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The Influence of Fat and Water on the Melted Cheese Characteristics of Mozzarella Cheese

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THE INFLUENCE OF FAT AND WATER ON THE
MELTED CHEESE CHARACTERISTICS OF MOZZARELLA CHEESE

by

Robert Lloyd Fife

A dissertation submitted in partial fulfillment
of the requirements for the degree

of

DOCTOR OF PHILOSOPHY

in

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Approved:

UTAH STATE UNIVERSITY
Logan, Utah

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ABSTRACT

The Influence of Fat and Water on the Melted Cheese Characteristics of Mozzarella Cheese

by

Robert Lloyd Fife, Doctor of Philosophy

Utah State University, 2003

Major Professor: Dr. Donald J. McMahon
Department: Nutrition and Food Sciences

The effect of reducing the fat content of low-moisture part-skim Mozzarella cheese from 19% to less than 5% on melted cheese properties, i.e., apparent viscosity, cheese melt, and cook color, was investigated. Functional properties of melt and stretch and cook color were evaluated at d 1, 7, 14, and 28. A rapid microwave oven method underestimated the moisture content of the low fat cheeses by approximately 10%. Low fat cheese did not melt as well as did the low-moisture part-skim Mozzarella cheese although the moisture content of the low fat cheese (moisture content ranged from 62.5% to 63.6%) was greater than the moisture content of the part-skim control (52.1%). Storage for 28 d only marginally increased the meltability of low fat cheese. Lower fat content increased cook color. The amount of intact αs-CN decreased by at least 48% in all cheeses as a result of proteolysis during 28 d of storage.
The relative proportion of bound, entrapped, and expressible water was determined for a reduced-fat (8% fat) and control (19% fat) Mozzarella cheese on d 1, 7, 14, and 21 of refrigerated storage. Changes in the state of water were related to changes in cheese microstructure of a commercial Mozzarella cheese and to changes in cheese meltability of the control cheese. The amount of expressible water was proportional to fat content. Throughout storage, fat/serum channels became smaller and the protein matrix expanded into the areas between fat globules. The meltability of both cheeses increased during storage. Both cheeses contained 0.71 g bound water/g protein. Expressible water decreased in both cheeses until by d 21 no water was expressible. Entrapped moisture increased from approximately 10% to 60% for the control cheese and from approximately 33% to 50% for the reduced-fat cheese.

An objective test was developed for measuring stretch, a characteristic of melted cheese. Three nonfat and four low-moisture part-skim cheeses were evaluated using the new test and the results compared with conventional test methods. Two new melted cheese parameters were defined: melt strength, the maximum load (g) obtained during the test, and stretch quality, the average load (g) as the cheese fibers stretched and elongated. Melt strength correlated with apparent viscosity. Stretch quality was determined for selected nonfat and low-moisture part-skim cheeses. A three-pronged probe was used to pull cheese vertically from a melted cheese pool. Use of this elongation stretch test, along with more traditional melted cheese tests, provides more complete information about the functional properties of Mozzarella cheese.
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Robert L. Fife
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LIST OF ABBREVIATIONS

AV = apparent viscosity
C:F = casein-to-fat ratio
CN = casein
DSC = differential scanning calorimeter
FM = melt strength
LMPS = low-moisture part-skim
MFFC = moisture in fat-free cheese
NF = nonfat
SEM = standard error of the mean
SL = stretch length
SQ = stretch quality
SQ5 = stretch quality at 5 cm of extension
SQ20 = stretch quality at 20 cm of extension
UW = University of Wisconsin
CHAPTER 1
GENERAL INTRODUCTION

Low-moisture part-skim Mozzarella cheese is the most important type of Mozzarella cheese used as an ingredient by the pizza restaurant industry. As such, its organoleptic and functional characteristics are of interest to manufacturers of pizza cheese and to consumers. Organoleptic characteristics such as color, blistering character, shredability, flavor, and watering-off or oiling-off tendency are of primary importance. However, these attributes are of little concern if the pizza cheese will not melt or stretch.

Most research has focused on low-moisture part-skim Mozzarella cheese that contains approximately 21% fat and 48% moisture. However, those interested in consuming less dietary fat continue to seek alternatives to low-moisture part-skim Mozzarella cheese. Since reduced-fat, low-fat, and nonfat Mozzarella cheese represent potential alternatives to part-skim Mozzarella cheese, a greater understanding of these cheese types is important.

As fat is reduced, Mozzarella cheese becomes rubbery and tough, is less elastic, and lacks the ability to melt and stretch. Interestingly, these changes also occur if water, rather than fat, is removed. Assuming that a relationship exists between the amount of fat and water in cheese and cheese meltability, understanding the role each plays as their relative proportions change in reduced-fat cheeses is important.

Melt and stretch are the result of heat transfer and thermal phase change characteristics of the cheese and the flow properties of the melt. As such, they are highly
interrelated and often difficult to measure. Helical viscometry, a test used to measure
melted cheese properties, has been used to access the stretchability of Mozzarella cheese
although this test primarily measures melt properties rather than stretch properties. Melt
and stretch may not always be correlated with one another. Therefore, it would be useful
to develop a test that builds upon the principles of helical viscometry, while objectively
measuring stretch characteristics. Thus, the two most important melted cheese
characteristics, melt and stretch, could more appropriately be used to characterize melted
cheese properties.

The objectives of this research were to:

1. Determine how reductions in fat level to less than 5% effect the meltability of
   Mozzarella cheese.
2. Determine the status of water in Mozzarella cheese and its influence on cheese
   meltability.
3. Develop a tensile test for objectively measuring the stretchability of melted cheese.
CHAPTER 2

LITERATURE REVIEW

INTRODUCTION

During 1998, Italian cheese production in the United States surpassed 3 billion pounds. Mozzarella cheese production increased to 2.4 billion pounds, which made up 79% of U.S. Italian cheese production volume (National Cheese Institute, 1999). Of the Mozzarella cheese produced, most was used by the pizza restaurant industry, the largest user of Mozzarella cheese in the U.S. (Alvarez, 1986; Pilcher and Kindstedt, 1990). The pizza restaurant industry uses cheese with fat content typical of, or higher than, low-moisture, part-skim Mozzarella, i.e. 30 to 45% fat on a dry weight basis (Food and Drug Administration, 2000). Since low-moisture part-skim Mozzarella cheese represents a large proportion of the total Mozzarella cheese production, it is not surprising that studies have focussed on its attributes and characteristics (Kindstedt, 1995; Kindstedt and Kiely, 1990; Masi and Addeo, 1986; Mistry and Anderson, 1993; Oberg et al., 1991; Oberg et al., 1993; Yun et al., 1995). Two important characteristics of Mozzarella cheese used as pizza topping are the ability to melt and stretch; less important characteristics include, cheese color, blistering character, shredability and oil/water retention (Park et al., 1984). Although melt and stretch are of particular importance, it has been difficult to objectively quantify these characteristics in a manner that is useful to cheese manufacturers and to those that use Mozzarella cheese.
Dietary awareness of consumers and their desire to follow nutritional guidelines (National Academy of Sciences, 1989) by reducing total fat intake have prompted the cheese industry to investigate fat reduction in Mozzarella cheese (McMahon et al., 1993). Reduced-fat, low fat, and nonfat cheese variations have been developed as potential alternatives to part-skim Mozzarella cheese.

**Reduced-Fat Mozzarella**

As fat or moisture content is increased or decreased, the ability of the cheese to melt and stretch is altered. Masi and Addeo (1986) found that increases in the fat and moisture content of Mozzarella cheese are accompanied by a decrease in the modulus of elasticity (an indication of rigidity), resulting in softening of the cheese body and difficulty in shredding. Konstance and Holsinger (1992) found that reducing moisture or fat results in increased hardness, elevated springiness values, and decreased meltability. Defects associated with low fat Mozzarella cheese include a rubbery, tough texture, lack of flavor, paleness or green tint, inability to melt, and poor stretchability (Mistry and Anderson, 1993; Tunick et al., 1993).

One method for minimizing melt and stretch defects in low fat Mozzarella cheese is to increase cheese moisture. Tunick et al. (1991) increased the moisture content of Mozzarella cheese by eliminating the “cook” step and incorporating a “wash” step into their cheese making procedure. Merrill et al. (1994) described a method for manufacturing
reduced-fat Mozzarella cheese containing 50% less fat than part-skim Mozzarella. They incorporated a number of changes in their cheese making procedure that increased the amount of moisture. Higher pasteurization temperature, milk pre-acidification, larger curd size, reduced cook temperature, minimal stirring during cooking, and less frequent turning during cheddaring were used to retain more moisture in the curd. No significant differences in apparent viscosity (AV), melting, or browning, occurred between the control cheese and the reduced-fat cheese over 28 d of refrigerated storage when these procedural changes were followed.

Although Tunick et al. (1991) and Merrill et al. (1994) were able to produce reduced-fat cheese with functional characteristics similar to low-moisture part-skim cheese, the effect of reducing the fat content of part-skim Mozzarella cheese more than 50% was not determined. An understanding of fat reduction below 50% would be important since low fat and nonfat Mozzarella cheese contain fat levels 75% to 90% below that found in low-moisture part-skim Mozzarella cheese (Food and Drug Administration, 2000).

**Cheese Structure**

When milk is renneted, casein micelles are converted from a stable colloid into a protein gel with individual casein micelles joined together into chains. As cheese making proceeds, and whey is expelled from the curd, the protein chains become thicker through continued association with one another until they form a protein matrix in which water and fat are distributed (Oberg et al., 1993; Schmidt, 1982). For Mozzarella cheese, this
process is the same as Cheddar cheese up until the time the Mozzarella cheese curds are transferred to the cooker-stretcher to produce a pasta filata cheese (Oberg et al., 1993) rather than being cheddared (Green et al., 1981).

In pasta filata cheese, the proteins in the cheese curds coalesce into larger strands that are oriented in the direction of stretching. This coalescence results in a redistribution of the water and fat during stretching and molding with the larger strands (or fibers) of protein being separated by channels containing water, water-soluble cheese components, bacteria, and fat globules (Oberg et al., 1993; Prentice et al., 1993). Changes in the microstructure and functionality of Mozzarella cheese during storage (Cooke et al., 1995; Kiely et al., 1993; Paulson et al., 1998) suggest that the proteins are not in a quiescent state immediately after stretching and molding but undergo further structural rearrangement.

**Proteolysis During Storage**

The improved meltability of Mozzarella cheese observed during storage is generally associated with the proteolytic hydrolysis of $\alpha_{s1}$-casein ($\alpha_{s1}$-CN) by chymosin, plasmin, and proteolytic enzymes originating from the starter cultures (Fox et al., 1979; Kiely et al., 1993; Kindstedt and Guo, 1997; McMahon et al., 1993; Tunick et al., 1993). Hydrolysis of $\beta$-casein ($\beta$-CN) in cheese made using chymosin occurs at a much slower rate and is not usually associated with improved meltability. However, Bogenrief and Olson (1995) demonstrated that the meltability of Cheddar cheese correlated more with $\beta$-CN hydrolysis than with $\alpha_{s1}$-CN hydrolysis. In cheese made using *Cryphonectria*
parasitica rennet, the meltability was greater, at all stages of maturation, compared to cheeses made with chymosin. They attributed the improved meltability to increased β-CN hydrolysis.

It has also been shown (Yun et al., 1993) that increased β-CN hydrolysis improves cheese meltability, increases free oil, and decreases apparent viscosity in Mozzarella cheese. However, we have observed (unpublished data) that changes in cheese meltability can occur during storage without any significant decrease in intact β-CN levels. This suggests that proteolysis may not be the only causal effect for changes in the meltability of Mozzarella cheese during storage and that other changes occurring concomitantly in the cheese during storage should be investigated. One such change that has been observed, as shown by a decrease in expressible serum, is a redistribution of water in cheese during storage (Guo and Kindstedt, 1995). However, it is not known what causes this change in water or what subsequent role water plays in cheese functionality.

Water Distribution in Cheese

The various states in which water may exist in a material such as cheese can be described in terms of the spatial relationship between water and the solid constituents of the food (in cheese, these solids are predominantly protein although water interactions also exist between other cheese constituents). Bound water has been characterized as nonsolvent water or chemisorbed water (Geurts et al., 1974); sorbent- or solute-associated water and unfreezable water (Berlin, 1981); constitutional water (i.e., water
that occupies interstitial protein spaces); vicinal water (i.e., water that covers a protein as a single layer) (Fennema, 1985), and also characterized in terms of hydrodynamically influenced layers of water that surround a protein (Farrell, 1988). Such isotropically bound water is intimately associated with (or is in close proximity to) the protein surface. This water exhibits slower rotational and translational speeds than pure water, is not available as a solvent, and cannot be frozen at -40°C (Farrell et al., 1989).

In contrast, bulk water is water that is more loosely associated with the proteins. Bulk water retains a large solvent capacity and is freezable at -40°C. Farrell et al. (1989) described bulk water as being anisotropic, because the rotational and translational speeds of such water molecules are not significantly affected by their interaction with proteins. Bulk water may be further divided into entrapped or free water depending upon its mobility within the protein matrix. Water that is impeded by the macrostructure of the protein matrix, such that it cannot be expressed by centrifugation, is considered to be entrapped. Water that is not impeded by the protein matrix and may be expressed by centrifugation is known as expressible water (Fennema, 1985). Thus, the distribution and movement of water in cheese may be characterized as being bound to proteins, entrapped by those proteins, or expressible by centrifugation.

Although water distribution can be characterized as described, it is important to note that other constituents within the cheese matrix may hold moisture and subsequently affect cheese functionality. Recently, Perry et al. (1997; 1998) used exopolysaccharide-producing starter cultures to increase the moisture content of low fat Mozzarella cheese.
They were able to increase the moisture content of low fat Mozzarella cheese by adding starter culture with a cell wall exopolysaccharide. When cheese was made using 10 or 454 kg vats (Perry et al., 1997), cheese moisture was increased by 3% and 2%, respectively (Perry et al., 1998). In each case, increased moisture content was accompanied by an increase in meltability. This increase was probably associated with water that was trapped, hydrogen bonded, or bound to the polysaccharide capsule.

**MEASURING MELTED CHEESE CHARACTERISTICS**

Two important functional characteristics of pizza cheese are the ability to melt when heated and stretch when pulled. Conceptually, these two attributes are clearly understood although neither term carries a precise rheological definition. Consumers generally evaluate cheese melt as the degree to which shredded cheese flows after being heated in an oven. Additionally, they evaluate cheese stretch by monitoring the number of cheese strands that form and extend as they lift a pizza slice from their plate. Melt refers to the ability of cheese to flow after heating. It is implied that no force other than gravity is applied to the cheese. Stretch refers to the ability of melted cheese to form fibrous strands that extend when the cheese is pulled apart (Kindstedt, 1995). The application of an external force to melted cheese mimics the manner in which consumers evaluate cheese stretch. A number of methods have been developed to measure cheese melt and stretch. Common among those methods used to evaluate cheese melt is the
absence of an externally applied force, while the presence of an externally applied force is common to methods used to evaluate stretch.

**Measuring Cheese Melt**

*Meltdown tests.* One of the most common methods used to assess cheese meltability was originally developed for the process cheese industry in the late 1950’s by L. D. Schreiber (Kosikowski, 1977). A cylindrical cheese sample, 41 mm in diameter and 5 mm in height, was heated in a convection oven for 5 min at 232°C. After heating, sample diameter expansion was measured as described by Kosikowski (1977). Degree of meltability was taken as the mean expansion of the cheese sample. This method was inexpensive, easy to perform, and continues to be used within the cheese industry (Kosikowski and Mistry, 1997).

Arnott et al. (1957) also used a cylindrical cheese sample (17 mm in diameter, 17 mm in height) to determine the meltability of process cheese. Rather than measuring the increase in cheese cylinder diameter, they measured the decrease in cheese cylinder height immediately after removing the sample from a convection oven (15 min at 100°C). The relative percent decrease in height was calculated and reported on a scale of 0 to 100%. Since neither this test nor the Schreiber test was standardized, additional investigators have used various sample dimensions and heating conditions to measure cheese meltability. Examples of modifications to these methods are described by Chang (1976), Hokes et al. (1982), Kovacs and Igoe (1976), Sood and Kosikowski (1979), and Weik et al. (1958).
Park et al. (1984) compared the Schreiber (Kosikowski, 1977) and Arnott et al. (1957) methods for measuring the meltability of sharp Cheddar, process American, and Mozzarella cheese. Both the Schreiber and Arnott methods provided results that were consistent, reproducible, and could differentiate among the meltability of the three types of cheese. However, the two meltability indices, i.e. diameter expansion and decrease in relative cheese cylinder height, did not correlate when compared with one another. They concluded that sample geometry and slow heat transfer, caused by different testing temperatures, effected the melt consistency within the sample and caused the lack of correlation between the two indices.

Park et al. (1984) also used a microwave oven to reduce the amount of time required to heat the cheese, thus reducing the effect of slow heat transfer. Cheese samples of the same dimension as were used in the Schreiber (Kosikowski, 1977) and Arnott et al. (1957) methods were heated in a microwave oven from 0 to 60 s (power setting not specified). Specimens melted after 20 to 60 s of heating. Meltability was determined as described in the original test methods. Again, no clear correlation was found between the two meltability indices. The meltability of the three types of cheese was compared using differential scanning calorimetry. When thermograms of the three cheese types were compared, little difference between the meltability patterns was observed, indicating that there was not a measurable thermal event taking place that could differentiate between the cheeses. Although differential scanning calorimetry could not detect a difference in melting properties of these cheeses, Park et al. (1984) recognized that physical factors such as geometry and testing conditions produced differences in cheese melt properties.
They concluded that both the Schreiber (Kosikowski, 1977) and Arnott et al. (1957) methods were capable of measuring differences in cheese melt.

