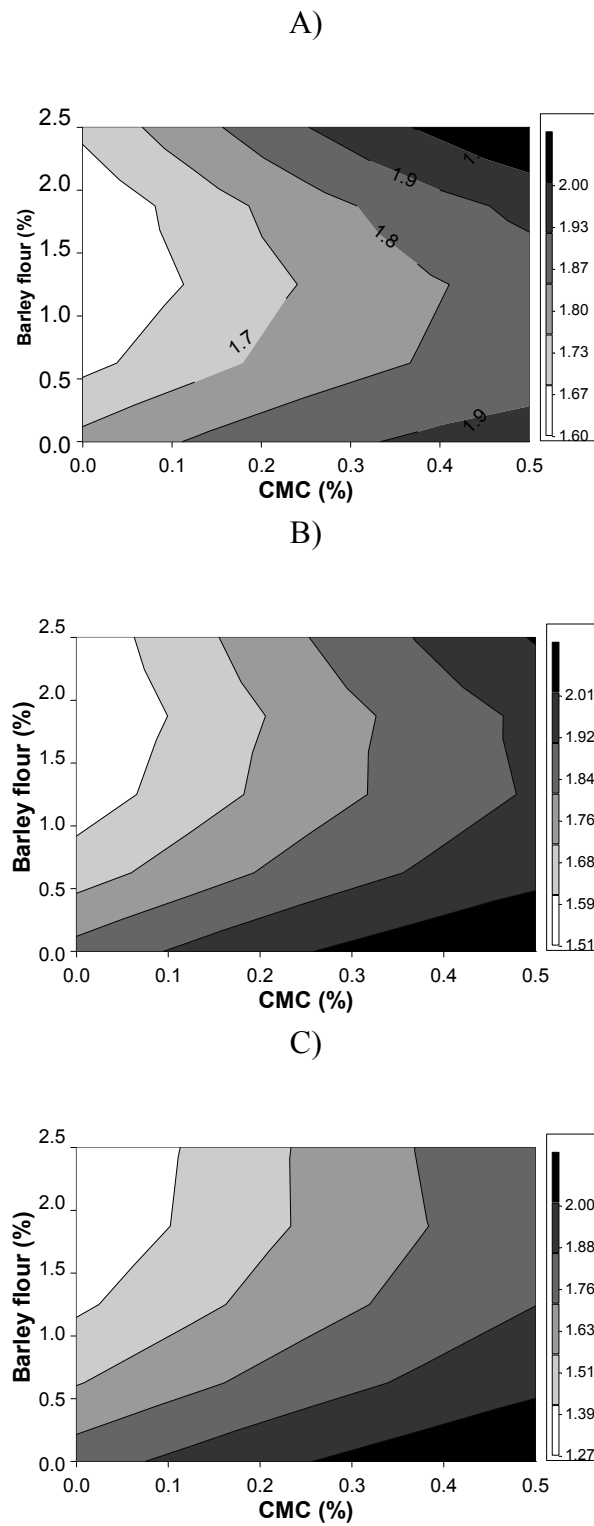


Fig. B. 4. Effect of CMC and Barley Flour on Rupture Distance (mm) of Corn Tortillas Stored at 4C for 14 Days. A) 0 MAU, B) 825 MAU and C) 1650 MAU. $LSD(\alpha=0.05)=0.27$.