Olson and Price (1958) measured the distance cheese flowed (mm) from a reference line in a horizontal glass tube as a means of measuring cheese melt. A cheese sample, 30 mm in diameter and 20 mm in height, was placed into one end of a glass tube and each end was closed with a stopper. The tube was placed horizontally into a convection oven at 110°C for 8 min. During the 8-min heating time the cheese melted and flowed down the tube. The distance of flow was measured and used to compare cheese meltability.

*Modified meltdown tests*. Breene et al. (1964) combined the concepts of the Schrieber (Kosikowski, 1977) and Arnott et al. (1957) tests to measure cheese meltability. They used decrease in height of a cheese cylinder (15 mm in diameter, 5 mm in height) as well as increase in diameter expansion to measure the meltability of pizza cheese manufactured without starter culture. A cheese sample was prepared by sectioning a 15-mm diameter cylinder of cheese into 5-mm slices. The sample was placed on a dish, tempered at 4°C, and placed into a boiling water bath for 5 min during which time the cheese melted. Reduction in sample height and the diameter expansion were measured, with the average of two diameters measured at right angles to each other being used to calculate diameter expansion. Melt was reported as percent decrease in cheese sample height and percent increase in horizontal expansion.

Oberg et al. (1992a) modified the melt test of Olson and Price (1958) by using a specific amount of cheese, rather than a sample of specified dimensions, to measure the
Fifteen grams of ground cheese was placed in one end of a glass tube (30 x 250 mm). A rubber stopper was placed in the end of the tube containing the cheese, and the other end closed with a stopper containing an exhaust hole. The cheese was tempered at 4°C for 30 min while the tube was positioned vertically, then the tube was positioned horizontally in a convection oven at 110°C for 60 min. During the 60 min, the cheese melted and flowed down the tube. After cooling to room temperature, the distance the cheese flowed was measured and used as a measure of cheese meltability.

Bogenrief and Olson (1995) modified both the Schreiber test (Kosikowski, 1977) and the Olson and Price (1958) test to determine the effect of β-CN hydrolysis on the meltability of cheddar cheese over 180 d refrigerated storage. A sample 30 mm in diameter and 20 mm in height (Olson and Price, 1958) was heated in a microwave oven for 45 s at full power. The maximum width and width at 90 degrees to the maximum of the melted cheese was measured immediately after removal from the oven. Bogenrief and Olson (1995) found that this method provided the same information about the effect of β-CN hydrolysis on cheddar cheese melt as did the conventional Schreiber test. Bogenrief and Olson (1995) modified the melt test of Olson and Price (1958) by changing the heating medium from air to water. A sample, 30 mm in diameter and 20 mm in height, was placed in a glass tube as previously described (Olson and Price, 1958). A water bath (95°C), rather than a convection oven, was used to heat cheese samples. A stopper with a vent tube was used to close the distal end of the glass tube and allow for air expansion.
while the tube was under the surface of the water. Cheese flow at 5, 8, and 12 min was recorded rather than at 8 min only. The extent to which cheese flowed at 5 min was used to compare cheese meltability. Bogenrief and Olson (1995) showed that cheese melt at 5 min increased over 180 d maturation while noting that the same conclusions could be made using cheese melts at either 8 or 12 min of heating.

**UW Meltmeter.** Wang et al. (1998) developed the University of Wisconsin (UW) Meltmeter for evaluating the melt/flow characteristics of cheese. Conceptually, this method was a mechanized version of the Arnott et al. (1957) method. The meltmeter consists of an aluminum body with a doughnut shape heater inside. This doughnut shaped heater was in contact with a stationary piston located where the doughnut hole would be. This piston had a diameter equivalent to the doughnut “hole.” The outer ring, i.e., the doughnut shaped heater, could be moved up or down independently of the stationary piston. When testing a sample, the annular ring was raised such that its top surface was positioned above the top surface of the stationary piston, thus forming a heated sample well. A circular plate, oriented above the sample well, was lowered onto the upper surface of the raised doughnut heater (see Wang et al., 1998 or Kuo et al., 2000). This circular plate acted as a sample well cap, applied a constant weigh to the top surface of a sample, and provided a means for determining cheese height. To perform a test, a solid cheese sample was placed in the heated sample well, the circular plate was lowered onto the upper surface of the outer ring to cap the sample well and the sample was tempered until the desired sample temperature (typically, 60°C) was reached. After the desired sample temperature had been reached, the outer doughnut ring was lowered,
leaving the sample and circular plate poised on top of the stationary piston. Once the “walls” formed by the outer ring were lowered, the sample began to flow under the constant force of the circular plate. The sample height was recorded as a function of time, and meltability was calculated as the difference between the initial height and the height of the melted cheese after 1 s. Meltability calculated in this manner correlated well with biaxial elongational viscosity.

Measuring Cheese Stretch

Fork test. Attributing the fork test to a single source is difficult and many companies modify the general methodology as needed. Ground cheese is sprinkled on a pizza crust containing pizza sauce. After baking, a fork is inserted into the melted cheese and raised vertically until the cheese strands break (U. S. Department of Agriculture, 1980). Strand length, at the time the strands break, is defined as the stretchability of the cheese. Although test parameters are controlled, most technicians perform the fork test differently. Where and how the fork is inserted, tine orientation, the amount of tine covered by the cheese, and the speed used to lift the cheese, varies. In addition, defining the point at which the “majority” of the strands break depends on the experience and judgment of the technician. For these reasons, it is not uncommon for evaluations of the same cheese to differ. In fact, an experienced operator can make subtle changes in the test that will allow the desired result to be obtained. Other approaches have been used to measure cheese stretchability in order to minimize the subjectivity of the fork test.
**Weissenberg test.** Olson and Nelson (1980) used the tendency of a viscoelastic material to climb a smooth rod rotating in the material (the Weissenberg effect) to evaluate the elasticity (stringiness) of molten Mozzarella cheese. Shredded cheese samples (30 g) were placed into the bottom of an aluminum pan that was lined with wire mesh. A rod covered with filter paper was lowered into the pan and positioned just above the shredded cheese. The screen and filter paper provided a rough surface to which the cheese could adhere. Additional cheese (60 g) was distributed around the rod. The pan was placed into a water bath for 30 min at 65°C to melt the cheese. The height of the melted cheese was measured then the rod was rotated at 10 revolutions per minute, causing the cheese sample to climb the rod. Cheese was evaluated by measuring the height that the cheese climbed, the time required to fracture the cheese, place of fracture of the cheese in the pan, and the textural characteristics of the cheese.

Olson and Nelson (1980) related cheese column height to elasticity, fracture time to toughness, place of fracture to stringiness, and texture (smooth or rough) to meltability. They concluded that the Weissenberg method could be used to measure the elasticity of molten Mozzarella cheese and that those measurements could be used to predict the performance of cheese on a pizza. An advantage claimed for this method was that separate measurements could be obtained for stretch, elasticity, and melt. However, the assessment of the degree to which the cheese stretched or melted was highly subjective and is the major disadvantage of the method.

**Spinning flow.** Cavella et al. (1992) evaluated the applicability of a spinning flow method for determining the stretchability of Mozzarella cheese. Instrumentation
consisted of a piston-type capillary rheometer equipped with a microprocessor and pick-up system. Cheese plugs were loaded into a rheometer barrel contained within a heating chamber. Samples were melted then pressed through a small hole in the bottom of the capillary tube by a plunger. A cheese strand formed as the molten cheese was extruded through the hole. This cheese strand was then directed horizontally past a load cell and between two rotating pick-up wheels (imagine a sock being pulled between two wringers of an old-style washing machine). The amount of force that was required to stretch the cheese and the strand length were determined. Plunger speed, the distance between the pickup wheels and the exit section of the capillary, and the rate at which the pick up speed increased, were experimental variables. Cavella et al. (1992) used this method to determine the force, stress, and strand elongation at strand failure within the temperature range in which it was possible to stretch Mozzarella cheese (approximately 57°C to 82°C). Test reproducibility was comparable to that observed when the same method was used to test polymers of non-biological origin (Petrie, 1979). Cavella et al. (1992) concluded that spinning methodology could be used to evaluate the stretchability of Mozzarella cheese although the practicality of the method was not determined.

**Horizontal uniaxial extension.** Ak et al. (1993) developed a method that used fracture strain at strand failure to measure the stretchability of Mozzarella cheese. Thin dumbbell-shaped pieces of cheese (thickness, 6 mm; width, 8 mm; length, 60 mm) were preconditioned in an incubator to 10, 20, 30, or 40°C. Samples were placed in a heated oil bath and secured by clamps attached to the large ends of the cheese. One clamp remained stationary while the other was attached to a load cell by a cable and pulley system. When
the apparatus was activated, the cheese was pulled horizontally (50, 200, or 500 mm/min), causing it to stretch.

Tensile strength, deformability modulus, and fracture strain were successfully determined for Mozzarella cheese after 7, 14, 21, and 28 d refrigerated storage. Unfortunately, test temperatures higher then 40°C caused many of the cheese samples to sag, invalidating the stretch results. However, most cheese samples stretched to 200% of their original length while some stretched to 650%. Since test temperatures were significantly below those found in pizza ovens, this method had limited application for manufacturers of pizza cheese.

**Vertical uniaxial extension.** Ak and Gunasekaran (1995) modified the horizontal uniaxial extension method (Ak et al., 1993) such that cheese samples were stretched vertically and at a higher temperature (60°C). One end of a dumbbell shaped sample was clamped to a load cell while the other end was attached to a weight. Typical dimensions of the test-section were 6 mm in thickness, 7 mm in width, and 60 mm in length. Two pulling-weights (14 g and 18 g) were used. Strain at cheese strand failure was used as the measure of stretchability.

Unlike samples used for the horizontal uniaxial test, cheese samples used for this method were not tempered before submerging them in hot oil. As such, the temperature of the sample increased throughout the test, causing the stress and strain rates to be inconsistent. In addition, many samples stretched to the maximum capacity of the test. This made strain at strand fracture difficult to determine since the strands did not break. Ak and Gunasekaran (1995) suggested that heavier weights be used in order to cause the
elongating cheese strand to fracture at an earlier time. However, they chose not to attach heavier weights to their samples since it required modifying the sample shape or altering the instrumentation. Even so, they were able to show that the transient elongational viscosity of Mozzarella cheese could be determined by this method and that this parameter might provide helpful information on elongational properties of different cheeses.

**Compressive elongation.** Apostolopoulos (1994) developed a fundamental test to evaluate the elongation properties of melted cheese. A non-lubricated cylindrical cheese sample was placed on a hot plate and covered with a metal cap until the sample temperature reached 65°C. After the cheese melted, it was immediately compressed by another plate attached to a tensile testing machine at 20 mm/min. The elongation viscosity was calculated and used as an index of the ability of the cheese to form strings when stretched.

**Vertical tensile extension.** Apostolopoulos (1994) also developed a tensile test that better represented the way consumers evaluated the stretchability of pizza cheese. A circular plate was used as a template to hold the pizza crust. A smaller circular piece (with a vertical rod attached) was cut out allowing the center of the template to be raised independently of the edge. A similarly cut pizza crust was placed on the template and a standard weight of cheese sprinkled onto the crust. The complete apparatus was heated in a microwave oven for 15 s to melt the cheese. On removal from the oven, the rod was attached to the head of a tensile testing machine and pulled vertically, stretching the melted cheese. The extensibility of the cheese was taken as the distance of travel until all
the cheese strands broke. Sensory evaluation by trained panelists indicated that the extensibility test correlated well to the way consumers evaluated cheese on a pizza.

Peña et al. (1996) described a vertical tensile extension method for determining the stretchability of cheese. Their work primarily involved optimizing the type of probe used to stretch molten Cheddar, Oaxaca, and Asadero cheese rather than evaluating vertical extension as a method for evaluating cheese stretchability. Various probes (fork prongs, beam, and wire rectangle) were pulled through molten cheese (65°C) at 0.2, 0.5, 0.7, 0.1, and 1.5 mm/s. Cheese was lifted to a maximum extension of 90 mm. Unlike the vertical extension method of Apostolopoulos (1994), Peña et al. (1996) used the peak force exerted on the probe as a measure of cheese stretchability. They determined that the wire rectangle probe, pulled at 1 mm/s, provided peak force values with the lowest variability.

**Horizontal tensile extension.** Guinee and O’Callaghan (1997) modified the vertical tensile extension method of Apostolopoulos (1994) such that two halves of a cheese-covered pizza were pulled horizontally, rather than vertically. Cheese strand length at strand failure was used as the measure of cheese stretchability. They determined that the amount of the cheese per unit area of pizza crust and the holding time prior to stretching greatly influenced their results. Using this method, they were able to differentiate between types of cheese used as pizza topping and concluded that cheese extension was a realistic measure of stretchability.

**Helical viscometry.** Lee et al. (1978) included cheese viscosity as part of their study correlating sensory evaluation with texture profile analysis parameters such as
hardness, brittleness, chewiness, springiness, adhesiveness, and lumpiness. A Brookfield viscometer equipped with a helipath stand was used to evaluate the melted cheese properties of eleven cheeses, including Mozzarella. A ground cheese sample (unspecified amount) was tamped into two 16-mm x 100-mm glass test tubes. A T-spindle was pushed into the cheese contained in one test tube and positioned such that the T-spindle rested above the bottom of the tube. A thermocouple was positioned in the second test tube to monitor cheese temperature. Each test tube was placed into a water bath. The viscometer was actuated while the cheese was in a solid state. Viscosity readings registered off-scale since cheese samples were at room temperature. As the temperature of the water bath was gradually increased, the cheese softened and the viscosity readings began to decrease. Force/temperature data were plotted from 10 to 80°C. Viscosity values for a given test temperature at the first sign of softening, the temperature when the viscosity reading reached 50% of full-scale value, and the temperature at which cheese viscosity no longer decreased, were used to compare the meltability of the different cheeses. Although the Brookfield viscometer was equipped with a helipath, it was not activated during this test.

Kindstedt et al. (1989a) used a Brookfield viscometer in which the helipath stand was activated to evaluate cheese stretchability. Ground cheese was tamped into a large glass test tube and the cheese melted in a water bath prior to inserting a T-spindle. Upon activating the viscometer, full-scale values (or the maximum viscosity values) were recorded. The helipath stand was then activated, causing the T-spindle to cut through the cheese in a helix-like pattern. Unlike the earlier method (Lee et al., 1978), full-scale values
only occurred when the viscosity of the molten cheese exceeded the maximum torque spring capacity. Although a viscometer was used to measure resistance on the torque spring, values depended on the strain rate employed during the test, the length of the spindle, and the temperature of the melted cheese. As such, force measurements were expressed as AV rather than viscosity (van Vliet, 1991).

Kindstedt et al. (1989a) interpreted apparent viscosity to be a measure of cheese stretchability and elasticity and suggested that the maximum peak value of the force-time resistance profile be used to differentiate between cheeses. To determine the usefulness of helical viscometry, Kindstedt et al. (1989b) compared low-moisture part-skim Mozzarella cheese manufactured at one cheese plant with low-moisture Mozzarella cheese manufactured at another. Using helical viscometry, they demonstrated that the two cheese plants produced cheese with different AV (as would be expected because they made Mozzarella cheese with different fat levels). Additionally, they showed that the AV of cheese being produced within one plant was consistent, while the AV of cheese being produced within the other plant was inconsistent. They correlated the inconsistent AV within the latter cheese plant with poor milk quality and production problems. Kindstedt et al. (1989b) concluded that helical viscometry represented a useful method for studying cheese-melting properties.

Oberg et al. (1991) studied the effect of proteolytic activity on the physical properties of Mozzarella cheese, including stretchability, over 28 d refrigerated storage. They modified the method of Kindstedt et al. (1989b) by using a Brookfield viscometer with a more sensitive torque spring (50 to 2 x 10^6 centipoise vs. 100 to 8 x 10^6 centipoise)
and by using the sum of the viscosity readings from 6 to 10 min (while the cheese stretched above the cheese pool) to objectively compare cheese stretchability. Oberg et al. (1991) determined that there was an inverse relationship between cheese melt, as determined by the melt test of Olson and Price (1958) and stretch, as determined by helical viscometry; i.e., melt increased while stretch decreased. This observation confirmed earlier work by Keller et al. (1974). They found an inverse relationship between cheese melt and cheese viscosity while studying the effect of acid coagulants on the mineral retention and rheological properties of Mozzarella cheese made by direct acidification. Keller et al. (1974) measured melt by using a modified Arnott et al. (1957) method, and viscosity during compression as described by Davis (1937). Oberg et al (1991) also observed that the tail sections of the stretch profiles were more pronounced when using a more sensitive torque spring thus improving their ability to discern between cheese types.

Oberg et al. (1992a) used apparent viscosity to measure differences in cheese stretchability between Mozzarella cheeses manufactured using different milk-clotting enzymes. Rather than using the sum of the apparent viscosity measurements obtained as the cheese strand stretched above the molten cheese pool, they used the relative peak area at each of 10 readings, 1 min apart, as their measure of cheese stretchability. This method was also used to determine the effects of freezing, thawing, and shredding on the stretchability of low-moisture part-skim Mozzarella cheese (Oberg et al., 1992b).
CONCLUSIONS

Fat and water content affect the melt and stretch of Mozzarella cheese. As such, a better understanding of their role in determining how Mozzarella cheese melts and stretches is important to cheese manufacturers and consumers. Melt and stretch are highly interrelated characteristics. A variety of test methods have been developed to measure melt and stretch. Methods that measure melt generally involve measuring the degree to which molten cheese flows. This relationship between cheese melt and melted cheese flow appears to provide a direct measure of cheese melt. However, most methods used to measure cheese stretch use indirect measurements to determine cheese stretchability. It would be useful to develop a stretch test that mimics the manner in which cheese manufacturers and consumers access cheese stretch while reducing the subjectivity of the fork test.

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CHAPTER 3
FUNCTIONALITY OF LOW FAT
MOZZARELLA CHEESE

ABSTRACT

Low fat Mozzarella cheese was made from milks containing casein to fat ratios of 3.0, 5.0, 7.0, and 8.0. Prior to addition of rennet, milk was pasteurized at 79°C for 28 s and then acidified to pH 6.0 with lactic acid. Three replicates of each cheese were made in 7-L vats and stored at 4°C. Functional properties as pizza cheese were evaluated. Cheese moisture and fat contents were evaluated at 1 d. Apparent viscosity and extent of flow of melted cheese, cook color, and proteolysis were evaluated at 1, 7, 14, and 28 d. Moisture content by a rapid microwave oven method underestimated the moisture content of low fat cheeses; the probable moisture was calculated by component analysis. The part-skim Mozzarella control with 19% fat contained 51% moisture while the moisture contents of the low fat cheeses containing 2 to 5% fat were 63%. Low fat cheeses did not melt as well as did the part-skim Mozzarella cheese, although the differences between the cheeses with 2% and 5% fat were insignificant. Storage for 28 d only marginally increased the meltability of low fat cheese. Lower fat content also increased cook color. The amount of intact $\alpha_s$-casein decreased by at least 48% in all cheeses as a result of proteolysis during 28 d of storage.

INTRODUCTION

Dietary awareness of consumers and their desire to follow nutritional guidelines (National Academy of Sciences, 1989) by reducing total fat intake have prompted the cheese industry to investigate fat reduction in Mozzarella cheese (McMahon et al., 1993). Reduced-fat, low fat, and nonfat cheese variations are now being developed as potential alternatives to part-skim Mozzarella.

The pizza restaurant industry uses cheese with fat content typical of, or higher than, low-moisture part-skim Mozzarella and represents the largest category of Mozzarella users in the US (Alvarez, 1986; Pilcher and Kindstedt, 1990). In a recent survey, over 50% of pizza restaurants reported occasional to frequent quality problems, including melting (67%), poor shredding (55%), and blistering or browning problems (50%) (Pilcher and Kindstedt, 1990).

Two important characteristics of Mozzarella cheese used as pizza topping are the ability to melt and stretch (Park et al., 1984). As fat or moisture is removed, functional defects appear. Masi and Addeo (1986) found that increases in the fat and moisture of Mozzarella cheese are accompanied by a decrease in the modulus of elasticity (an indication of rigidity), resulting in softening of the cheese body and difficulty in shredding. Konstance and Holsinger (1992) found that reducing moisture or fat results in increased hardness, elevated springiness values, and decreased meltability. Defects associated with low fat Mozzarella cheese include a rubbery, tough texture, lack of flavor,
paleness or green tint, inability to melt, and poor stretchability (Mistry and Anderson, 1993; Tunick et al., 1993).

Merrill et al. (1994) described a method for manufacturing reduced-fat Mozzarella cheese containing 50% less fat than part-skim Mozzarella. A higher pasteurization temperature, milk preacidification, larger curd size, reduced cook temperature, minimal stirring during cooking, and less frequent turning during cheddaring were used to retain more moisture in the curd. No significant differences occurred between the two types of cheese in moisture, apparent viscosity (AV), melting, or browning over 28 d of refrigerated storage.

The objective of this study was to determine the effect of lowering fat content of part-skim mozzarella cheese by 75% to 90% on cheese functionality over 28 d of refrigerated storage. A melt test was used to measure the extent of cheese flow when cheese was melted, and an apparent viscosity test was used to provide information on stretchability. Browning of the cheese when heated was used to evaluate cook color, and degree of proteolysis during storage was also measured.

**MATERIALS AND METHODS**

**Milk, Cultures, and Rennet**

Pasteurized (79°C for 29 s) skim milk and 2% milk were obtained from the Gary H. Richardson Dairy Products Laboratory at Utah State University. Milk was cooled to 4°C; the protein and fat levels were determined and then were standardized to a casein to fat ratio (C:F) of either 1.2, 3.0, 5.0, 7.0, or 8.0. Lyophilized direct-set cultures of
*Lactobacillus helveticus* LH 100 and *Streptococcus thermophilus* TA 061 and single-strength calf rennet extract were obtained from Rhodia Food (Madison, WI).

**Manufacturing Procedure**

Low fat cheeses were made from milk with C:F of 3.0, 5.0, 7.0, and 8.0 using the method of Merrill et al. (1994) and compared with a control cheese made from milk having C:F of 1.2. Five stainless steel vats (22 x 22 x 22 cm), each containing 7 L of milk, were used to manufacture cheese. The milks with C:F 3.0, 5.0, 7.0, and 8.0 were adjusted to pH 6.0 (at < 10°C) using 85% lactic acid (EM Industries, Inc., Cherry Hill, NJ) that had been diluted 1:2 (vol/vol) with distilled water. The pH of the control milk was not adjusted. The 7-L vats were warmed simultaneously in a water bath to 34°C, inoculated with 0.75 g of each culture, and allowed to ripen for 45 min. After ripening, 3 ml of rennet, diluted in 30 ml distilled water, were added. The curd was cut using 1.9-cm knives at 10 min (for milk with C:F 3.0, 5.0, 7.0, and 8.0) and 50 min (for milk with C:F of 1.2 because it was not pre-acidified) after rennet addition, healed for 15 min, and then gently agitated for 30 s. The temperature of all vats (both control and low fat) was raised to 38°C over 10 min with periodic gentle agitation. The whey was drained at pH 6.0 (whey pH), and the curds were hand-cheddared (turning every 20 min) at 38°C until the curd pH reached 5.2. Curds were cut and stretched in hot water (83°C) until they were elastic and smooth. The stretched cheese (approximately 700 g from each vat) was placed into stainless steel molds (9 x 9 x 9 cm) and immersed in ice water for 30 min to cool. The six resulting cheese loafs from each vat were removed from the molds, immersed in individual
refrigerated (4°C) brines (saturated NaCl, pH 5.0) for 4 h, then individually vacuum-packed. Cheeses were stored at 4°C.

**Chemical and Physical Analysis of Cheese**

Moisture content was measured by microwave oven (model AVC 80; CEM Corp., Matthews, NC), using 50% power for 5 min, and fat content was determined using a modified Babcock method (Richardson, 1985). The extent of melting was measured by the tube test of Olson and Price (1958), modified by increasing the oven temperature to 150°C. Cook color was measured by reflectance colorimetry (Minolta Chroma Meter CR-100; Minolta Corp., Ramsey, NJ) (Oberg et al., 1992b). Total protein (N x 6.38) was measured by the semi-micro-Kjeldahl method [AOAC method 920.123; (AOAC, 1990)] and ash using a dry ash method [AOAC method 935.42; (AOAC, 1990)]. Melt characteristics and AV of melted cheese, cook color, and proteolysis were measured at 1, 7, 14, and 28 d. Curd pH, moisture, total protein, and fat were measured at 1 d.

**Apparent viscosity.** A Brookfield DV II+ helical viscometer (Brookfield Engineering Laboratories, Inc., Stoughton, MA) fitted with a T-bar spindle (T-F with a 9.0 mm crossbar) was used to measure AV of melted cheese using the method of Kindstedt et al. (1989) with modifications. A 25-cm section of rod was added to the helipath rod so that the total distance of travel was 25 cm (total helipath rod height, 50 cm). A jacketed test tube holder was attached to the rod to allow use of a remote water bath. Hot water (80°C) was circulated through the jacket during evaluations to maintain a uniform sample temperature. Fifteen grams of shredded cheese were tamped into a 25-
mm x 150-mm test tube, tempered for 10 min at 80°C, and then placed in the water jacketed test tube holder. The T-bar spindle was lowered until it reached a position just above the bottom of the test tube and the viscometer was activated (1.5 rpm). An IBM-compatible computer, equipped with Brookfield DV Gather + 1.0 software, was used to record AV every 5 s for 10 min. The mean of the readings on the AV profile curve from 0.5 to 1.5 min were reported as AV.

**Gel electrophoresis.** One-dimensional SDS-PAGE was carried out on a PhastSystem™ using PhastGel™ homogeneous 12.5 gels (Pharmacia LKB Biotechnology, Piscataway, NJ). Cheese samples (25 mg) were solubilized by adding 1 ml of Tris (10 mM)-EDTA (1 mM) buffer, pH 8.0, 350 μl of SDS (10%), and 50 μl of β-mercaptoethanol; samples were then placed in boiling water for 5 min, mixed by vortexing for 5 s, and boiled for 5 min (Pharmacia, 1990b; 1990c). Bromophenol blue (3 μl of 4.5%, wt/vol, solution) was added as tracking dye, and 0.5 μl of sample was loaded on the gel (Creamer, 1991). Skim milk, whole casein (CN), αs-CN, β-CN, and κ-CN served as controls. Gels were stained with Coomassie blue-R (Pharmacia, 1990a). Relative peak areas were determined using PhastImage™ software (Pharmacia LKB Biotechnology).

**Statistical Analysis**

Analysis of variance was run for the variables: melt, AV, and cook color. Three independent replicates for each C:F were run using a split plot design; C:F as the whole-plot effect, and storage time was the split-plot effect. Statistical analysis was done using Minitab 7.2 (Minitab Inc., State College, PA).
RESULTS

Composition

Low fat cheeses were made from milks with C:F of 3.0, 5.0, 7.0, and 8.0 that had fat contents between 2.2 and 5.0% (Table 3.1). The low fat cheeses contained from 4 to 6% more protein than the control, but ash contents were similar. The microwave oven method [which had been successfully used in previous studies for part-skim Mozzarella cheese (Oberg et al., 1991; 1992a; 1992b)] estimated the moisture content of all cheeses to be between 51.2 and 54.6%. However, when a mass balance was used to validate the moisture measurements, it was observed that the microwave oven method had underestimated the moisture contents of the low fat cheeses. Probable moisture content (Emmons, 1994) was calculated based on measurements of protein, fat and ash, and

Table 3.1. Mean (±SEM) percentages of fat, protein, ash, moisture contents estimated by a microwave oven method, probable moisture, and moisture in the fat-free cheese (MFFC) in Mozzarella cheeses made from milk of various casein to fat ratios (C:F).

<table>
<thead>
<tr>
<th>C:F</th>
<th>Fat</th>
<th>Protein</th>
<th>Ash</th>
<th>Microwave Oven¹</th>
<th>Probable moisture²</th>
<th>MFFC³</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>X</td>
<td>SEM</td>
<td>X</td>
<td>SEM</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>1.2</td>
<td>19.3</td>
<td>0.4</td>
<td>24.6</td>
<td>0.3</td>
<td>3.0</td>
<td>0.03</td>
</tr>
<tr>
<td>3.0</td>
<td>5.0</td>
<td>0.6</td>
<td>28.2</td>
<td>0.1</td>
<td>2.9</td>
<td>0.01</td>
</tr>
<tr>
<td>5.0</td>
<td>3.1</td>
<td>0.5</td>
<td>30.4</td>
<td>0.4</td>
<td>3.0</td>
<td>0.01</td>
</tr>
<tr>
<td>7.0</td>
<td>3.0</td>
<td>0.4</td>
<td>30.4</td>
<td>0.6</td>
<td>3.0</td>
<td>0.02</td>
</tr>
<tr>
<td>8.0</td>
<td>2.5</td>
<td>0.5</td>
<td>30.1</td>
<td>0.3</td>
<td>3.1</td>
<td>0.01</td>
</tr>
</tbody>
</table>

¹ Microwave oven method for part-skim and reduced-fat Mozzarella cheese.
² Calculated as 100% - (percentage of fat + percentage of protein + percentage of ash + 1.0%). The 1.0% accounts for lactic acid in the cheese and other solids lost during ashing.
³ Calculated as the percentage of probable moisture / (100% - percentage of fat).
allowing 1% for lactic acid and other solids in the cheese that were volatilized during ashing (Table 3.1). On this basis, the moisture contents of the low fat cheeses ranged from 62.5 to 63.6%. Moisture contents were similar even though fat contents ranged from 2 to 5%. When moisture in the fat-free cheese (MFFC) was calculated based on probable moisture content, the MFFC of the low fat cheeses were comparable with the MFFC of the control cheese (all between 64.5 and 66.2%).

**Melt**

When an overall analysis of variance was conducted for the 28-d storage period, melting was not statistically different \( (P > 0.05) \) among the cheeses with different fat contents (Table 3.2). Some differences in melt were observed at individual storage times (Figure 3.1). At d 1, the cheeses melted to the same extent (i.e., \( P > 0.05 \)). After 7 d of storage, the melting improved, but the low fat cheeses melted less well \( (P < 0.05) \) than the control. No further increases in melting were observed. At d 14, the melt of the control cheese was still higher \( (P < 0.05) \) than that of the low fat cheeses, although no difference \( (P < 0.05) \) was observed in the melt among the cheeses after 28 d, perhaps because of the large variance in melting of the control cheese. Variation in melting was less for the low fat cheeses. For all cheeses, melting was significantly affected \( (P < 0.05) \) by storage time.

**Apparent Viscosity**

The AV remained relatively constant (Figure 3.2) as the T-bar spindle raised through the melted cheese (area 1) and, in some instances, increased slightly as the T-bar
Table 3.2. Analyses of variance for melt, stretch, and cook color for part-skim and low fat Mozzarella cheeses as a function of casein to fat ratio (C:F) and storage time at 4°C.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>df</th>
<th>Melt</th>
<th>Stretch</th>
<th>Color</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replicates (R)</td>
<td>2</td>
<td>2.99&lt;sup&gt;NS&lt;/sup&gt;</td>
<td>15.20 x 10&lt;sup&gt;12&lt;/sup&gt;</td>
<td>89.06</td>
</tr>
<tr>
<td>C:F</td>
<td>4</td>
<td>2.28&lt;sup&gt;NS&lt;/sup&gt;</td>
<td>1.30 x 10&lt;sup&gt;12&lt;/sup&gt;NS</td>
<td>57.59*</td>
</tr>
<tr>
<td>R x C:F (error A)</td>
<td>8</td>
<td>0.99</td>
<td>2.20 x 10&lt;sup&gt;12&lt;/sup&gt;</td>
<td>10.80</td>
</tr>
<tr>
<td>Time (T)</td>
<td>3</td>
<td>6.25*</td>
<td>60.80 x 10&lt;sup&gt;12&lt;/sup&gt;**</td>
<td>200.70&lt;sup&gt;NS&lt;/sup&gt;</td>
</tr>
<tr>
<td>R x T (error B)</td>
<td>6</td>
<td>1.04</td>
<td>2.48 x 10&lt;sup&gt;12&lt;/sup&gt;</td>
<td>158.82</td>
</tr>
<tr>
<td>C:F x T</td>
<td>12</td>
<td>0.25&lt;sup&gt;NS&lt;/sup&gt;</td>
<td>0.52 x 10&lt;sup&gt;12&lt;/sup&gt;NS</td>
<td>6.24&lt;sup&gt;NS&lt;/sup&gt;</td>
</tr>
<tr>
<td>R x C:F x T (error C)</td>
<td>24</td>
<td>0.16</td>
<td>0.54 x 10&lt;sup&gt;12&lt;/sup&gt;</td>
<td>8.61</td>
</tr>
<tr>
<td>Corrected total</td>
<td>59</td>
<td></td>
<td></td>
<td></td>
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</tbody>
</table>

<sup>1</sup><sup>P</sup> > 0.05, *<sup>P</sup> < 0.05, **<sup>P</sup> < 0.01

Figure 3.1. Mean (±SEM) melt measurements of Mozzarella cheese (n = 3) made from milk with a casein to fat ratio of 1.2 (solid bar), 3.0 (open bar), 5.0, (diagonal striped bar), 7.0 (horizontally striped bar), or 8.0 (gray bar) during 28 d of storage.
Figure 3.2. Apparent viscosity profiles of low fat (A) and part-skim (B) Mozzarella cheese made from milk with a casein to fat ratio of 7.0 and 1.2, respectively, over 28 d of storage. Solid circle, d 1; open square, d 7; solid triangle, d 14; and open diamond, d 28.
spindle approached and exited the cheese column surface (area 2). Mean AV decreased sharply as the T-bar spindle was raised above the cheese surface (area 3), drawing a strand of cheese behind it. As the T-bar spindle continued to rise, the cheese strand stretched and became thinner.