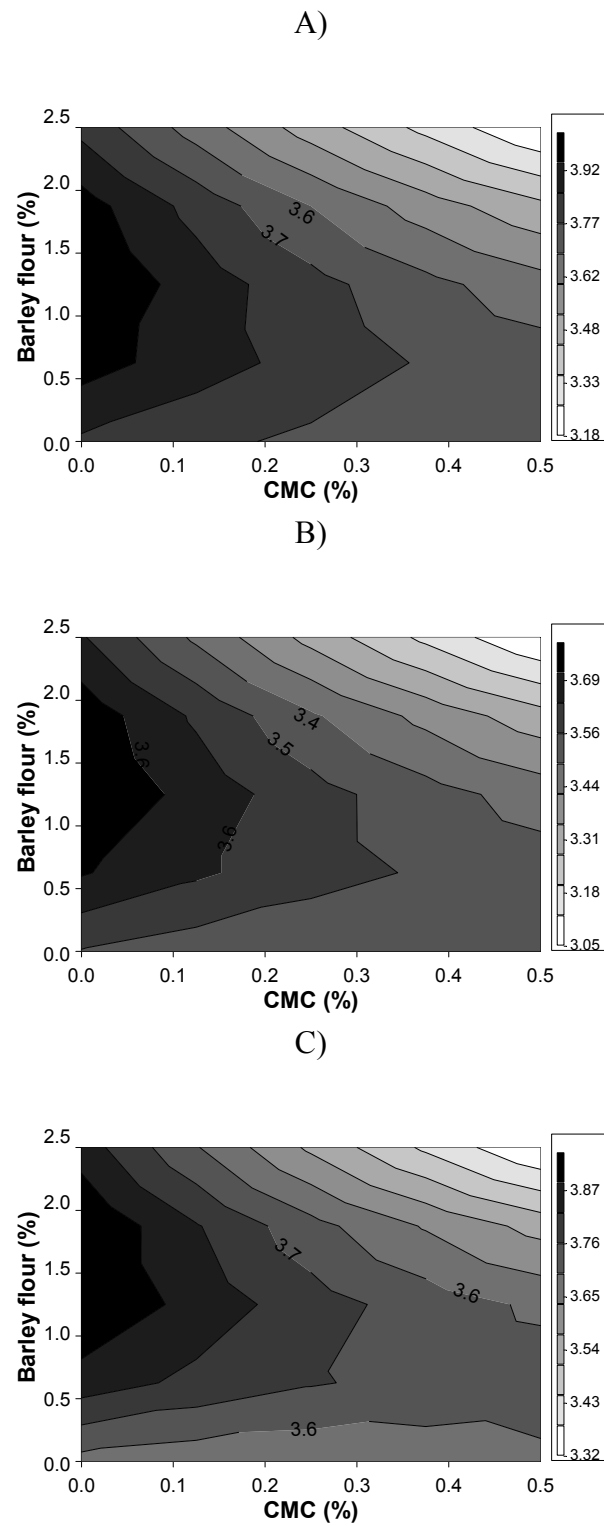


Fig. B. 5. Effect of CMC and Barley Flour on Young's Modulus (N/S) of Corn Tortillas Stored at 4C for 14 Days. A) 0 MAU, B) 825 MAU and C) 1650 MAU. $LSD(\alpha=0.05)=0.33$.

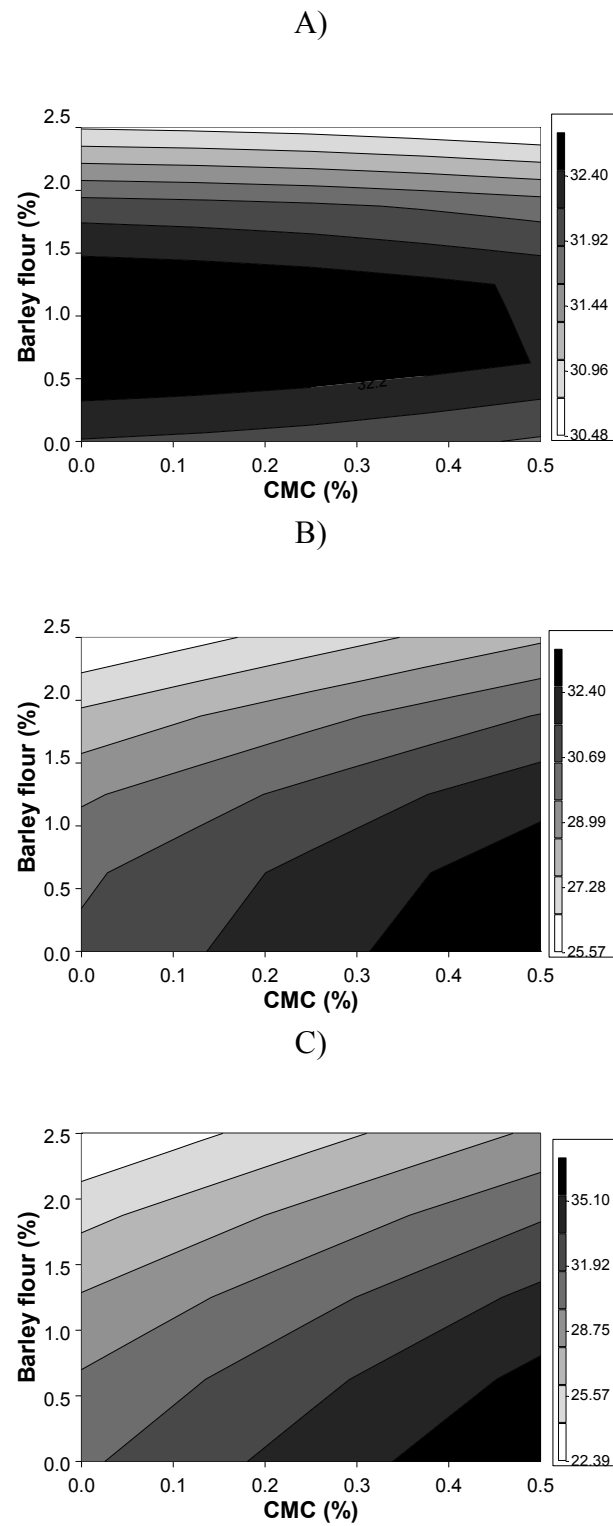


Fig. B. 6. Effect of CMC and Barley Flour on Final Stiffness (10^5 Pa) of Corn Tortillas Stored at 4C for 14 Days. A) 0 MAU, B) 825 MAU and C) 1650 MAU. $LSD(\alpha=0.05)=1.98$.