No significant difference ($P > 0.05$) was found among the AV of the melted cheeses (Table 3.2) although the low fat cheeses tended to have higher AV than did the control cheeses (Figure 3.3). The AV decreased ($P < 0.01$) during storage for all cheeses. The strand characteristics during the AV test were different between the control and low fat cheeses. Stretched low fat cheese appeared to be more fibrous, tougher, less pliable, and more brittle than those formed by the control cheese. Although the visual

![Graph showing apparent viscosity over time](image)

**Figure 3.3.** Mean (±SEM) apparent viscosity of Mozzarella cheese ($n = 3$) made from milk with a casein to fat ratio of 1.2 (solid bar), 3.0 (open bar), 5.0 (diagonal striped bar), 7.0 (horizontally striped bar), and 8.0 (gray bar) during 28 d of storage.
characteristics differed, AV profiles of the low fat cheeses and part-skim control cheese were similar.

**Cook Color**

Cook color was affected \( (P < 0.05) \) by fat content of the cheese (Table 3.2). All low fat cheeses had lower \( b^* \) values (a greenish tint) than the part-skim control cheese. The overall effect of storage time on cook color was not significant \( (P > 0.05) \), although all of the cheeses had lower \( b^* \) values after 28 d of refrigerated storage (Figure 3.4).

**Proteolysis**

Peak densitometry indicated that approximately 50% of the \( \alpha_s \)-CN in all samples was hydrolyzed after 28 d of storage (Figure 3.5). The part-skim Mozzarella cheese (which contained 19.3% fat) had a 52% loss of \( \alpha_s \)-CN. The low fat Mozzarella cheese (containing 5.0, 3.1, 2.5, and 2.2% fat) lost 47, 55, 77, and 61% of the \( \alpha_s \)-CN, respectively, in agreement with the observations of Kiely et al. (1993). Hydrolysis of \( \alpha_s \)-CN in all cheeses, regardless of fat level, was greatest between d 1 and d 14; little change occurred after d 14.

**DISCUSSION**

When a microwave oven is used to dry cheese, the amount of microwave energy absorbed per unit of weight is dependent on the fat, salt, and water contents of the cheese. Low fat cheeses, with less fat and lower salt concentration in the water
**Figure 3.4.** Mean (±SEM) cook color measurements of Mozzarella cheese (n = 3) made from milk with a casein to fat ratio of 1.2 (solid bar), 3.0 (open bar), 5.0 (diagonal striped bar), 7.0 (horizontally striped bar), and 8.0 (gray bar) during 28 d of storage.

**Figure 3.5.** Amount of intact αs-CN, expressed as a percentage of d1, during 28 d of storage at 4°C of Mozzarella cheeses (n = 1) made from milk with a casein to fat ratio of 1.2 (solid bar), 3.0 (open bar), 5.0 (diagonal striped bar), 7.0 (horizontally striped bar), and 8.0 (gray bar).
phase, absorb less energy than higher fat cheeses containing less moisture. Thus, low
fat cheeses would reach a lower maximal temperature than the higher fat cheeses, given the
same microwave power settings, and possibly contribute to underestimating the moisture
content. Increasing the power settings, however, does not correct this underestimation.
(In a follow-up study, the microwave oven method was modified by increasing power
setting from 50% to 100%. The moisture content values increased by 1.5% but still left
unaccounted for 10% of the proximate analysis.) The inability of the microwave oven
method we used to volatilize all of the water in the low fat cheeses indicates that some of
the water remained trapped within the protein matrix. Assuming the difference between
the measured moisture content and the probable moisture (based on protein, fat and ash
content) represents water that is trapped in the protein matrix (as vicinal, multilayer and
entrapped water), then the portion of such water in low fat cheese is much greater than
that in part-skim Mozzarella cheese.

This result demonstrates the importance of measuring other components in cheese
(protein, fat, minerals, and carbohydrates) as well as moisture. Emmons (1994) showed
that cheese moisture content may be underestimated even when using a forced-draft oven
method. Whatever method is used, procedural changes may be necessary when cheeses of
vastly different compositions are being compared, particularly when low fat cheeses are
being compared with full fat cheeses by a microwave oven method.

The melt test measures the ability of cheese particles to flow past one another
when heated. Fat or unbound water acts as a lubricant and increases the ability of cheese
particles to flow (Tunick and Shieh, 1995; Tunick et al., 1993). Assuming that a
relationship exists between how tightly water is bound and how well the cheese melts, the moisture content of low fat cheese becomes a predominant issue in developing low fat cheeses that melt well. Furthermore, even though moisture content in the low fat cheeses (62 to 64%) was higher than in the part-skim cheese (52%), when considered on a fat free basis, the moisture contents were all similar (65 to 66%). Thus, the protein to moisture ratio appears to remain constant even after fat content is reduced from 19 to 2%, reflecting the water-holding capacity of the proteins in cheese. Therefore, the state of the water in the cheese, as well as water on a fat-free basis, needs to be considered when functional properties of low fat Mozzarella cheese are investigated.

The trend of large increases in melting during the first 7 d of refrigerated storage of part-skim Mozzarella cheese, followed by a little increase from d 7 to d 28 (Figure 3.1), is consistent with prior observations (Oberg et al., 1991; 1992a; 1992b). There is, however, a difference in the rate of aging of low fat (2 to 5% fat) versus part-skim (19% fat) cheeses (Merrill et al., 1994; Tunick and Shieh, 1995). Because the rates of hydrolysis of $\alpha_s$-CN were similar (Figure 3.5) and because the fat content did not affect initial melting (Figure 3.1), the explanation for this difference in aging probably lies in some fundamental differences in how the protein and fat are structurally arranged in the cheese. A better understanding of cheese structure, especially on the molecular level, would help elucidate the parameters that control cheese melting.

The AV profiles that are derived from helical viscometry (Figure 3.2) are the combined result of cheese viscosity, the ability of milk proteins to form strands around the T-bar spindle, and the ability of those strands to deform when stretched (Kindstedt
and Kiely, 1990; Kindstedt et al., 1989). For both the control and low fat cheeses, the AV was constant while the T-bar spindle remained in the melted cheese. An increase in the AV as the T-bar spindle exited the cheese could result from partial drying of the cheese surface. Then, as the cheese strand attached to the T-bar spindle is stretched and reduced in thickness, the AV rapidly decreases. Differences in this “tail” section (area 3 of the AV profile) were considerable. The thickness of the strand and the time it remained attached to the T-bar spindle were recorded as differences in height and duration of the AV profile. Cheese that was capable of forming strong, yet elastic strands frequently remained attached to the T-bar spindle for the duration of the test (10 min) with fibers extending up to 25 cm. No correlation was apparent between the amount of cheese pulled up by the T-bar spindle and the extent to which it remained attached to the T-bar spindle. Only the portion of the AV profiles from 0.5 to 1.5 min were thus used to calculate mean AV values. This procedure eliminated variation from differences in surface-hardening and tail sections. However, further study of cheese strand characteristics after the T-bar spindle leaves the melted cheese column may better describe the term “stretch” as it is used by the pizza industry and may help to elucidate rheological characteristics of the cheese.

The primary structural protein in Cheddar cheese is \( \alpha_S \)-CN, which undergoes hydrolysis during refrigerated storage and is associated with textural changes during aging (Lawrence et al., 1987). \( \alpha_S \)-Casein probably performs a similar structural role in Mozzarella cheese. Residual milk-clotting enzymes, endogenous milk protease (plasmin),
and proteolytic activity of starter cultures have been shown (Kiely et al., 1993; Lawrence et al., 1987) to improve meltability through proteolytic degradation of the milk protein matrix. Hydrolysis of β-CN by chymosin proceeds more slowly. Basch et al. (1989) calculated half-lives for the loss of αs-CN and β-CN as 2 and 37 wk, respectively. Kiely et al. (1993) also found that, after 29 d, 50% of the αs-CN in low moisture, part-skim (20% fat) Mozzarella cheese was hydrolyzed, but β-CN remained virtually unchanged.

Interestingly, all five cheeses melted the same on d 1, which suggests that fat content is not the prime factor controlling cheese meltability. The part-skim Mozzarella had 5 to 10 times more fat, but protein to moisture ratios were the same. The marked increase in melt and the decrease in AV of the part-skim Mozzarella cheese during the 1st wk of storage has been assumed to be a function of proteolysis. However, the caseins in the low fat cheese underwent the same degree of proteolysis (or more) yet melting was not improved. Therefore, hydrolysis of αs-CN while probably contributing to improved melting, was not the prime factor in the increase in melting that occurred during storage of Mozzarella cheese. After 7 d, the control cheese showed no increase in melting, but between d 7 and d 14, the disappearance of αs-CN was greatest.

Cook color is influenced both by proteolytic activity (producing amino acids and small peptides) and carbohydrate utilization of starter cultures used for Mozzarella cheese production. Galactose-fermenting strains of *S. thermophilus* and *L. helveticus* reduce the amount of browning during cook compared with nongalactose-fermenting
strains (Olson, 1983). Cook color decreases with time when *L. delbrueckii ssp. bulgaricus* is replaced by *L. helveticus* in Mozzarella cheese starters (Oberg et al., 1992b).

The slightly higher $b^*$ values for the low fat cheeses, although not statistically significant, may have been due to the higher moisture content of the low fat cheeses, which resulted in slightly more retention of lactose in the cheese. Cook color would also be expected to decrease during storage as residual sugars in the cheese are utilized by bacteria.

When cool, the low fat cheeses had a slightly greenish tint that became less noticeable upon heating and returned when cooled. This greenish tint is associated with fewer light-scattering centers (less fat globules) and is a common defect of low fat cheeses. Some manufacturers overcome this by adding a whitening agent, such as titanium dioxide, and others have looked to microparticulated fat replacers as a potential way to impart more opaqueness to low fat and nonfat cheeses.

**CONCLUSIONS**

Low fat Mozzarella cheeses containing between 2 and 5% fat were produced. Their moisture contents were estimated, based on component analysis, to be 61 to 63%. An underestimation of moisture content in the low fat cheeses occurred when a rapid microwave oven test method was used to determine moisture content. This result reinforces the need to confirm moisture measurements by component analysis and mass balance, especially when low fat cheeses are being analyzed. Melting characteristics of the low fat Mozzarella were affected by the reduction in fat. By increasing water retention in low fat Mozzarella cheese, some improvement can be made in melting
characteristics. However, further work is needed to produce low fat Mozzarella cheese that meets the FDA requirements of less than 6% fat and has acceptable functionality during cooking. These low fat cheeses may require the addition of fat mimetics that entrap water and also provide lubricant properties typically provided by fat globules.

REFERENCES


The aim of this study was to determine what happens to water in Mozzarella cheeses during storage and to relate those changes to cheese microstructure and functionality. A reduced-fat (8% fat) Mozzarella cheese and a control cheese with 19% fat were made and evaluated over 21 d of refrigerated storage at 4°C. Fat, protein, ash, salt, and water were measured on d 1. Meltability, total water, freezable water, and expressible water were measured on d 1, 7, 14, and 21. Even though the reduced-fat cheese had a higher total water content than did the control cheese, the reduced-fat cheese contained less water on a fat-free basis. The amount of water expressible at 25°C was higher in the control cheese than in the reduced-fat cheese and was proportional to the fat content of the cheese. During storage, the expressed serum for both cheeses decreased to zero by d 21. Based on changes observed in microstructure of a commercial Mozzarella cheese (19% fat) during storage it was concluded that the expressed water was derived from water contained in the fat-serum channels that were interspersed throughout the protein matrix. The amount of bound water was lower in the control cheese than in the reduced-fat cheese and was proportional to the protein content of the cheese. Bound

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water levels remained constant throughout storage. During storage of the commercial Mozzarella cheese, the fat-serum channels became smaller with the protein matrix expanding into the areas between the fat globules. By d 21, the fat globules were completely encased by the protein matrix. This expansion of the protein matrix in the commercial cheese occurred over the same time span as the decrease in expressible water of the experimental cheese, indicating that the protein matrix was absorbing the water originally located in the fat-serum channels. Because no change in bound water was observed, the water that had been expressible at d 1 was being absorbed into the protein matrix as entrapped water. The meltability of both cheeses increased during storage at the same time as the percentage of entrapped water increased.

INTRODUCTION

When milk is renneted, casein micelles are converted from a stable colloid into a protein gel with individual casein micelles joined together into chains. As cheese making proceeds, and whey is expelled from the curd, the protein chains become thicker through continued association with one another until they form a protein matrix in which water and fat are distributed (Oberg et al., 1993; Schmidt, 1982). For Mozzarella cheese, this process is the same as Cheddar cheese up until the time the Mozzarella cheese curds are transferred to the cooker-stretcher to produce a pasta filata cheese (Oberg et al., 1993) rather than being cheddared (Green et al., 1981).

In pasta filata cheese, the proteins in the cheese curds coalesce into larger strands that are oriented in the direction of stretching. This coalescence results in a redistribution
of the water and fat during stretching and molding with the larger strands (or fibers) of protein being separated by channels containing water, water-soluble cheese components, bacteria, and fat globules (Oberg et al., 1993; Prentice et al., 1993). Changes in the microstructure and functionality of Mozzarella cheese during storage (Cooke et al., 1995; Kiely et al., 1993; Paulson et al., 1998) suggest that the proteins are not in a quiescent state immediately after stretching and molding but undergo further structural rearrangement.

The improved meltability of Mozzarella cheese observed during storage is generally associated with the proteolytic hydrolysis of $\alpha_{s1}$-CN by chymosin, plasmin, and proteolytic enzymes originating from the starters (Fox et al., 1979; Kiely et al., 1993; Kindstedt and Guo, 1997; McMahon et al., 1993). Hydrolysis of $\beta$-CN in cheese made using chymosin occurs at a much slower rate and is not usually associated with improved meltability. However, Bogenrief and Olson (1995) demonstrated that the meltability of Cheddar cheese correlated more with $\beta$-CN hydrolysis than with $\alpha_{s1}$-CN hydrolysis. In cheese made using Cryphonectria parasitica rennet, the meltability was greater, at all stages of maturation, compared to cheeses made with chymosin. They attributed the improved meltability to increased $\beta$-CN hydrolysis.

It has also been shown (Yun et al., 1993) that increased $\beta$-CN hydrolysis improves cheese meltability, increases free oil, and decreases apparent viscosity in Mozzarella cheese. However, we have observed (unpublished data) that changes in cheese meltability can occur during storage without any significant decrease in intact $\beta$-
CN levels. This suggests that proteolysis may not be the only causal effect for changes in the meltability of Mozzarella cheese during storage and that other changes occurring concomitantly in the cheese during storage should be investigated. One such change that has been observed, as shown by a decrease in expressible serum, is a redistribution of water in cheese during storage (Guo and Kindstedt, 1995). However, it is not known what causes this change in water or what subsequent role water plays in cheese functionality.

The various states in which water may exist in a material such as cheese can be described in terms of the spatial relationship between water and the solid constituents of the food (in cheese, these solids are predominantly protein although water interactions also exist between other cheese constituents). Bound water has been characterized as nonsolvent water or chemisorbed water (Geurts et al., 1974); sorbent- or solute-associated water and unfreezable water (Berlin, 1981); constitutional water (i.e., water that occupies interstitial protein spaces); vicinal water (i.e., water that covers a protein as a single layer) (Fennema, 1985), and also characterized in terms of hydrodynamically influenced layers of water that surround a protein (Farrell, 1988). Such isotropically bound water is intimately associated with (or is in close proximity to) the protein surface. This water exhibits slower rotational and translational speeds than pure water, is not available as a solvent, and cannot be frozen at -40°C (Farrell et al., 1989).

In contrast, bulk water is water that is more loosely associated with the proteins even though it retains a large solvent capacity and is freezable at -40°C. Farrell et al. (1989) described bulk water as being anisotropic, because the rotational and translational
speeds of such water molecules are not significantly effected by their interaction with the proteins. Bulk water may be further divided into entrapped or free water depending upon its mobility within the protein matrix. Water that is impeded by the macrostructure of the protein matrix, such that it cannot be expressed by centrifugation, is considered to be entrapped. Water that is not impeded by the protein matrix and may be expressed by centrifugation is known as expressible water (Fennema, 1985). Thus, the distribution and movement of water in cheese may be characterized as being bound to proteins, entrapped by those proteins, or expressible by centrifugation.

The objective of this research was to explain changes observed in Mozzarella cheese microstructure and functionality over time in terms of changes in water distribution in the cheese and to determine whether those changes are the same for cheeses with high or low fat levels.

**MATERIALS AND METHODS**

**Milk, Cultures, and Rennet**

Skim milk and nonhomogenized 2% milk from the Gary H. Richardson Dairy Products Laboratory at Utah State University were pasteurized at 80°C for 29 s, cooled to 4°C, and used to standardize cheese milks to a casein to fat ratio of 1.2 or 2.4. Lyophilized direct-set cultures of *Lactobacillus helveticus* LH 100 and *Streptococcus thermophilus* TA 061 were obtained from Rhodia, (Madison, WI), and double-strength Chymax® rennet from Chris Hansen, Inc. (Milwaukee, WI).
Mozzarella Manufacturing Procedure

Double-O vats, with a capacity to hold 454 kg of milk per vat (DEC International, Damrow, Fond DuLac, WI) and equipped with vertical agitator and knives and Para Just AC motor speed controls (Parametrics, Orange, CT) were used to manufacture cheese curds. Curds were mechanically stretched in a cooker-stretcher (model ALIS LAB; Alfa-Laval Cheese Systems Ltd., Tetrapak, Inc., Chicago, IL), and the melted cheese formed into rectangular loaves (Alfa-Laval Aiform; Tetrapak, Inc., Chicago, IL).