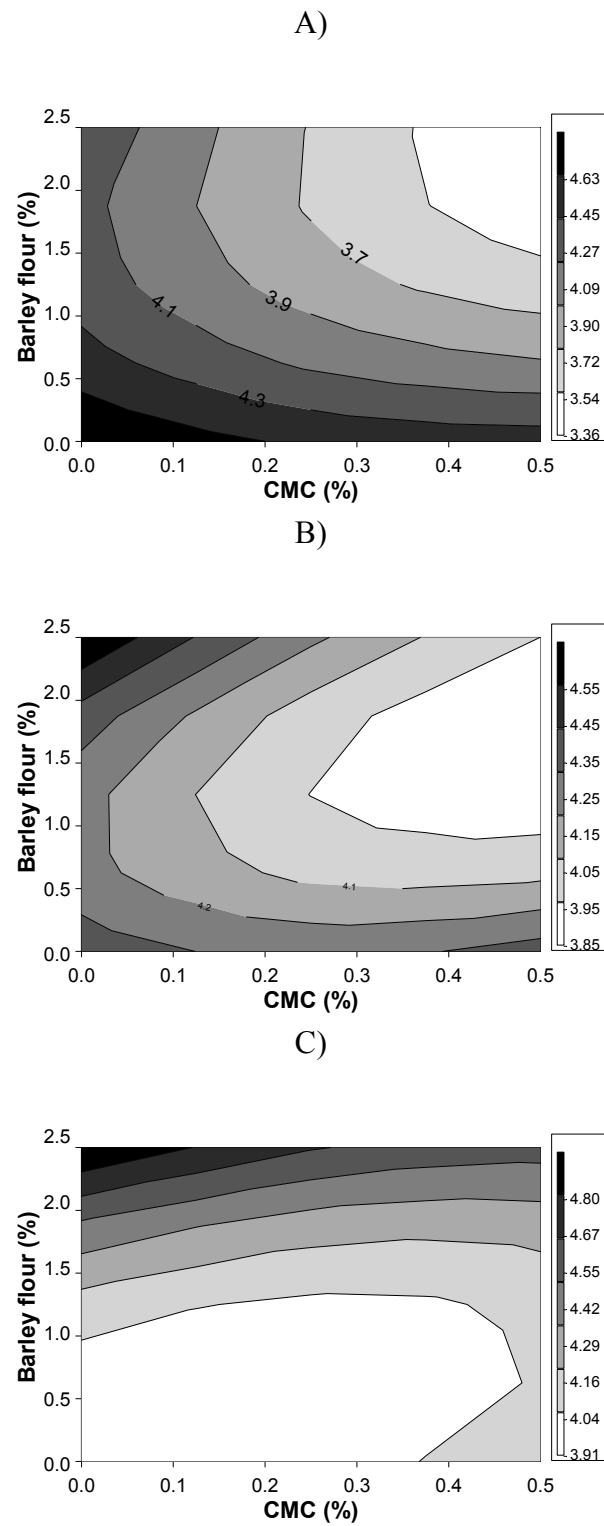


Fig. B. 7. Effect Of CMC And Barley Flour On Energy Dissipated (10^{-4} J/M³) Of Corn Tortillas Stored At 4C For 14 Days. A) 0 MAU, B) 825 MAU And C) 1650 MAU. LSD($\alpha=0.05$)=0.74.

APPENDIX C

TABLE C.1
Effect of β -glucan, Amylase and CMC on Rollability Score of Corn Tortillas
throughout Storage for 14 Days at 4°C^a

Barley β -glucan (%)	4 hr	1d	4d	7d	14d
DMF Control	5.0 ^a	4.5 ^c	3.0 ^e	2.4 ^f	2.8 ^d
Barley Flour Control	5.0 ^a	4.6 ^{abc}	3.4 ^{de}	3.6 ^{cd}	3.1 ^{cd}
0.12% β -glucan	5.0 ^a	4.7 ^{abc}	4.1 ^{ab}	3.6 ^{cd}	3.5 ^{abc}
0.24% β -glucan	5.0 ^a	4.6 ^{abc}	3.9 ^{bc}	3.6 ^{de}	3.2 ^{bcd}
0.96% β -glucan	5.0 ^a	4.9 ^a	4.2 ^{ab}	4.1 ^{abcd}	3.8 ^{abc}
2.4% β -glucan	5.0 ^a	4.8 ^{ab}	4.3 ^{ab}	4.3 ^a	3.9 ^{ab}
0.5% CMC	5.0 ^a	4.9 ^a	4.6 ^a	4.2 ^{ab}	4.1 ^a
With 550 MAU amylase					
0.12% β -glucan	5.0 ^a	4.8 ^a	3.5 ^{cd}	3.1 ^e	3.8 ^{abc}
0.24% β -glucan	5.0 ^a	4.6 ^{abc}	4.1 ^{ab}	3.7 ^{bcd}	3.2 ^{bcd}
0.96% β -glucan	5.0 ^a	4.9 ^a	3.9 ^{bc}	4.0 ^{abcd}	3.4 ^{abcd}
2.4% β -glucan	5.0 ^a	4.5 ^{bc}	4.3 ^{ab}	4.0 ^{abcd}	3.6 ^{abc}
0.5% CMC	5.0 ^a	4.8 ^{ab}	4.1 ^{ab}	4.1 ^{abc}	3.6 ^{abc}
LSD^b($\alpha=0.05$)	0	0.28	0.47	0.54	0.71
Mean	5.0	4.7	3.9	3.7	3.5
LSD_{time}($\alpha=0.05$)	0.15				

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE C.2
Effect of β -glucan, Amylase and CMC on Pliability Score of Corn Tortillas
throughout Storage for 14 Days at 4°C^a

	4 hr	1d	4d	1week	2week
DMF Control	5.0 ^a	4.2 ^c	3.0 ^{bc}	2.1 ^e	1.9 ^c
Barley Flour Control	5.0 ^a	4.3 ^{bc}	3.5 ^{ab}	2.9 ^{cd}	2.8 ^{ab}
0.12% β -glucan	5.0 ^a	4.3 ^{bc}	3.8 ^{ab}	3.6 ^{ab}	3.1 ^{ab}
0.24% β -glucan	5.0 ^a	4.3 ^{bc}	3.7 ^{ab}	3.1 ^{abcd}	2.5 ^{bc}
0.96% β -glucan	5.0 ^a	4.3 ^{bc}	3.6 ^{ab}	3.4 ^{abc}	3.3 ^a
2.4% β -glucan	4.8 ^b	4.3 ^{bc}	3.9 ^a	3.6 ^a	3.0 ^{ab}
0.5% CMC	5.0 ^a	4.5 ^a	3.9 ^a	3.5 ^{abc}	3.3 ^a
With 550 MAU amylase					
0.12% β -glucan	5.0 ^a	4.3 ^{bc}	2.6 ^c	2.3 ^e	2.4 ^{bc}
0.24% β -glucan	5.0 ^a	4.4 ^{ab}	3.4 ^{abc}	2.6 ^{de}	2.5 ^{bc}
0.96% β -glucan	4.9 ^{ab}	4.4 ^{ab}	3.6 ^{ab}	3.0 ^{bcd}	2.8 ^{ab}
2.4% β -glucan	5.0 ^a	4.3 ^{bc}	3.8 ^{ab}	3.4 ^{abc}	3.0 ^{ab}
0.5% CMC	5.0 ^a	4.5 ^{ab}	3.9 ^a	3.6 ^a	3.4 ^a
LSD^b($\alpha=0.05$)	0.12	0.19	0.80	0.60	0.73
Mean	5.0	4.3	3.6	3.1	2.8
LSD_{time}($\alpha=0.05$)	0.17				

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE C.3
Effect of β -glucan, Amylase and CMC on Rupture Force (N) of Corn Tortillas
throughout Storage for 14 Days at 4°C^a

	4 hr	1d	4d	7d	14d
DMF Control	4.97 ^{bc}	11.76 ^{bcd}	12.26 ^{ef}	12.88 ^{cd}	13.66 ^{bcd}
Barley Flour Control	4.91 ^c	11.06 ^{cd}	11.68 ^f	12.14 ^d	12.74 ^f
0.12% β -glucan	5.39 ^{abc}	12.82 ^{ab}	13.38 ^{bcd}	13.78 ^{bc}	14.07 ^{bcd}
0.24% β -glucan	4.95 ^{bc}	11.71 ^{bcd}	13.30 ^{cde}	13.93 ^{bc}	14.01 ^{bcd}
0.96% β -glucan	5.02 ^{abc}	12.72 ^{ab}	14.58 ^{ab}	15.46 ^a	14.08 ^{bcd}
2.4% β -glucan	5.49 ^{abc}	13.11 ^a	14.97 ^a	14.15 ^{abc}	14.49 ^{ab}
0.5% CMC	5.33 ^{abc}	11.76 ^{bcd}	13.04 ^{de}	13.33 ^{cd}	13.63 ^{bcd}
With 550 MAU amylase					
0.12% β -glucan	4.79 ^c	10.53 ^d	12.23 ^{ef}	12.14 ^d	13.19 ^{def}
0.24% β -glucan	5.30 ^{abc}	11.29 ^{cd}	12.51 ^{ef}	12.76 ^{cd}	13.57 ^{cdef}
0.96% β -glucan	5.95 ^a	13.11 ^a	13.75 ^{bcd}	14.80 ^{ab}	14.37 ^{abc}
2.4% β -glucan	5.86 ^{ab}	11.88 ^{abc}	14.26 ^{abc}	13.60 ^{cd}	15.09 ^a
0.5% CMC	5.08 ^{abc}	10.82 ^{cd}	12.42 ^{ef}	12.23 ^d	13.12 ^{ef}
LSD^b($\alpha=0.05$)	0.94	1.27	1.20	1.47	0.90
Mean	5.25	11.88	13.20	13.43	13.83
LSD_{time}($\alpha=0.05$)	0.38				

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE C.4
Effect of β -glucan, Amylase and CMC on Rupture Distance (mm) of Corn Tortillas throughout Storage for 14 Days at 4°C^a

	4 hr	1d	4d	7d	14d
DMF Control	3.70 ^d	1.90 ^c	1.70 ^{abcd}	1.87 ^{bcd}	1.68 ^{ab}
Barley Flour Control	4.22 ^{cd}	2.00 ^{bc}	1.46 ^d	1.69 ^d	1.50 ^b
0.12% β -glucan	4.97 ^{abc}	2.34 ^{ab}	1.71 ^{abc}	1.97 ^{abcd}	1.57 ^{ab}
0.24% β -glucan	4.56 ^{cd}	2.05 ^{bc}	1.64 ^{abcd}	2.08 ^{abc}	1.63 ^{ab}
0.96% β -glucan	3.54 ^d	2.19 ^{abc}	1.87 ^{ab}	2.25 ^a	1.65 ^{ab}
2.4% β -glucan	3.74 ^d	2.01 ^{bc}	1.89 ^a	1.87 ^{bcd}	1.66 ^{ab}
0.5% CMC	5.83 ^{ab}	2.48 ^a	1.83 ^{abc}	2.14 ^{ab}	1.88 ^a
With 550 MAU amylase					
0.12% β -glucan	4.83 ^{bc}	2.04 ^{bc}	1.61 ^{cd}	1.77 ^{cd}	1.62 ^{ab}
0.24% β -glucan	4.39 ^{cd}	2.10 ^{bc}	1.65 ^{abcd}	1.84 ^{bcd}	1.65 ^{ab}
0.96% β -glucan	3.73 ^d	2.24 ^{abc}	1.59 ^{cd}	1.88 ^{bcd}	1.53 ^b
2.4% β -glucan	3.92 ^{cd}	1.92 ^c	1.64 ^{bcd}	1.75 ^{cd}	1.64 ^{ab}
0.5% CMC	6.04 ^a	2.24 ^{abc}	1.72 ^{abc}	1.85 ^{bcd}	1.81 ^{ab}
LSD^b($\alpha=0.05$)	1.07	0.35	0.25	0.33	0.31
Mean	4.46	2.13	1.91	1.69	1.65
LSD_{time}($\alpha=0.05$)	0.18				