The control (i.e., low-moisture part-skim Mozzarella cheese) and reduced-fat Mozzarella cheeses were made by modifying the reduced-fat Mozzarella cheese-making procedure described by Merrill et al. (1994). After preacidification to pH 6.1 and adjustment of the milk temperature to 34°C, 20 units of each culture were added, and the milk was ripened for 45 min. Thirty-five milliliters of rennet (diluted in 350 ml distilled water) were added, and the coagulated milk was cut 15 min after rennet addition. Curds were healed for 5 min and then agitated for the remainder of the cheese-making procedure. After healing, the temperature was raised to 39°C over 30 min; then, approximately half of the whey was removed. When the curd pH reached 5.2, the remaining whey was drained, and curds were salted (1 kg of salt/1000 kg of milk) in three increments, 10-min apart. The salted curds were cooked and stretched at 80°C in 5% (wt/vol) brine solution and then formed into 2.2-kg rectangular loaves. The loaves were immersed in cold water for 1 h, cut longitudinally in half, individually vacuum-packaged, and stored at 4°C.
Cheese Composition

Fat was measured in duplicate using a modified Babcock method (Richardson, 1985). Cheese moisture was measured in triplicate by vacuum-oven AOAC method 926.08 (AOAC, 1990). Ash was measured in duplicate by gravimetric method AOAC 935.42, protein in duplicate by AOAC method 920.123, and salt in duplicate by chloride analysis by AOAC method 971.19 (model 926 salt analyzer; Corning, Medfield, MA) (AOAC, 1990). Moisture content as measured by vacuum oven was used as total moisture for all calculations of water partitioning in the cheese.

Melt

Melt was measured by modifying the method of Bogenrief and Olson (1995). Cheese plugs weighing approximately 15 g were placed into glass tubes, which were sealed with rubber stoppers. Sample tubes were immersed in hot (95°C) mineral oil, and the distance (mm) the cheese melted was measured at 0, 4, 8, 12, and 16 min. Meltability measurements at 12 min were used to compare control and reduced-fat cheeses.

Water

Freezable and bound water. Freezable water, defined as water freezable at -40°C, was measured using a differential scanning calorimeter (DSC) (model 2910; TA Instruments Inc., New Castle, DE) equipped with a refrigerated cooling system. Nitrogen was used to purge the DSC cell (50 ml/min) and refrigerated cooling system (150 ml/min) prior to and during sample analysis. Cheese samples, approximately 10 to 20 mg, were
accurately weighed, then sealed in aluminum pans (TA Instruments Inc.), and placed inside the DSC cell. A pan containing indium, which served as the reference, was also placed inside the DSC cell. Both pans were simultaneously equilibrated to 25°C, cooled at the rate of 10°C/min to -40°C, and then heated at a rate of 10°C/min to 40°C. Universal Analysis 1.5B software (TA Instruments Inc.) was used to measure the area under the ice melting peak, and the enthalpy (Joules per gram) of the transition determined. The percentage of freezable moisture in the sample was then calculated using the latent heat of fusion of water (334.4 J/g) (Ollivon, 1991). Bound water was defined as the water that did not freeze at -40°C (Berlin, 1981).

Expressible water. Samples were centrifuged at 12,500 x g for 75 min (25°C) (Guo and Kindstedt, 1995). The expressed fluid was decanted into a weighing pan, and the fat was removed using a micro-pipette. The expressible water was calculated as the expressed fluid minus fat.

Entrapped water. Entrapped water was calculated as the difference between the freezable water and the expressible water.

Statistical Analyses

Cheese was made in duplicate on two different days with two vats of cheese (a control cheese and a reduced-fat cheese) being manufactured on the same day. Differences between means were tested using Students t test. Significance was declared at \( P < 0.05 \) unless otherwise stated.
Cheese Microstructure

In a separate experiment, scanning electron microscopy was used to examine changes in the microstructure of a commercially prepared low moisture part-skim Mozzarella cheese (19% fat, 48.8% moisture, 1.3% salt, with a pH of 5.2) during storage. Samples were obtained at 1, 7, 14, and 21 d and prepared for electron microscopy using the method described by Oberg et al. (1993).

RESULTS AND DISCUSSION

Cheese Microstructure

*Initial structure.* As the commercial Mozzarella cheese curd was heated, stretched, and molded, the proteins formed into fibers that were oriented in a roughly parallel manner as shown in Figure 4.1 and as previously reported (Oberg et al., 1993). While the cheese was still hot (ca. 54°C), the proteins had the appearance of continuous, interconnected, smooth-walled fibers separated by channels that contained molten fat globules, serum, bacteria, and water-soluble cheese components. Although most of the fat and bacteria were removed during sample preparation for electron microscopy, some bacteria (along with residual fat globule membrane material) could still be found within the fat-serum channels between the protein fibers of the cheese.

*d 1.* After the commercial cheese had been brined, the appearance of the protein matrix surface surrounding the fat-serum channels changed (Figure 4.2). The original smooth appearance of the fat-serum channel walls, present when the cheese was hot, changed to having a rough-textured appearance. These channel walls were textured
Figure 4.1. Scanning electron micrograph of commercial part-skim Mozzarella cheese taken immediately after hot-water stretching. Cheese matrix proteins appear as smooth, elongated fibers separated by serum channels, which once contained fat, water, and bacteria. Bacterial cells and residual fat globule membrane material adhere to the fat-serum channel walls.

Figure 4.2. Scanning electron micrograph of commercial part-skim Mozzarella cheese 1 d after cooling and brining. Fat-serum channel walls show indentations formed by solidified fat globules of varying size and starter bacteria. Bacterial cells and residual fat globule membrane material adhere to the fat-serum channel walls.
by numerous circular indentations that ranged in size from approximately 1 to 10 μm in diameter. In addition, there were also many smaller circular and elliptical indentations approximately 0.5 to 1 μm in diameter. The larger indentations corresponded to the size of fat globules, and the smaller indentations corresponded to that of the cocci starter culture. Some cells (often as diplococci) were observed to still occupy some of these indentations.

Based on these observations, it appeared that the protein matrix pressed upon the rigid components of the fat-serum channels and molded around any solid object adjacent to the fat-serum channel walls. While the cheese was still warm, the fat globules provided no resistance, because the fat was still molten, and left no indentations in the fat-serum channel walls. However, as the cheese cooled, the fat globules solidified and acted as a template around which the pliable protein matrix molded.

It was also concluded from the formation of these indentations that the fat globules were closely packed in the fat-serum channels. If not, they would have provided little resistance to the protein matrix, and very shallow, if any, indentations would have been formed. Therefore, fat appears to have the important function during the cooking and stretching steps of the cheese-making process of interrupting the fusion of the protein matrix and providing space in which excess serum can be retained.

Prior to heat treatment, the cheese curd consists of a protein matrix that contains randomly distributed pockets of serum and fat (Ollivon, 1991). When the curd is heated and stretched, the protein matrix becomes molten and starts to fuse together, most likely through hydrophobic interactions. This fusion continues unless it is interrupted by any
material that is incompatible with the protein matrix, such as fat globules, bacteria, or microparticulated fat replacers (McMahon et al., 1993). The mixing that occurs during stretching then causes the fat globules and much of the bacteria to be pushed together into increasingly smaller spaces until the fat globules are sufficiently packed to resist the pressure from the protein matrix. Thus, closely packed pockets of fat globules and bacteria are formed that are then oriented into channels between the protein fibers as the molten cheese is extruded and molded into its final shape. Consequently, the size and amount of fat-serum channels that are present in Mozzarella cheese will be a function of fat content of the cheese rather than the moisture content. Any excess moisture would be pushed out of the cheese until the fat globules are closely packed, which is one reason why it is difficult to increase the moisture content of lower fat Mozzarella cheese and not have continued syneresis of serum from the cheese after packaging.

d 7 through 21. When the microstructure of the commercial cheese was examined during refrigerated (4°C) storage, the appearance of the fat-serum channels continued to change (Figures 4.3 to 4.5). The fat globule impressions in the protein matrix surface of the channel walls became more pronounced and changed from small indentations to large depressions. By d 7 of storage, the protein matrix extended 0.5 to 1.0 μm into the fat-serum channels, so that the spherical shape of the fat globules were distinctly defined. After 14 d, the protein matrix had loosely surrounded the fat globules. Thin strands of protein material, encroaching from all sides, connected the fat-serum channel walls and partially occupied the space previously filled by the interstitial serum between the
Figure 4.3. Scanning electron micrograph of commercial part-skim Mozzarella cheese after 7 d of storage at 4°C. Fat globule indentations are more pronounced than at d 1, indicating that the cheese protein matrix has expanded into the fat-serum channels.

Figure 4.4. Scanning electron micrograph of commercial part-skim Mozzarella cheese after 14 d storage at 4°C. Thin fibers of cheese protein matrix surround many fat globules and occupy the inter fat globule spaces once filled by serum.
Figure 4.5. Scanning electron micrograph of low (A) and high (B) magnification of commercial part-skim Mozzarella cheese after 21 d of storage at 4°C. The hydrated cheese protein matrix fills the spaces between the solidified fat globules. Impressions of discrete fat globules attest to the completeness of the cheese protein matrix hydration and subsequent expansion into the fat-serum channels. Starter bacterial cells embedded in the matrix are evident.
closely packed fat globules. In the micrographs of d-14 cheese, the fat-serum channels had a honeycomb appearance.

By d 21, the interstitial spaces between the fat globules of the commercial cheese appeared to be completely filled by the protein matrix. Rather than being contained within fat-serum channels, the columns of fat globules were completely encased within the protein matrix. From the micrographs, it could be concluded that there had been no change in the position of the fat globules during storage; rather, any changes in cheese microstructure during storage were a result of the redistribution of protein and water.

**Cheese Composition**

Composition of the cheeses made for the study of moisture status in cheese is shown in Table 4.1. Actual fat contents for the cheeses were within the range expected for low-moisture part-skim Mozzarella cheese (i.e., 30 to 45 % fat dry weight) and its 50%-fat reduced equivalent. This fat reduction corresponded to increases in the protein, ash, and moisture contents that were similar to increases in other reduced-fat cheese made using this method (Fife et al., 1996).

Interestingly, despite the higher moisture content in the reduced-fat cheese, the moisture was 3.4% lower when expressed on a fat-free basis. When compared on a protein basis, the amount of water in the cheese decreased from 1.9 g of water/g of protein to 1.6 g of water/g of protein, which would probably have an influence on meltability of the reduced-fat cheese. The increase in the ash content is probably a result of increased protein content, which would increase protein-bound minerals such as colloidal calcium
Table 4.1. Mean (±SEM) percentages of fat, protein, ash, salt, moisture, and moisture in fat-free component (MFFC) and pH in part-skim and reduced-fat Mozzarella cheese after 1 d of refrigerated (4°C storage).

<table>
<thead>
<tr>
<th></th>
<th>Fat</th>
<th>Protein</th>
<th>Ash</th>
<th>Salt</th>
<th>Moisture</th>
<th>MFFC</th>
<th>pH</th>
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<tr>
<td></td>
<td>X</td>
<td>SEM</td>
<td>X</td>
<td>SEM</td>
<td>X</td>
<td>SEM</td>
<td>X</td>
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<tr>
<td>Part-skim</td>
<td>19.2</td>
<td>0.9</td>
<td>26.4</td>
<td>0.7</td>
<td>2.6</td>
<td>0.1</td>
<td>1.4</td>
</tr>
<tr>
<td>Reduced-fat</td>
<td>8.0</td>
<td>0.9</td>
<td>32.7</td>
<td>0.1</td>
<td>3.2</td>
<td>0.0</td>
<td>1.4</td>
</tr>
</tbody>
</table>

Note: All values are expressed as mean (±SEM) percentages.
phosphate, and increased moisture (replacing fat) which would increase the total amount of soluble minerals such as ionic Ca, Na and K. Because of its higher moisture content, the reduced-fat cheese had a slightly lower salt-in-moisture content (2.6%) compared to the part-skim cheese (2.8%).

**Water Partitioning**

*Expressed serum.* When expressed on the basis of cheese weight or total moisture content (Figure 4.6), the control cheese contained twice as much expressible serum than the reduced-fat cheese. The control cheese contained 13.3 g of expressed serum/g of cheese compared to 6.2 g of expressed serum/g of cheese in the reduced-fat cheese. This represents 54% of the water in the control cheese but only 22% of the water in the reduced-fat cheese. When the fat content of the cheeses was taken into account it was found that there was no significant difference \( P > 0.40 \) between the cheeses and that on average they contain 0.75 g of expressed serum/g of fat. This further implies that the serum that can be expressed from cheese by centrifugation is associated in some manner with the fat globules.

During storage, the amount of expressible serum decreased so that by d 14, very little serum was expressed, and by 21, there was no expressible serum (Figure 4.6). Guo and Kindstedt (1995) have observed this trend. They reported that 30 to 40% of the water in low-moisture part-skim cheese was expressible after 2 d of storage. Reduction of the expressible serum to 0% occurred by d 8, d 12, and d 16 in the three commercially
Figure 4.6. Percentage of moisture in control (open square) and reduced-fat (closed square) Mozzarella cheese that was bound (A), expressed (B), or entrapped (C) during 21 d storage at 4°C.
prepared cheeses that they analyzed. Based on our observations and those reported by Guo and Kindstedt (1995), it appears that the majority of the expressible water initially in Mozzarella cheese becomes impeded by the macrostructure of the protein matrix during the first 2 wk of storage at 4°C.

Guo and Kindstedt (1995) had concluded from increases in protein concentration in the expressed serum, that a continual solubilization of casein (mainly β-CN) from the protein matrix into the expressed serum had occurred. Subsequently, they proposed that the solubilization of casein resulted from changes to the protein brought about by addition of salt to the cheese and that the serum in the fat-serum channels became converted into a hydrated paracasein gel (Kindstedt and Guo, 1997). From our observations of changes in microstructure during storage of commercial Mozzarella cheese, a different conclusion was made. It appears that serum, along with the protein contained therein, is absorbed into the protein matrix and becomes an integral part of the protein matrix by d 21. There was no observable discontinuity between the material that filled the serum channels and that of the protein fibers (Figure 4.5B).

In an effort to understand the processes occurring during cheese storage, we reexamined the data reported by Guo and Kindstedt (1995). When expressed on a mass basis rather than a concentration basis, it became apparent that there was an initial increase in amount of caseins which was then followed by a gradual decline in protein as the volume of expressed serum decreased. The solubilized protein was predominantly β-CN which has increased solubility at cold temperatures (Farrell, 1988) as does $\alpha_s$-CN to
a lesser extent. This solubilization is probably also from changes to the protein brought about by addition of salt to the cheese as suggested by Kindstedt and Guo (1997).

**Bound water.** On d 1, the control cheese contained 18.1 g of bound water/100 g of cheese while the reduced-fat cheese contained 23.8 g of bound water/g of cheese. A similar increase in bound water content with decrease in fat content was shown when the bound water was calculated as a percentage of the total moisture in the cheese (Figure 4.6). However, when the protein content of the cheeses was taken into account it was found that there was no significant difference ($P > 0.50$) between the cheeses, and that on average they contained 0.71 g of bound water/g of protein.

During the 21 d storage, the level of bound water in cheese remained constant. Therefore, the process of transfer of water from the fat-serum channels into the protein matrix was not being driven by an increase in the amount of water that was chemically bound to the proteins. Instead, there must be some other driving force that is causing the transfer of water into the protein matrix.

**Entrapped water.** At d 1 of storage, the reduced-fat cheese contained a greater proportion of its water as entrapped water than the control cheese (Figure 4.6). This was expected because (i) the higher protein content of the reduced-fat cheese means there is more protein matrix per unit weight of cheese, and (ii) the higher fat content of the control cheese results in the presence of more fat-serum channels interspersed throughout the protein matrix of the control cheese than occurs in a reduced-fat cheese (Oberg et al.,
During storage the amount of entrapped water increased for both cheeses indicating that the expressible water was being absorbed into the protein matrix.

Recently, Cooke et al. (1995) demonstrated that the spaces between electron-dense centers (i.e., the protein sub-aggregates that comprise the backbone of the protein matrix) shown in transmission electron micrographs of low fat Mozzarella cheese increased with storage time (6 wk). They postulated that the increase in spacing, as well as an increase in the size of the electron-dense center, was a result of a reorganization of the protein matrix in response to proteolysis. More recently, Paulson et al. (1998) showed that the spaces between electron-dense centers (protein sub-aggregates) of nonfat Mozzarella cheese changed in response to salt concentration. It is our hypothesis, that the increase in spacing between protein sub-aggregates results from a reorganization of the protein matrix brought about by salting of the cheese and subsequent absorption of water from the fat-serum channels into the protein matrix during refrigerated storage. The spaces between the protein sub-aggregates would represent regions of entrapped water within the protein matrix.

Because of the larger reservoir of water contained in the fat-serum channels of the control cheese, there was more water that could be absorbed into the protein matrix in the control cheese than in the reduced-fat cheese. Although physically impeded by the macrostructure of the protein matrix, entrapped water is available to participate in both protein hydration and act as a solvent. Such entrapped water is considered to be part of the bulk water phase in cheese as the water molecules retain the anisotropic properties of
free water even though they cannot be expressed by centrifugation (i.e., their rotational and translational speeds are not significantly affected by their interactions with the proteins). If it is assumed that there is no difference in the proteins between the control and reduced-fat cheese, the challenge for the cheese maker is to produce reduced-fat cheeses with the same water to protein ratio as the reference cheese. To do this would require that a similar reservoir of water was initially present in the reduced-fat cheese even though the volume occupied by fat-serum channels decreases as fat content is lowered.