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE C.5
Effect of β -glucan, Amylase and CMC on Young's Modulus (N/s) of Corn Tortillas
throughout Storage for 14 Days at 4°C^a

	4 hr	1d	4d	7d	14d
DMF Control	1.01 ^{bcd}	3.45 ^{ab}	3.76 ^{ef}	3.69 ^{cde}	4.09 ^{de}
Barley Flour Control	0.84 ^{efg}	3.03 ^{cd}	3.83 ^{ef}	3.65 ^{de}	4.08 ^{de}
0.12% β -glucan	0.83 ^{efg}	3.27 ^{bc}	4.04 ^{bcd}	3.76 ^{cd}	4.54 ^{ab}
0.24% β -glucan	0.85 ^{def}	3.41 ^b	4.12 ^{bcd}	3.69 ^{cde}	4.29 ^{bcd}
0.96% β -glucan	1.06 ^{abc}	3.37 ^b	4.14 ^{bc}	3.78 ^{bcd}	4.31 ^{bcd}
2.4% β -glucan	1.16 ^{ab}	3.68 ^a	4.25 ^{ab}	3.99 ^{ab}	4.44 ^{abc}
0.5% CMC	0.75 ^{fg}	2.93 ^d	3.86 ^{cdef}	3.47 ^e	3.90 ^e
With 550 MAU amylase					
0.12% β -glucan	0.78 ^{efg}	2.99 ^d	3.85 ^{def}	3.64 ^{de}	4.21 ^{cd}
0.24% β -glucan	0.93 ^{cde}	3.09 ^{cd}	3.83 ^{ef}	3.63 ^{de}	4.34 ^{bcd}
0.96% β -glucan	1.20 ^a	3.37 ^b	4.31 ^{ab}	4.05 ^a	4.52 ^{ab}
2.4% β -glucan	1.11 ^{ab}	3.45 ^{ab}	4.41 ^a	3.89 ^{abc}	4.65 ^a
0.5% CMC	0.67 ^g	2.90 ^d	3.76 ^f	3.47 ^e	3.84 ^e
LSD^b($\alpha=0.05$)	0.17	0.24	0.28	0.23	0.30
Mean	0.93	3.25	3.73	4.01	4.27
LSD_{time}($\alpha=0.05$)	0.08				

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE C.6
Effect of β -glucan, Amylase and CMC on Final Stiffness (10^5 Pa) of Corn Tortillas throughout Storage for 14 Days at 4°C^a

	Initial	7d	14d
DMF Control	3.13 ^{bc}	32.81 ^{bcd}	32.83 ^{cde}
Barley Flour Control	3.27 ^{bc}	30.05 ^{de}	33.41 ^{bcd}
0.12% β -glucan	3.40 ^b	30.73 ^{cde}	33.85 ^{abcd}
0.24% β -glucan	3.00 ^c	33.50 ^{abc}	33.52 ^{bcd}
0.96% β -glucan	3.25 ^{bc}	33.76 ^{ab}	34.87 ^{abc}
2.4% β -glucan	3.47 ^b	32.34 ^{bcd}	35.62 ^{ab}
0.5% CMC	3.13 ^{bc}	28.86 ^e	30.76 ^e
With 550 MAU amylase			
0.12% β -glucan	3.11 ^{bc}	28.29 ^e	31.50 ^{de}
0.24% β -glucan	3.16 ^{bc}	33.25 ^{bc}	32.18 ^{de}
0.96% β -glucan	3.90 ^a	36.21 ^a	36.37 ^a
2.4% β -glucan	3.94 ^a	32.16 ^{bcd}	35.00 ^{abc}
0.5% CMC	3.26 ^{bc}	28.11 ^e	26.90 ^f
LSD^b($\alpha=0.05$)	0.37	2.94	1.76
Mean	3.33	31.67	33.06
LSD_{time}($\alpha=0.05$)		0.71	

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE C.7
Effect of β -glucan, Amylase and CMC on Energy Dissipated (10^{-4} J/m³) of Corn Tortillas throughout Storage for 14 Days at 4°C^a

	4 hr	7 d	14 d
DMF Control	6.67 ^{bc}	0.37 ^{cd}	0.39 ^{bc}
Barley Flour Control	4.87 ^{bcd}	0.38 ^{cd}	0.44 ^{ab}
0.12% β -glucan	4.30 ^{bcd}	0.39 ^{bcd}	0.47 ^a
0.24% β -glucan	4.11 ^{bcd}	0.34 ^d	0.40 ^{abc}
0.96% β -glucan	5.50 ^{bcd}	0.37 ^{cd}	0.45 ^{ab}
2.4% β -glucan	6.84 ^b	0.34 ^d	0.43 ^{ab}
0.5% CMC	5.09 ^{bcd}	0.42 ^{bc}	0.47 ^a
With 550 MAU amylase			
0.12% β -glucan	9.95 ^a	0.53 ^a	0.35 ^c
0.24% β -glucan	4.22 ^{bcd}	0.38 ^{bcd}	0.47 ^{ab}
0.96% β -glucan	3.79 ^{cd}	0.37 ^{cd}	0.42 ^{abc}
2.4% β -glucan	2.89 ^d	0.45 ^b	0.43 ^{ab}
0.5% CMC	4.25 ^{bcd}	0.40 ^{bcd}	0.43 ^{ab}
LSD^b($\alpha=0.05$)	2.99	0.08	0.07
Mean	5.21	0.40	0.43
LSD_{time}($\alpha=0.05$)		0.55	

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

APPENDIX D

TABLE D.1
Effect of Additives and Storage on Rollability Score of Corn Tortillas Stored at 4°C^a

β -glucan (%)	CMC (%)	4 hr	1d	4d	7d	14d
0	0	4.9 ^b	4.4 ^{cd}	3.6 ^{de}	3.1 ^d	2.1 ^c
0	0.25	5 ^a	4.7 ^{abc}	4.3 ^{abc}	3.7 ^{bc}	3.3 ^{ab}
0	0.5	5 ^a	4.9 ^a	4.5 ^a	4.4 ^a	3.8 ^a
0.03	0	5 ^a	4.8 ^{ab}	4.4 ^a	3.6 ^{cd}	2.9 ^b
0.06	0	5 ^a	4.7 ^{abc}	4.2 ^{abc}	3.8 ^{bc}	3.4 ^{ab}
0.06	0.125	5 ^a	4.8 ^{ab}	4.3 ^{ab}	4.2 ^{ab}	3.8 ^a
0.12	0	5 ^a	4.8 ^{ab}	3.5 ^e	3.5 ^{cd}	3.5 ^{ab}
0.12	0.5	5 ^a	4.9 ^a	4.3 ^{abc}	4.4 ^a	4.0 ^a
0.18	0.375	5 ^a	4.9 ^a	3.9 ^{cd}	4.7 ^a	3.8 ^a
0.24	0	5 ^a	4.1 ^d	4.0 ^{bcd}	3.8 ^{bc}	3.0 ^b
0.24	0.25	5 ^a	4.9 ^a	4.2 ^{abc}	4.3 ^a	3.4 ^{ab}
0.24	0.5	5 ^a	4.5 ^{bcd}	4.4 ^{ab}	4.5 ^a	4.0 ^a
LSD^b($\alpha=0.05$)		0.02	0.44	0.39	0.54	0.66
Mean		5.0	4.7	4.1	4.0	3.4
LSD_{time}($\alpha=0.05$)				0.17		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE D.2
Effect of Additives and Storage on Pliability Score of Corn Tortillas Stored at 4°C^a

β -glucan (%)	CMC (%)	4 hr	1d	4d	7d	14d
0	0	4.9 ^b	3.6 ^{bc}	2.8 ^c	2.4 ^c	1.8 ^d
0	0.25	4.9 ^b	4.0 ^{ab}	3.5 ^{ab}	3.2 ^b	2.8 ^{bc}
0	0.5	5.0 ^a	4.2 ^a	3.6 ^{ab}	3.6 ^{ab}	3.1 ^{abc}
0.03	0	4.9 ^b	3.8 ^{abc}	3.9 ^a	3.2 ^b	2.6 ^c
0.06	0	5.0 ^a	3.8 ^{abc}	3.3 ^{abc}	3.2 ^b	2.9 ^{bc}
0.06	0.125	5.0 ^a	3.9 ^{ab}	3.4 ^{abc}	3.5 ^{ab}	3.1 ^{abc}
0.12	0	5.0 ^a	3.9 ^{ab}	3.1 ^{bc}	3.3 ^b	2.9 ^{bc}
0.12	0.5	5.0 ^a	4.2 ^a	3.8 ^a	3.8 ^a	3.6 ^a
0.18	0.375	5.0 ^a	4.1 ^a	3.5 ^{ab}	3.9 ^a	3.0 ^{abc}
0.24	0	5.0 ^a	3.4 ^c	3.4 ^{abc}	2.5 ^c	2.8 ^{bc}
0.24	0.25	5.0 ^a	4.0 ^{ab}	3.8 ^a	3.3 ^b	3.2 ^{abc}
0.24	0.5	5.0 ^a	4.0 ^{ab}	3.9 ^a	3.6 ^{ab}	3.3 ^{ab}
LSD^b($\alpha=0.05$)		0.04	0.44	0.63	0.49	0.63
Mean		5.0	3.9	3.5	3.3	2.9
LSD_{time}($\alpha=0.05$)				0.15		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE D.3
Effect of Additives and Storage on Rupture Force (N) of Corn Tortillas Stored at 4°C^a