**Melt**

The meltability of the control and reduced-fat cheese during 21 d storage is shown in Figure 4.7. The control cheese melted to a greater extent than did the reduced-fat cheese although the difference between the cheeses diminished as storage time increased. These observations are similar to previously reported trends for low moisture part-skim, reduced-fat, and low fat Mozzarella cheeses (Fife et al., 1996; Guo and Kindstedt, 1995; McMahon et al., 1996; Merrill et al., 1994; Tunick and Shieh, 1995).

The largest increase in meltability of the control cheese was from d 1 to 7, followed by minimal increase from d 7 to 21. The meltability of the reduced-fat cheese also increased during storage with the largest increase occurring from d 1 to 14. There was only a slight change in meltability of the reduced-fat cheese from d 14 to 21. This corresponds to the transfer of water from the fat-serum channels to the protein matrix of the reduced-fat cheese being virtually complete by d 14.
Figure 4.7. Meltability after 12 min at 95°C of control (open bar) and reduced-fat (solid bar) Mozzarella cheese that was stored for 21 d at 4°C.

The improvement in cheese meltability during the first 21 d of storage can also be explained in terms of changes in water and protein states in the cheese. At the beginning of storage, the cheeses had the least meltability. This suggests that, when heated, there are relatively strong interactions maintained between protein molecules within the protein matrix so that they resist the tendency to flow; even though expressible water and fat are present within the fat-serum channels.

Thus, the presence of considerable free water and fat in the fat-serum channels is not enough to ensure good meltability. During the initial weeks of storage, water is transferred from the fat-serum channels into the protein matrix as the proteins become
more hydrated, and some interactions between proteins are replaced with interactions of proteins with the bulk phase water molecules. This increased hydration of proteins has been attributed to salting-in of proteins in the cheese (Paulson et al., 1998). During most of the cheese-making process, the protein matrix of the renneted curd is dehydrated through the action of cutting, agitation, heating, and acidification of the curd. It appears (Paulson et al., 1998) that the presence of added salt results in the protein becoming more hydrated and less tightly aggregated. As the proteins that constitute the matrix became more hydrated, their hydrodynamic volume increases and the matrix begins to extend into the spaces between fat globules in the fat-serum channels. In regard to the functionality of the cheese, a more hydrated protein structure would allow the proteins to slip past one another more easily and, when combined with the lubricating properties of the fat (Tunick et al., 1993; Tunick and Shieh, 1995), result in improved meltability. Thus, the overall meltability of Mozzarella cheese can be considered as the combined effects of (i) fat content and (ii) the balance between protein-to-protein interactions and interactions between proteins and the bulk water entrapped within the protein matrix.

An initial high level of interactions between proteins would restrict the ability of the proteins to flow when heated. Then, as expressible water is absorbed into the protein matrix, increased protein hydration allows the proteins to flow more easily when heated and results in improved meltability. Further improvements in meltability could therefore be obtained by allowing the protein matrix to become fully hydrated. It would appear that this state of full hydration was not reached in the reduced-fat cheese because the
initial reservoir of water contained in the fat-serum channels was insufficient for the quantity of protein present in the cheese.

CONCLUSIONS

During the first 3 wk of storage, the expressible water contained within the fat-serum channels of Mozzarella cheese was absorbed into the cheese protein matrix where it became entrapped water. Presumably, this change was caused by a rearrangement of the protein molecules contained within the protein matrix that was induced by salting and refrigeration of the cheese. The water absorption was accompanied by a swelling of the protein matrix, which continued until the spaces between the fat globules were completely filled with matrix proteins. The increase in entrapped water during storage was limited in the reduced-fat cheese because of the lower volume of expressible water that was initially present in the fat-serum channels. Improvements in the meltability of the cheese occurred at the same time as the matrix proteins became more hydrated. This implies that a reduction in the extent of protein-to-protein interaction within the protein matrix is a principal reason for increased meltability of mozzarella cheese during storage.

REFERENCES


CHAPTER 5
TEST FOR MEASURING THE STRETCHABILITY OF MELTED CHEESE

ABSTRACT

The purpose of this research was to develop an objective test for measuring the stretchability of melted Mozzarella cheese. Three nonfat and four low-moisture part-skim cheeses were obtained from commercial sources or manufactured at Utah State University. Each cheese was analyzed for fat, protein, moisture, Ca, and Na. Functional characteristics of melt were measured by a modified tube test, and stretch by both helical viscometry and a fork test used by commercial manufacturers of pizza cheese. Values obtained from these tests were compared with the new stretch test that used a texture profile analyzer. To determine the proper incubation temperature for the new stretch test, cheese was placed into a stainless steel cup and tempered (30 min) in a water bath at either 60, 70, 80, or 90°C until melted. The tempered cup was placed in a water-jacketed holder mounted on the texture profile analyzer. A three-pronged probe was lowered into the melted cheese and pulled vertically until all of the melted cheese strands broke or the maximum beam stroke was reached. Melt strength, the maximum load (g) obtained during the tensile test, stretch quality, the average load (g) as cheese fibers stretch and elongate and stretch length were determined. There was a significant difference in stretch quality between the nonfat and low-moisture part-skim cheeses.
There was also a significant difference in stretch quality among the nonfat and low-moisture part-skim cheeses. Melt strength correlated with apparent viscosity. Other melted cheese parameters were independent of one another. This elongation stretch test, along with traditional melted cheese tests, provides more complete information about the functional properties of Mozzarella cheese.

INTRODUCTION

One of the most important characteristics of Mozzarella cheese used as pizza topping is the ability to stretch when melted. Stretch refers to the ability of melted cheese to form fibrous strands that extend under tension (Kindstedt, 1995). Several methods have been used over the years to evaluate the elongational properties of cheese (Ak et al., 1993; Ak and Gunasekaran, 1995; Cavella et al., 1992). One method used by commercial cheese manufacturers for evaluating the stretchability of Mozzarella cheese is the fork test (U.S. Department of Agriculture, 1980). This test is performed by placing shredded cheese on a pizza crust covered with pizza sauce. After baking, a fork is inserted into the melted cheese and raised vertically until the attached cheese strands break. Strand length when the majority of the strands finally break represents the stretchability of the cheese. Although test parameters are controlled, most technicians do not perform the fork test uniformly. Variations occur in where and how the fork is inserted, tine orientation, amount of tine covered by the cheese, and the speed at which the fork is lifted. In addition, defining the point at which the “majority” of the attached
strands break depends on the judgment of the technician. For these reasons, it is not uncommon for evaluations of the same cheese to differ when the fork test is utilized.

Other stretch tests have been developed to reduce the subjective nature of the fork test. Apostolopoulos (1994) developed a tensile test that mimicked the way consumers assess cheese stretchability when they eat a pizza. A circular plate was used to hold the pizza crust. A smaller circular piece (with a vertical rod attached) was cut out allowing the center of the plate to be raised independently of the outer rim. A similarly cut pizza crust was placed on the plate and a standard weight of cheese sprinkled on top of the crust. The complete apparatus was heated in a microwave oven for 15 s to melt the cheese. On removal from the oven, the vertical rod was attached to the head of a tensile testing machine and pulled vertically, stretching the melted cheese. The extensibility of the cheese was recorded as the distance of travel until all the cheese strands broke. A comparison to sensory evaluations indicated that the extensibility test correlated well with the way consumers evaluate cheese on a pizza. More recently, Guinee and O’Callaghan (1997) modified the method so that the two halves of a cheese-covered pizza were pulled horizontally, rather than vertically. Stretch was defined as the distance the cheese stretched until complete strand failure.

Another way cheese stretchability was evaluated was to use a helical viscometer. Helical viscometry measures the torque on a rotating T-bar spindle as the spindle is raised through a column of melted cheese. The amount of torque depends on the strain rate employed during the test, therefore, torque units are expressed as apparent viscosity
(AV) (van Vliet, 1991). Lee et al. (1978) used helical viscometry to correlate melted cheese characteristics, sensory evaluations, and texture profile analysis of eleven cheeses, including Mozzarella cheese. Kindstedt et al. (1989a) investigated the application of helical viscometry for measuring melted cheese properties of Mozzarella cheese and developed a test protocol. Kindstedt et al. (1989b) demonstrated the usefulness of this method by comparing the apparent viscosities of Mozzarella cheese manufactured by two different cheese plants. Oberg et al. (1991) used helical viscometry to study the effect of proteolytic activity on the physical properties of Mozzarella cheese, including stretchability, over 28 d refrigerated storage. They used helical viscometry to measure differences in apparent viscosity of Mozzarella cheeses manufactured using different milk-clotting enzymes and to determine the effects of freezing, thawing, and shredding on the stretchability of low-moisture part-skim Mozzarella cheese (Oberg et al., 1992).

In addition to the information obtained from AV measurements while the T-bar spindle remains in the cheese, information on stretch characteristics could be obtained as cheese stretches above the melted cheese pool (Kindstedt et al., 1989b; Oberg et al., 1991). The purpose of this research was to develop an objective test for measuring the stretchability of melted Mozzarella cheese that would allow cheese makers and pizza makers to predict how cheese would perform as a pizza topping.
MATERIALS AND METHODS

Cheese

Three nonfat (NF) and four low-moisture part-skim (LMPS) Mozzarella cheeses were used to evaluate the stretch test. One NF cheese, cheese NF-3, was manufactured at Utah State University using direct acidification. The remaining cheeses were obtained from commercial sources. With the exception of cheese NF-3, the manufacturing dates of the commercial cheeses were unknown, although each cheese was coded with an expiration date. Since our objective was to determine whether the USU stretch test could be used to differentiate between the stretchability of the various cheeses, regardless of cause, storage-related events such as proteolysis were not monitored. Each cheese was tested for fat, moisture, protein, and mineral content. Fat was measured in duplicate using a modified Babcock method (Marshall, 1992). Cheese moisture was measured in triplicate by vacuum-oven AOAC method 926.08 and protein measured in duplicate by AOAC method 920.105 (AOAC, 1990). Mineral content was measured using inductively coupled plasma-atomic spectroscopy (U. S. EPA, 1992).

Cheese Functionality

Melt was measured using a modified (oil bath, 95°C) tube test method (McMahon et al., 1999). The distance the cheese flowed at 12 min was used to compare cheese meltability. Apparent viscosity, an indicator of cheese stretchability, was measured using the method of Kindstedt et al. (1989a), with modifications by Fife et al.
(1996) in which AV was measured while the T-bar was fully immersed in the molten cheese. A fork test, used by pizza cheese manufacturers, was also used to evaluate cheese stretchability (Figure 5.1). For each cheese, 114 g of pizza sauce (Kraft Food Service, Inc., Glenview, IL) was evenly distributed on a thawed 30-cm diameter pizza crust (Krusteaz®, Continental Mills, Seattle, WA) followed by 284 g of shredded cheese. Cheese was shredded using a Professional Salad Shooter (National Presto Ind., Inc., Eau Claire, WI). Pizzas were baked at 250°C for 6 min in a conveyer-style oven.

Figure 5.1. Fork test as used by pizza cheese manufacturers. Fork is inserted into melted pizza topping and lifted vertically until the cheese strands break. The distance the cheese strands extend, as measured on a ruled scale, is defined as stretch.
Baked pizzas were allowed to cool at room temperature (approximately 22°C) for 1.5 min. A fork was inserted into the melted cheese then lifted vertically until the cheese strands broke. The degree to which the cheese strands extended was defined as stretch. The difference in cheese melt among samples was compared using one-way ANOVA. The difference in cheese apparent viscosity was also compared using one-way ANOVA. If the F-test for the model was significant ($P < 0.05$) the means were compared using the least significant difference test ($P < 0.05$).

**USU Stretch Test**

A Stevens Farnell Quality Testing System (Model 25, Stevens Farnell, Dunmorow, UK) in tensile mode was used to evaluate the cheese stretchability. A water-jacketed sample cup holder was mounted to the base of the instrument and attached to a circulating water bath (Techne Inc., Prinston, NJ) fitted with a Tempunit® TU-16D temperature control (Figure 5.2). The water bath was used to temper the cheese samples prior to testing and maintain sample temperature in the steel cups during the stretch test.

Cheese plugs (height: 3.5 cm; diameter: 3.0 cm) were placed into stainless steel cups (height: 5.0 cm; diameter: 3.4 cm), tempered in the water bath for 30 min (at 60, 70, 80 or 90°C) (Figure 5.3), then placed in the water-jacketed sample cup holder (Figure 5.4). A three-prong spindle (see Figure 5.5) was lowered into the melted cheese until it was 0.3 cm from the bottom of the cup. The sample cup was rotated 1/6 turn (to push
Figure 5.2. Stevens Farnell Quality Testing System shown with water-jacketed sample holder, and tri-probe. Sample holder was attached to the instrument base by two C-clamps. Wing nuts on sample holder were used to secure the sample cup.

Figure 5.3. Sample cups containing cheese temper in a water bath. Slotted wings were used to secure the cups to the sample cup holder. It was determined that cheese plugs, rather than ground cheese, provided more homogeneous melted cheese texture and therefore more consistent results than did ground cheese samples. Aluminum cover reduced evaporation.
Figure 5.4. Water-jacketed sample holder, sample cup containing molten cheese, and tri-probe. Sample cup was positioned such that the sample cup wings were oriented approximately 45 degrees from the wing nut posts. After lowering the probe into the cheese, the sample cup was rotated clockwise (white arrows) until the sample cup wing slots rested securely against the wing nut posts. The wing nuts were then tightened, securing the sample cup in place. By orienting the sample cup in this fashion, an uncut area of the cheese was provided for evaluation.
Figure 5.5. Schematic view of tri-probe used for USU stretch test. Scale (mm). All hooks 15 mm in length, radius (R), 1.3 mm.
Figure 5.6. Water-jacketed sample holder, sample cup containing molten cheese, and tri-probe. As the beak was raised, cheese fibers attach to the probe and stretch as the probe is raised vertically to its maximum of 30 cm.

the spindle into an uncut area of the cheese) and locked into position. After 30 s, the cheese was pulled vertically (1.7 cm/s) (Figure 5.6) until all the strands broke or the maximum beam stroke (30 cm) was reached. Four parameters were selected from the resulting force-time data set. Melt strength ($F_M$) was defined as the maximum load (in grams) obtained during the USU stretch test. As the spindle exits the cheese reservoir, strands of cheese are drawn upward and the amount and elasticity of the cheese determines the load exerted against the spindle as it moves upward. The load (in grams), at 5 cm and 20 cm of extensibility, was used as an indicator of stretch quality (SQ) and designated as $SQ_5$ and $SQ_{20}$, respectively. The final parameter was called stretch length
(SL). This is the distance the cheese strands remained attached to the spindle or the distance at which the load on the spindle was \( \leq \) to 5 g. The entire force-time data set was referred to as the stretch profile of the cheese.

**Statistical Analysis**

Two nonfat and four low-moisture part-skim Mozzarella cheeses were chosen at random from local supermarkets. An additional nonfat Mozzarella cheese was manufactured at Utah State University. The data was analyzed as a split plot design with cheese as the whole plot factor and either viscosity, stretch strength, stretch quality, or stretch length as the response variable. The statistical analysis was done in PROC Mixed in SAS using the (default) restricted maximum likelihood estimation method (SAS, 1999). Significance was declared at \( P \leq 0.05 \).

**RESULTS**

**Cheese Composition**

The fat and moisture content of the NF cheeses were within expected ranges, i.e., \(< 0.1\% \) fat and 56 to 61\% moisture (Table 5.1). This reduction in fat content and increase in moisture content, when compared to LMPS cheese, was accompanied by an increase in the protein content of the NF cheeses. The calcium, sodium and potassium contents of NF-1 and NF-2 were slightly higher than the mineral contents of the LMPS cheeses (Table 5.2). However, cheese NF-3, a cheese made using a direct acid method, contained less calcium than did the other cheeses. This reduction in calcium content is
Table 5.1. Mean (±SD) fat, moisture, and moisture-in-nonfat-substance (MNFS) in nonfat (NF) and low-moisture part-skim (LMPS) Mozzarella cheese (n = 2). Protein and calcium-to-protein ratio of NF and LMPS Mozzarella cheese (n = 1).