β -glucan (%)	CMC (%)	4 hr	1d	4d	7d	14d
0	0	5.19 ^{bc}	12.03 ^{abcd}	13.11 ^{bc}	11.74 ^d	13.15 ^d
0	0.25	5.48 ^b	11.43 ^{cd}	13.66 ^{ab}	13.66 ^{abc}	14.39 ^{ab}
0	0.5	6.13 ^a	12.17 ^{abc}	12.81 ^{abc}	13.36 ^{abc}	14.27 ^{abc}
0.03	0	5.21 ^{bc}	11.29 ^d	12.91 ^c	12.90 ^{cd}	13.13 ^{cd}
0.06	0	4.96 ^c	11.74 ^{bcd}	13.66 ^{abc}	13.27 ^{bc}	13.91 ^{abcd}
0.06	0.125	5.20 ^{bc}	12.73 ^{abc}	13.20 ^{bc}	13.31 ^{abc}	14.02 ^{abcd}
0.12	0	5.22 ^{bc}	13.08 ^a	13.36 ^{abc}	12.56 ^{cd}	14.29 ^{abcd}
0.12	0.5	5.06 ^c	11.90 ^{abcd}	12.83 ^{abc}	13.01 ^{abc}	13.56 ^{abcd}
0.18	0.375	5.13 ^b	12.64 ^a	14.17 ^a	13.90 ^{ab}	13.17 ^{bcd}
0.24	0	5.36 ^{bc}	12.47 ^{abcd}	13.02 ^c	14.34 ^{abc}	14.36 ^{abcd}
0.24	0.25	5.10 ^c	12.63 ^{ab}	12.85 ^{bc}	13.69 ^{abc}	13.27 ^{cd}
0.24	0.5	5.03 ^{bc}	13.01 ^a	13.77 ^{abc}	14.21 ^a	13.96 ^a
LSD^b($\alpha=0.05$)		0.45	1.11	1.27	1.05	1.10
Mean		5.26	12.25	13.25	13.37	13.83
LSD_{time}($\alpha=0.05$)				0.31		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE D.4
Effect of Additives and Storage on Rupture Distance (mm) of Corn Tortillas Stored at 4°C^a

β -glucan (%)	CMC (%)	4 hr	1d	4d	7d	14d
0	0	3.60 ^g	1.98 ^{bc}	1.72 ^{de}	1.57 ^d	1.56 ^{bc}
0	0.25	4.90 ^{def}	1.97 ^{bc}	1.80 ^{cde}	1.88 ^{abc}	1.71 ^{ab}
0	0.5	5.28 ^{abcd}	2.18 ^{ab}	1.88 ^{bcd}	2.10 ^a	1.72 ^{ab}
0.03	0	4.33 ^f	1.81 ^c	1.69 ^{de}	1.90 ^{abc}	1.46 ^c
0.06	0	5.00 ^{cde}	1.77 ^c	1.81 ^{cde}	1.84 ^{bc}	1.64 ^{abc}
0.06	0.125	5.03 ^{bcd}	2.02 ^{bc}	1.81 ^{cde}	1.87 ^{abc}	1.60 ^{bc}
0.12	0	5.02 ^{ce}	2.20 ^{ab}	1.80 ^{cde}	1.75 ^{cd}	1.63 ^{abc}
0.12	0.5	5.62 ^{abc}	2.15 ^{ab}	2.13 ^{ab}	2.09 ^{ab}	1.71 ^{ab}
0.18	0.375	5.52 ^{abcd}	2.21 ^{ab}	2.22 ^a	1.87 ^{abc}	1.57 ^{bc}
0.24	0	4.58 ^{ef}	2.00 ^{bc}	1.55 ^e	1.79 ^{cd}	1.50 ^c
0.24	0.25	5.68 ^a	2.25 ^{ab}	1.94 ^{abcd}	1.97 ^{abc}	1.65 ^{abc}
0.24	0.5	5.66 ^{ab}	2.33 ^a	2.05 ^{abc}	2.09 ^a	1.81 ^a
LSD^b($\alpha=0.05$)		0.64	0.29	0.31	0.25	0.21
Mean		5.02	2.07	1.89	1.86	1.63
LSD_{time}($\alpha=0.05$)				0.12		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE D.5
Effect of Additives and Storage on Young's Modulus (N/s) of Corn Tortillas Stored at 4°C^a

β -glucan (%)	CMC (%)	4 hr	1d	4d	7d	14d
0	0	1.04 ^a	3.45 ^{bc}	3.77 ^{abcd}	3.77 ^{bcd}	4.08 ^c
0	0.25	0.89 ^{bc}	3.24 ^c	3.86 ^a	3.86 ^{abc}	4.40 ^{ab}
0	0.5	0.93 ^b	3.23 ^c	3.53 ^{abcd}	3.53 ^e	4.32 ^{abc}
0.03	0	0.92 ^b	3.41 ^{bc}	3.54 ^{abcd}	3.54 ^e	4.34 ^{abc}
0.06	0	0.78 ^{def}	3.78 ^a	3.78 ^{ab}	3.78 ^{bcd}	4.44 ^{ab}
0.06	0.125	0.80 ^{cde}	3.57 ^{ab}	3.79 ^{abcd}	3.79 ^{abcd}	4.42 ^{ab}
0.12	0	0.78 ^{def}	3.42 ^{bc}	3.82 ^{abc}	3.82 ^{abc}	4.43 ^{ab}
0.12	0.5	0.70 ^{ef}	3.28 ^{bc}	3.57 ^d	3.57 ^{de}	4.22 ^{bc}
0.18	0.375	0.77 ^{def}	3.47 ^{bc}	3.95 ^{cd}	3.95 ^{ab}	4.33 ^{abc}
0.24	0	0.87 ^{bcd}	3.38 ^{bc}	4.01 ^{ab}	4.01 ^a	4.56 ^a
0.24	0.25	0.69 ^f	3.34 ^{bc}	3.79 ^{cd}	3.79 ^{abcd}	4.16 ^{bc}
0.24	0.5	0.73 ^{ef}	3.25 ^c	3.71 ^{cd}	3.71 ^{cde}	4.30 ^{abc}
LSD^b($\alpha=0.05$)		0.10	0.30	0.30	0.23	0.26
Mean		0.83	3.40	3.76	3.78	4.33
LSD_{time}($\alpha=0.05$)				0.08		

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE D.6
Effect of Additives and Storage on Final Stiffness (10⁵ Pa) of Corn Tortillas Stored at 4°C^a

β -glucan (%)	CMC (%)	4 hr	7d	14d
0	0	2.34 ^{bc}	34.36 ^{ab}	38.76 ^a
0	0.25	2.53 ^{bc}	34.67 ^a	38.47 ^{ab}
0	0.5	2.35 ^{bc}	33.04 ^{bc}	37.24 ^{abc}
0.03	0	2.23 ^c	32.56 ^c	37.60 ^{ab}
0.06	0	2.47 ^{bc}	32.96 ^c	37.56 ^{ab}
0.06	0.125	2.39 ^{bc}	32.53 ^c	37.04 ^{bcd}
0.12	0	2.40 ^{bc}	31.75 ^{cd}	35.73 ^{cde}
0.12	0.5	2.43 ^{bc}	29.97 ^e	34.76 ^e
0.18	0.375	2.50 ^{bc}	32.77 ^c	35.44 ^{de}
0.24	0	2.57 ^b	31.81 ^{cd}	36.98 ^{bcd}
0.24	0.25	3.06 ^a	31.08 ^{de}	34.49 ^e
0.24	0.5	2.43 ^{bc}	32.01 ^{cd}	35.01 ^e
LSD^b($\alpha=0.05$)		0.31	1.36	1.63
Mean		2.47	32.46	36.60
LSD_{time}($\alpha=0.05$)			0.45	

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

TABLE D.7
Effect of Additives and Storage on Energy Dissipated (10^{-4} J/m³) of Corn Tortillas
Stored at 4°C^a

β-glucan (%)	CMC (%)	Initial	7d	14d
0	0	5.40 ^{bc}	0.44 ^b	0.26 ^c
0	0.25	3.86 ^{bcd}	0.36 ^b	0.30 ^{bc}
0	0.5	8.62 ^a	0.38 ^b	0.31 ^{abc}
0.03	0	3.27 ^{cd}	0.34 ^b	0.31 ^{abc}
0.06	0	4.06 ^{bcd}	0.35 ^b	0.30 ^{bc}
0.06	0.125	5.40 ^{bc}	0.36 ^b	0.32 ^{abc}
0.12	0	9.01 ^a	0.43 ^b	0.36 ^{ab}
0.12	0.5	4.41 ^{bcd}	0.38 ^b	0.38 ^a
0.18	0.375	6.47 ^{ab}	0.34 ^b	0.35 ^{ab}
0.24	0	5.20 ^{bcd}	0.45 ^b	0.34 ^{ab}
0.24	0.25	2.13 ^d	1.31 ^a	0.38 ^a
0.24	0.5	3.42 ^{bcd}	0.34 ^b	0.37 ^{ab}
LSD^b(α=0.05)		3.14	0.50	0.07
Mean		5.10	0.46	0.33
LSD_{time}(α=0.05)			0.58	

^a Means in the same column followed by the same letter are not significantly different.

^b LSD = Least significant difference for means separation.

APPENDIX E

APPENDIX F

TABLE F.1
Statistic Indicators for Yield, Tortilla Weight And Thickness

Parameter	Yield	Tortilla weight	Tortilla thickness
p-value	0.19	0.06	0.28
R ²	0.60	0.69	0.56
CV (%)	19.6	2.42	3.81

TABLE F.2
Statistic Indicators for Subjective and Objective Texture Parameters at 4 hr After Production

Parameter	Rollability	Pliability	Rupture force	Rupture distance	Young's modulus	Final stiffness	Energy dissipated
p-value	0.40	0.77	<0.0001	<0.0001	<0.0001	0.0018	<0.0001
R ²	0.25	0.17	0.46	0.46	0.83	0.30	0.63
CV (%)	6.27	11.23	12.18	14.86	8.01	11.67	18.97

TABLE F. 3**Statistic Indicators for Subjective and Objective Texture Parameters at 14 Days of Storage at 4°C**

Parameter	Rollability	Pliability	Rupture force	Rupture distance	Young's modulus	Final stiffness	Energy dissipated
p-value	<0.0001	<0.0001	<0.0001	0.1223	<0.0001	<0.0001	0.002
R ²	0.63	0.83	0.56	0.18	0.43	0.55	0.28
CV (%)	18.43	10.10	8.157	15.44	9.20	6.54	21.33

VITA

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