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<tr>
<th>Cheese</th>
<th>Fat</th>
<th>SD</th>
<th>Moisture</th>
<th>SD</th>
<th>Protein</th>
<th>MNFS</th>
<th>C:P</th>
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<td>58.9</td>
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</tr>
<tr>
<td>NF-2</td>
<td>&gt; 0.1</td>
<td>0.0</td>
<td>56.3</td>
<td>0.1</td>
<td>26.9</td>
<td>56.3</td>
<td>0.030</td>
</tr>
<tr>
<td>NF-3</td>
<td>&gt; 0.1</td>
<td>0.0</td>
<td>60.3</td>
<td>0.1</td>
<td>31.9</td>
<td>60.3</td>
<td>0.016</td>
</tr>
<tr>
<td>LMPS-1</td>
<td>20.0</td>
<td>0.0</td>
<td>45.6</td>
<td>0.0</td>
<td>25.3</td>
<td>57.0</td>
<td>0.030</td>
</tr>
<tr>
<td>LMPS-2</td>
<td>20.5</td>
<td>0.4</td>
<td>49.9</td>
<td>0.8</td>
<td>23.0</td>
<td>62.8</td>
<td>0.031</td>
</tr>
<tr>
<td>LMPS-3</td>
<td>19.5</td>
<td>0.4</td>
<td>47.3</td>
<td>0.1</td>
<td>24.5</td>
<td>59.0</td>
<td>0.024</td>
</tr>
<tr>
<td>LMPS-4</td>
<td>20.5</td>
<td>0.0</td>
<td>48.1</td>
<td>0.1</td>
<td>23.2</td>
<td>60.5</td>
<td>0.027</td>
</tr>
</tbody>
</table>

Table 5.2. Mineral content of selected minerals in nonfat (NF) and low-moisture part-skim (LMPS) Mozzarella cheese (n = 1).

<table>
<thead>
<tr>
<th>Cheese</th>
<th>Ca</th>
<th>Na</th>
<th>P</th>
<th>K</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NF-1</td>
<td>0.86</td>
<td>0.62</td>
<td>0.59</td>
<td>0.09</td>
</tr>
<tr>
<td>NF-2</td>
<td>0.78</td>
<td>0.69</td>
<td>0.54</td>
<td>0.07</td>
</tr>
<tr>
<td>NF-3</td>
<td>0.52</td>
<td>0.55</td>
<td>0.41</td>
<td>0.08</td>
</tr>
<tr>
<td>LMPS-1</td>
<td>0.76</td>
<td>0.48</td>
<td>0.52</td>
<td>0.05</td>
</tr>
<tr>
<td>LMPS-2</td>
<td>0.72</td>
<td>0.58</td>
<td>0.47</td>
<td>0.04</td>
</tr>
<tr>
<td>LMPS-3</td>
<td>0.59</td>
<td>0.36</td>
<td>0.41</td>
<td>0.07</td>
</tr>
<tr>
<td>LMPS-4</td>
<td>0.62</td>
<td>0.51</td>
<td>0.44</td>
<td>0.07</td>
</tr>
</tbody>
</table>
typical of cheeses made using direct acidification (Paulson et al, 1998) and resulted in a lower C:P ratio than the other cheeses. The fat and moisture content of the LMPS cheeses were within expected ranges, i.e., 30 to 45% fat dry weight, and 45 to 52% moisture, respectively. The moisture-in-nonfat-substance of all cheeses ranged from 56 to 63%.

**Melt Test**

After submerging our sample tubes into hot oil, the temperature of the cheese samples increased from room temperature to a temperature nearing 95°C over a period of 16 min. During the first 4 min, the cheese softened, and began to flow in the glass tube. The outside of the cheese plug, being closer to the heat source, melted first and accounted for the majority of the cheese flow during the early stages of the test. By 8 min, the entire cheese plug reached a temperature that allowed the entire cheese mass to flow. Between 8 and 12 min, the cheese continued to flow evenly. However, between 14 and 16 min, bubbles began to appear on the surface of many cheese samples. Once the bubbles appeared, the flow rate of the molten cheese decreased dramatically. The appearance of the bubbles was probably caused as the water in the cheese reached the boiling point at our testing elevation (1470 m above sea level) and is consistent with observations of McMahon et al. (1999).

The test cheeses had a wide range of meltability, with 12-min melt distances ranging from 60 to 170 mm (Figure 5.7). There was a significant difference ($P < 0.05$) between the meltability of the various cheeses based on fat level. There was also a
Figure 5.7. Mean (±SD) melt distance of nonfat (NF) and low-moisture part-skim (LMPS) Mozzarella cheese at 12 min (95°C).

There was a wide range in AV of the cheeses and there was also a significant difference ($P < 0.05$) in the AV among the various cheeses (Figure 5.8). In general, the AV of all cheeses decreased with increasing test temperatures. However, the AV of one
Figure 5.8. Apparent viscosity of nonfat (NF) and low-moisture part-skim (LMPS) Mozzarella cheese at 60, 70, 80, and 90°C. NF-1, open diamond, NF-2, open circle, NF-3, open triangle, LMPS-1, solid diamond, LMPS-2, solid square, LMPS-3, solid circle, and LMPS-4, open square.

NF cheese, NF-2, exhibited anomalous behavior and AV increased with increasing test temperatures.

**Fork Test**

There was a significant difference ($P < 0.05$) between the stretchability of the various cheeses based on fat level (Figure 5.9). In general, cheeses with lower fat content stretched to a greater degree than cheeses with higher fat content. However, one LMPS cheese, cheese LMPS-4, stretched to the same degree as did the NF cheeses.
Figure 5.9. Mean (±SD) stretch distance, as determined by a fork stretch test, of nonfat (NF) and low-moisture part-skim (LMPS) Mozzarella cheese.

**USU Stretch Test**

*Stretch profile.* The stretch profiles of three selected cheeses, NF-2, LMPS-4, and LMPS-1, at 70°C are shown in Figure 5.10. The stretch profiles were divided into two regions (R1 and R2) based on the relative position of the probe to the melted cheese pool. The initial part of the stretch profile, (R1), covered the portion of the stretch profile from crosshead actuation until the probe lifted from the melted cheese pool. This occurred by 3 cm of extension, i.e., approximately 2 s after crosshead actuation. As the
Figure 5.10. Stretch profiles of nonfat (NF) and low-moisture part-skim Mozzarella cheese at 70°C; cheese NF-2 (A), cheese LMPS-4 (B), and cheese LMPS-1 (C). Region 1 (R1), from cross-head actuation until probe exits the melted cheese pool, region 2 (R2) from melted cheese pool surface until peak stroke maximum (30 cm) is reached.
probe is pulled through the melted cheese, a resistant load, based on cheese mass, viscosity, and elasticity, is exacted on the probe. This load increases rapidly from the time of crosshead actuation until the probe travels through approximately one half of the melted cheese pool. This load then decreases as the probe passes through and exits the upper half of the melted cheese pool.

Region 2 (R2) covered the portion of the stretch profile after the probe left the melted cheese pool surface. As the probe leaves the cheese surface, the molten cheese either begins to stretch or falls back into the melted cheese pool. As the probe is raised further, the cheese attached to the probe continues to stretch, becoming more elongated. Cheese not retained on the probe falls back into the melted cheese pool. As these events occur, the load decreases, as indicated by a rapid reduction in load cell value (Figure 5.10). By approximately 5 cm of extensibility, most of the excess cheese originally drawn up by the probe, falls back into the melted cheese pool, leaving only a small portion of the original cheese mass attached directly to the probe. Cheese attached to the probe begins to cool and stiffen slightly as the probe is raised further into the air. The elongating cheese column directly beneath the probe also begins to cool and stiffen slightly as the probe continues to rise from the cheese pool. Since no excess cheese remains on the probe to feed the lengthening cheese strand, additional cheese is drawn into the cheese column from the melted cheese pool as the probe is pulled upward. Cheese elongation and simultaneous introduction of new cheese into the cheese column occur at a given rate depending on the characteristics of the cheese being tested. Thus,
the load exerted on the load cell remains relatively constant throughout R2 of the stretch profile unless the cheese strands break. In some instances, attached cheese strands thinned to hair-like fibers that extended to the maximum distance (30 cm). The load cell was not sensitive enough to detect their presence.

**Stretch parameters.** Maximum load recorded within R1 was defined as melt strength ($F_M$) and occurred while the probe was being raised through the melted cheese pool (Figure 5.11). Two load measurements within R2 were used to characterize stretch quality (SQ). Measurements of SQ at 5 and 20 cm of extension (designated as SQ$_5$ and SQ$_{20}$ respectively) gave an indication of the extent that cheese formed into an initial strand (or strands) that could be pulled from the reservoir of molten cheese. The final parameter that was used to characterize the cheese was the stretch length (SL). This was defined as the maximum distance the probe traveled before all the cheese strands broke. Examples of the stretch profiles of individual cheeses are given in Figure 5.10 and Figure 5.11. In Figure 5.10, the variation in the stretch profiles between different types of cheese stretched at the same temperature (70°C) can be observed. For this example, the melt strength of cheese NF-2 (Figure 5.10A) was twice that of cheese LMPS-2 (Figure 5.10C). Cheese LMPS-4 (Figure 5.10B) had an intermediate melt strength value. When comparing stretch quality values of these same cheeses, cheese LMPS-4 had higher SQ$_5$ and SQ$_{20}$ values than did either cheese NF-2 or LMPS-2. From these observations, cheeses that have a higher melt strength value do not necessarily have a corresponding high stretch quality value. In this example (Figure 5.10), all cheeses stretched to the
Figure 5.11. Stretch profiles of low-moisture part-skim Mozzarella cheese (cheese LMPS-1) at 60, 70, 80, and 90°C. Melt strength (maximum load (g) obtained during the tensile stretch test), stretch quality (average load (g) obtained at 5 cm (SQ5) and 20 cm (SQ20)), and stretch length (SL) (maximum distance (cm) the probe traveled until all the strands broke or the beak stroke maximum (30 cm) was reached).
maximum extension of 30 cm at 70°C however, there were cheeses in which the strands broke before 30 cm of extension was reached (see Tables 5.3, 5.4, 5.5, and 5.6).

When individual cheeses were tested at different temperatures (60, 70, 80, and 90°C), different stretch profiles were obtained at each temperature. An example of the stretch profiles of cheese LMPS-1 tested at 60, 70, 80, and 90°C is given in Figure 5.11. In this example, \( F_M \), \( SQ_5 \), and \( SQ_{20} \) decreased as cheese LMPS-1 was tested at increasingly greater temperatures. Although this represented the general trend for both the NF and LMPS cheeses (see Tables 5.3, 5.4, 5.5, and 5.6), the \( F_M \) of one cheese, cheese NF-2, increased with increasing test temperature in the same manner that AV increased.

Linear correlation between the melted cheese parameters is shown in Table 5.7. There was a reasonably strong relationship between AV and \( F_M \), \( R^2 = 0.78 \), and \( SQ_5 \) and \( SQ_{20} \), \( R^2 = 0.80 \). However, correlation between other test parameters was less than \( R^2 = 0.45 \).

**DISCUSSION**

**Composition**

With the exception of cheese NF-3, which was produced using a direct acid method, the NF and LMPS cheeses used to evaluate the USU stretch test were chosen at random from commercial sources. The fat and moisture contents of all cheeses were within expected ranges (Code of Federal Regulations, 2000), as were the mineral
Table 5.3. Mean (±SD) melt strength ($F_M$), stretch quality at 5 cm ($SQ_5$), stretch quality at 20 cm ($SQ_{20}$) of extension, and stretch length (SL) of cheese at 60°C ($n = 2$).

<table>
<thead>
<tr>
<th>Cheese</th>
<th>$F_M$</th>
<th>SD</th>
<th>$SQ_5$</th>
<th>SD</th>
<th>$SQ_{20}$</th>
<th>SD</th>
<th>SL</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>NF-1</td>
<td>243</td>
<td>41</td>
<td>105</td>
<td>13</td>
<td>33</td>
<td>1</td>
<td>30.6</td>
<td>0.5</td>
</tr>
<tr>
<td>NF-2</td>
<td>182</td>
<td>60</td>
<td>13</td>
<td>8</td>
<td>6</td>
<td>1</td>
<td>16.7</td>
<td>15.4</td>
</tr>
<tr>
<td>NF-3</td>
<td>136</td>
<td>16</td>
<td>44</td>
<td>8</td>
<td>12</td>
<td>1</td>
<td>29.9</td>
<td>0.0</td>
</tr>
<tr>
<td>LMPS-1</td>
<td>254</td>
<td>35</td>
<td>54</td>
<td>5</td>
<td>16</td>
<td>4</td>
<td>31.0</td>
<td>0.7</td>
</tr>
<tr>
<td>LMPS-2</td>
<td>117</td>
<td>2</td>
<td>9</td>
<td>1</td>
<td>9</td>
<td>0</td>
<td>18.6</td>
<td>16.6</td>
</tr>
<tr>
<td>LMPS-3</td>
<td>218</td>
<td>18</td>
<td>55</td>
<td>1</td>
<td>21</td>
<td>1</td>
<td>30.6</td>
<td>0.0</td>
</tr>
<tr>
<td>LMPS-4</td>
<td>259</td>
<td>21</td>
<td>110</td>
<td>18</td>
<td>29</td>
<td>4</td>
<td>30.0</td>
<td>0.4</td>
</tr>
</tbody>
</table>

Table 5.4. Mean (±SD) melt strength ($F_M$), stretch quality at 5 cm ($SQ_5$), stretch quality at 20 cm ($SQ_{20}$) of extension, and stretch length (SL) of cheese at 70°C ($n = 2$).

<table>
<thead>
<tr>
<th>Cheese</th>
<th>$F_M$</th>
<th>SD</th>
<th>$SQ_5$</th>
<th>SD</th>
<th>$SQ_{20}$</th>
<th>SD</th>
<th>SL</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>NF-1</td>
<td>145</td>
<td>10</td>
<td>32</td>
<td>4</td>
<td>13</td>
<td>2</td>
<td>30.2</td>
<td>0.0</td>
</tr>
<tr>
<td>NF-2</td>
<td>178</td>
<td>28</td>
<td>7</td>
<td>3</td>
<td>4</td>
<td>0</td>
<td>9.0</td>
<td>1.5</td>
</tr>
<tr>
<td>NF-3</td>
<td>92</td>
<td>4</td>
<td>17</td>
<td>0</td>
<td>8</td>
<td>0</td>
<td>29.9</td>
<td>0.4</td>
</tr>
<tr>
<td>LMPS-1</td>
<td>87</td>
<td>27</td>
<td>13</td>
<td>6</td>
<td>8</td>
<td>1</td>
<td>29.3</td>
<td>1.0</td>
</tr>
<tr>
<td>LMPS-2</td>
<td>84</td>
<td>19</td>
<td>28</td>
<td>17</td>
<td>13</td>
<td>8</td>
<td>21.3</td>
<td>13.8</td>
</tr>
<tr>
<td>LMPS-3</td>
<td>98</td>
<td>3</td>
<td>27</td>
<td>2</td>
<td>13</td>
<td>2</td>
<td>30.1</td>
<td>1.1</td>
</tr>
<tr>
<td>LMPS-4</td>
<td>105</td>
<td>6</td>
<td>26</td>
<td>4</td>
<td>15</td>
<td>3</td>
<td>29.5</td>
<td>0.1</td>
</tr>
</tbody>
</table>
Table 5.5. Mean (±SD) melt strength (F_M), stretch quality at 5 cm (SQ_5), stretch quality at 20 cm (SQ_20) of extension, and stretch length (SL) of cheese at 80°C (n = 2).

<table>
<thead>
<tr>
<th>Cheese</th>
<th>F_M</th>
<th>SD</th>
<th>SQ_5</th>
<th>SD</th>
<th>SQ_20</th>
<th>SD</th>
<th>SL</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>NF-1</td>
<td>92</td>
<td>3</td>
<td>48</td>
<td>8</td>
<td>14</td>
<td>2</td>
<td>30.2</td>
<td>0.1</td>
</tr>
<tr>
<td>NF-2</td>
<td>279</td>
<td>33</td>
<td>11</td>
<td>9</td>
<td>12</td>
<td>8</td>
<td>17.8</td>
<td>18.1</td>
</tr>
<tr>
<td>NF-3</td>
<td>53</td>
<td>1</td>
<td>11</td>
<td>0</td>
<td>6</td>
<td>1</td>
<td>7.2</td>
<td>0.7</td>
</tr>
<tr>
<td>LMPS-1</td>
<td>50</td>
<td>11</td>
<td>9</td>
<td>2</td>
<td>7</td>
<td>1</td>
<td>16.5</td>
<td>15.5</td>
</tr>
<tr>
<td>LMPS-2</td>
<td>40</td>
<td>5</td>
<td>5</td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>4.9</td>
<td>0.0</td>
</tr>
<tr>
<td>LMPS-3</td>
<td>158</td>
<td>6</td>
<td>38</td>
<td>1</td>
<td>17</td>
<td>5</td>
<td>16.7</td>
<td>8.5</td>
</tr>
<tr>
<td>LMPS-4</td>
<td>53</td>
<td>3</td>
<td>1</td>
<td>4</td>
<td>10</td>
<td>2</td>
<td>30.0</td>
<td>0.0</td>
</tr>
</tbody>
</table>

Table 5.6. Mean (±SD) melt strength (F_M), stretch quality at 5 cm (SQ_5), stretch quality at 20 cm (SQ_20) of extension, and stretch length (SL) of cheese at 90°C (n = 2).

<table>
<thead>
<tr>
<th>Cheese</th>
<th>F_M</th>
<th>SD</th>
<th>SQ_5</th>
<th>SD</th>
<th>SQ_20</th>
<th>SD</th>
<th>SL</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>NF-1</td>
<td>93</td>
<td>6</td>
<td>34</td>
<td>4</td>
<td>26</td>
<td>8</td>
<td>29.2</td>
<td>1.8</td>
</tr>
<tr>
<td>NF-2</td>
<td>263</td>
<td>1</td>
<td>5</td>
<td>2</td>
<td>6</td>
<td>3</td>
<td>16.8</td>
<td>17.3</td>
</tr>
<tr>
<td>NF-3</td>
<td>51</td>
<td>8</td>
<td>15</td>
<td>4</td>
<td>8</td>
<td>1</td>
<td>8.1</td>
<td>2.5</td>
</tr>
<tr>
<td>LMPS-1</td>
<td>47</td>
<td>18</td>
<td>3</td>
<td>4</td>
<td>3</td>
<td>1</td>
<td>3.5</td>
<td>0.8</td>
</tr>
<tr>
<td>LMPS-2</td>
<td>41</td>
<td>3</td>
<td>6</td>
<td>6</td>
<td>3</td>
<td>1</td>
<td>4.9</td>
<td>1.2</td>
</tr>
<tr>
<td>LMPS-3</td>
<td>73</td>
<td>7</td>
<td>17</td>
<td>20</td>
<td>3</td>
<td>0</td>
<td>5.1</td>
<td>1.3</td>
</tr>
<tr>
<td>LMPS-4</td>
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<td>3</td>
<td>18</td>
<td>4</td>
<td>15</td>
<td>0</td>
<td>29.7</td>
<td>0.3</td>
</tr>
</tbody>
</table>
Table 5.7. Correlation coefficients ($R^2$) for apparent viscosity (AV), melt strength ($F_M$), stretch quality at 5cm ($SQ_5$) and stretch quality at 20cm ($SQ_{20}$) of extension, stretch length as determined by USU stretch test ($SL_T$), stretch length as determined by fork test ($SL_F$), and melt distance at 12 min ($M_{12}$).

<table>
<thead>
<tr>
<th></th>
<th>AV (%)</th>
<th>$F_M$ (g)</th>
<th>$SQ_5$ (g)</th>
<th>$SQ_{20}$ (g)</th>
<th>$SL_T$ (cm)</th>
<th>$SL_F$ (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AV (%)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>$F_M$ (g)</td>
<td>0.777</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>$SQ_5$ (g)</td>
<td>0.427</td>
<td>0.334</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>$SQ_{20}$ (g)</td>
<td>0.302</td>
<td>0.241</td>
<td>0.804</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>$SL_T$ (cm)</td>
<td>0.293</td>
<td>0.120</td>
<td>0.282</td>
<td>0.391</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>$SL_F$ (cm)</td>
<td>0.015</td>
<td>0.038</td>
<td>0.218</td>
<td>0.270</td>
<td>0.005</td>
<td>-</td>
</tr>
<tr>
<td>$M_{12}$ (mm)</td>
<td>0.110</td>
<td>0.183</td>
<td>0.093</td>
<td>0.088</td>
<td>0.053</td>
<td>0.058</td>
</tr>
</tbody>
</table>

\(^1\)Compared with other parameters at 80°C.

The contents of the commercially prepared cheeses (U.S. Department of Agriculture, 2001). However, the calcium content of cheese NF-3 was approximately 28% lower than that of the other NF cheeses or of the LMPS cheeses. We attributed this lower calcium content to the dissociation of calcium from colloidal calcium phosphate prior to renneting (Fox and McSweeney, 1998; Guinee et al., 1993). This dissociation of calcium from calcium phosphate was probably the result of a milk pre-acidification step in the manufacturing method used to make cheese NF-3. As the milk pH is lowered from pH 6.7 to 5.4 ionic calcium dissociates from colloidal calcium phosphate, increasing the concentration of ionic calcium in the milk. Upon coagulation, this ionic calcium is lost in the whey rather than being retained in the curd. Thus, the concentration of calcium in cheese made using a milk pre-acidification step is generally lower than in cheese made using a method in which a pre-acidification step is lacking (Paulson et al., 1998).
Melt Distance

The melt test measured the degree to which cheese changed from a semi-solid state to a liquid state when exposed to heat. As this change occurs the rigid structure associated with shredded or solid cheese is replaced by a soft, continuous, flowing mass. The extent to which this cheese mass flows may be measured and used as an indication of cheese meltability (Arnott et al., 1957; Kindstedt, 1993; Kosikowski and Mistry, 1997; Olson and Price, 1958).

The extent to which cheese melts is determined by the integrity of the casein matrix. During gel formation, casein micelles aggregate to form protein strands. Initially, these strands are relatively short but eventually fuse together into an overlapping yet porous network of protein that is filled with large amounts of serum (Guinee et al., 1993). As serum is expelled during syneresis, casein strands continue to aggregate until they form a dense protein matrix containing small amounts of serum and fat. The extent to which this protein matrix responds to heat-related phase change is determined by the number of strands per unit area, matrix homogeneity, and the number and type of bonds between the basic aggregates within a strand (Guinee et al., 1993). During the aging process, the proteolytic action of residual cheese clotting enzymes, plasmin, and bacterial proteases, hydrolyze casein, forming short chain proteins and peptides. As the number of short chain proteins and peptides increases, bond rearrangements occur that allow proteins to flow past one another more easily when heated (Yun et al., 1993). Therefore, as proteolysis continues melt distance increases. With the exception of
cheese NF-3, a non-commercial cheese for which the manufacturing date was known, the manufacturing dates of the commercial cheeses were unknown, therefore, comparison between cheese meltability based on proteolysis was meaningless. Since our objective was to determine whether the USU stretch test could differentiate between the stretchability of cheeses, regardless of cause, differences in melted cheese characteristics based on proteolysis were not determined.

In addition to casein hydrolysis, the presence of fat, in the case of LMPS cheese, and increased water content, i.e., unbound water, in the case of NF cheese, also affect cheese meltability. Fat and unbound water acts as a lubricant and increases the ability of cheese particles to flow (Tunick et al., 1993; Tunick and Shieh, 1995). With the exception of cheese LMPS-1, the NF and LMPS cheeses melted to approximately the same degree (see Figure 5.7). This was not surprising since NF Mozzarella cheese is generally manufactured in such a manner as to mimic the functional properties (melt and stretch) of LMPS Mozzarella cheese. Cheese LMPS-1 melted to a greater degree than did the other LMPS cheeses although the fat content of cheese LMPS-1 was similar to the fat content of the other LMPS cheeses. However, the moisture content of cheese LMPS-1 was approximately 3.0% lower than that of the other LMPS cheeses. This lower moisture content resulted in a lower moisture-in-nonfat-substance and suggested a decrease in the amount of unbound water available as a lubricating constituent of cheese LMPS-1. However, since cheese LMPS-1 melted to a greater extent than did any other cheese, the lubricating effect of fat and water may not have been the reason for its
increased meltability. Rather, cheese LMPS-1 had probably undergone more proteolysis than the other LMPS cheeses although the actual cause for this difference remains unknown because the extent of proteolysis was not measured.

**Helical Viscometry**

Unlike the melt test, which measured the degree to which the protein matrix undergoes heat-related phase change, apparent viscosity measured the resistant force on the T-bar as it traveled through the molten cheese. As a rotating T-bar spindle is pulled upward through molten cheese, resistance on a torque spring increases. This resistance is caused by the cheese modulus and any inclination for a cheese mass to form around the spindle (Kindstedt et al., 1989a; 1989b). Since helical viscometry measures the combined effect of these melted cheese properties, stretchability as determined by other methods may not be directly related to apparent viscosity. Nevertheless, low apparent viscosity values have been interpreted as being synonymous with poor stretchability or poor elasticity (Oberg et al., 1992).

Apparent viscosity is generally determined at 60°C since this is the approximate temperature at which melted cheese is consumed on a pizza (Kindstedt et al., 1989b). When relative apparent viscosity of the NF and LMPS cheeses were compared at 60°C, no clear distinction between the cheeses based on fat content was apparent (see Figure 5.8). For example, cheese NF-2, had low AV at 60°C, while, cheese NF-1 stretched to the maximum extent of the viscometer torque spring at 60°C, i.e., had high AV.
We were interested in comparing the apparent viscosity of the test cheeses to the USU stretch test at temperatures greater than 60°C. Therefore, apparent viscosity values were determined for the cheeses at temperatures from 60°C to a maximum test temperature of 90°C. As the temperature of the test was increased, the apparent viscosity of the molten cheeses generally decreased. However, the apparent viscosity of cheese NF-2 increased between test temperatures 70°C and 80°C. This observation was unexpected. In retrospect, cheese NF-2 was a pre-shredded Mozzarella cheese that contained inulin, which has been shown to form gels at 80°C.

**Fork Test**

When the fork test was used to evaluate the stretchability of the NF and LMPS Mozzarella cheeses, a general trend among the cheeses was apparent. NF cheeses stretched to a greater degree than did LMPS cheeses, with the exception of cheese LMPS-4 (see Figure 5.9). These results were not consistent with those obtained by helical viscometry at 60°C (see Figure 5.8). When stretch at 60°C was compared using apparent viscosity, cheese NF-1 and cheese NF-2 stretched to the greatest and least degree respectively of all the cheeses. However, both cheeses stretched to approximately the same degree when compared using the fork test. Additionally, cheese LMPS-1, cheese LMPS-2, and cheese LMPS-3, stretched to a lesser degree than did the NF cheeses when compared using a fork test, while stretching to intermediate degrees between cheese NF-1 and NF-2, when comparing stretch by helical viscometry. These observations suggest that the fork test and helical viscometry measure different melted.
cheese properties. Since the fork test only measures maximum cheese extensibility, it provides no information on the other attributes of the cheese. For example, the NF cheeses tended to form thin yet strong strands while the LMPS cheeses tended to form strands that were much thicker yet stretched to a lesser degree.

**USU Stretch Test**

*Probe configuration.* Little information exists on the type of probe configuration best suited for stretching cheese. A tined fork of unspecified dimension is used to stretch cheese when using the fork test method (U. S. Department of Agriculture, 1980). Kindstedt et al. (1989a; 1989b) and Oberg et al. (1991) used a T-bar spindle to determine the apparent viscosity of melted cheese by helical viscometry. More recently, Peña et al. (1996) evaluated three types of spindle configuration, fork, solid beam, and rectangular wire frame, for measuring the stretchability of melted cheese using a tensile test method. They obtained the greatest correlation between textural rankings and sensory results when a rectangular wire frame configuration was used. Unfortunately, the dimensions and frame construction of the wire spindle did not lend themselves to the USU stretch test. Our experience with helical viscometry suggested that we use a T-bar probe configuration. However, the two horizontal tines of the T-bar failed to lift enough cheese to form a consistent strand. The addition of another crossbar, to form a probe with an “X” shape, also failed in that it lifted too much cheese from the sample well. A tri-probe with three hemispherical hooks provided an intermediate configuration between the T-bar spindle and the X-bar spindle (see Figure 5.4).
Strand cooling. Since the temperature immediately surrounding the elongating cheese strand was not controlled, the upper portion of the extending cheese strand cooled slightly as it was raised from the melted cheese pool. This cooling was noted by a change in color from opaque to partly translucent and by a slight stiffening of the upper portion of the strand. This probably promoted cheese incorporation into the elongating strand from the melted cheese pool. Guinee and O’Callaghan (1997) confirmed earlier observations of Apostolopoulos (1994) that cheese temperature decreased by 3°C to 4°C in cheese samples stretched by means of a pizza-template method. They concluded that this temperature variation did not effect stretch length provided the test duration was relatively short (typically < 30 s). It should be pointed out that the cheese mass used in the USU stretch test was small when compared to the tests of Apostolopoulos (1994) or Guinee and O’Callaghan (1997). Therefore, the cheese attached to the hook and the upper portion of the elongating cheese strand probably cooled more than previously observed even though the duration of the test was quite short, approximately 20 s.

Melt strength. While the tri-probe remained in the melted cheese pool, the resistant force exerted on the tri-probe was caused by many of the same force components as those exerted on the T-bar spindle used to determine apparent viscosity. Those forces included the upward pull of the spindle, cheese viscosity, cheese fiber formation around the spindle, and the capacity of the cheese fibers to resist deformation. However, the rotational forces associated with helical viscometry were absent in the
USU stretch test. This similarity between the force components was reflected in a relatively high correlation coefficient ($R^2 = 0.77$) between AV and melt strength (see Table 5.7). Typically, the $F_m$ exerted on the tri-probe occurred when the probe had been pulled through approximately half the melted cheese pool. Load values dropped off quickly as the probe was pulled through the upper half of the melted cheese pool with full egression from the cheese surface at approximately 3 s. This short duration in the melted cheese pool resulted in a noticeable peak (see Figure 5.10) rather than a plateau as seen in helical viscometry stretch profiles (Fife et al., 1996).

**Stretch quality.** In preliminary work, all cheeses stretched to a minimum of 5 cm of extension while most cheeses stretched more than 20 cm. Therefore, the average load exerted on the probe at 5 cm and 20 cm of extensibility was chosen as the minimum and maximum SQ values, respectfully. The average load exerted on the probe at these two points of extensibility could be used as a measure of SQ. However, for cheeses that did not extend to 20 cm of extensibility or where the strands of one or two duplicate tests broke prematurely, SQ values were subject to averaging error. Therefore, the load values at 5 cm of extensibility and at 20 cm of extensibility were used to give an indication of cheese stretchability. As seen in Tables 5.3, 5.4, 5.5, and 5.6, either $SQ_5$ or $SQ_{20}$ may be used to estimate the stretchability of cheese.

**Stretch length.** Maximum cheese extensibility has been the primary method for evaluating cheese stretchability. However, no methodology has been standardized nor has there been agreement on interpretation. The fork test used in this study, the vertical
extension method of Apostolopoulos (1994), and the horizontal extension method of Guinee and O’Callaghan (1997) use maximum cheese extensibility as an indicator of cheese stretchability. However, our experience with helical viscometry, the fork test as described here, and with the USU stretch test, point out the difficulties when using maximum extensibility as the sole determinant of cheese stretchability. Although strand length is not a component of helical viscometry analysis, strands of molten cheese are drawn upward from the melted cheese pool as the helipath raises the T-spindle. It is not uncommon for a single hair-sized strand of cheese to extend the entire travel-distance of the helipath (approximately 22 cm). Nor is it unusual to observe extremely thin strands of cheese extending from the melted cheese pool to the tri-probe during the USU stretch test (30 cm). Although these strands extended from the probe to the cheese pool, none are large enough to be detected by the instrumentation. When using the fork test to evaluate cheese stretchability, it was also common for wispy strands of cheese to remain attached to the fork tines long after the majority of the strands had broken.

Apostolopoulos (1994) and Guinee and O’Callaghan (1997) also used the break-point method for determining cheese stretchability although neither mentioned how the endpoint was determined. In each method, determining the point at which all the strands break is difficult. Therefore, other parameters such as $F_M$ and $SQ_5$ and $SQ_{20}$ become useful in determining melted cheese properties in general and stretchability properties in particular.
CONCLUSIONS

By utilizing information from stretch profiles generated by the Utah State University stretch test, additional information on melted cheese characteristics of Mozzarella cheese may be obtained. When used in conjunction with descriptive parameters such as apparent viscosity and strand length, melt strength, stretch quality at 5 cm, and stretch quality at 20 cm may be used to more fully describe melted cheese characteristics.

REFERENCES


As the fat content of low-moisture part-skim Mozzarella cheese is reduced from 19.8% to less than 5%, cheese meltability is affected. Cheeses that contain between 2.2% and 5% fat melt to a lesser degree than does low-moisture part-skim Mozzarella cheese containing 19.8% fat. As fat content is reduced, a concurrent increase in the moisture content from 52.1% moisture in low-moisture part-skim Mozzarella cheese to approximately 63.0% moisture in reduced-fat cheese is observed. Although the level of moisture can be increased in lower fat cheese, moisture in the fat-free cheese may actually decrease. The moisture in the fat-free cheese may be increased in lower-fat cheeses by altering the cheese making procedure. Since altering the fat content or moisture content of cheese dramatically effects functional characteristics such as melt and stretch, an understanding of the affect of fat reduction and the role of moisture, as it relates to cheese meltability, is important.

Increasing the water content of these lower fat cheeses improved their meltability. However none of the lower fat cheeses melted as well as the low-moisture part-skim control although the moisture content of these cheeses was higher. This suggests that although the moisture content in the lower fat cheeses may be higher, this moisture may not be available to act as a lubricant as the cheese is heated and begins to melt.
The amount of bound, entrapped, and expressible moisture was determined in reduced-fat and low-moisture part-skim Mozzarella cheese. During the first 3 wk of storage, the amount of bound water/g of protein (0.71 g) remained the same. By day 21, expressible water contained within the fat-serum channels of Mozzarella cheese was absorbed into the cheese protein matrix where it became entrapped water. Presumably, this change was caused by a rearrangement of the protein molecules contained within the protein matrix that was induced by salting and refrigeration of the cheese. The water absorption was accompanied by a swelling of the protein matrix, which continued until the spaces between the fat globules were completely filled with matrix proteins. The increase in entrapped water during storage was limited in the reduced-fat cheese because of the lower volume of expressible water initially present in the fat-serum channels. Improvements in the meltability of the cheese occurred as the matrix proteins became more hydrated. This implies that a reduction in the extent of protein-to-protein interaction within the protein matrix is a principal reason for increased meltability of Mozzarella cheese during storage.

There are many methods used to measure cheese meltability that provide objective data. However, the closely related melted cheese characteristic of stretch is either measured by cheese extensibility or by visual observation. A multi-parameter tensile test was developed to measure cheese stretchability. This USU Stretch test utilized information from multiple areas of the stretch profile of melted cheese. During a test, strands of cheese are drawn upward and the amount and elasticity of the cheese
determines the load exerted against the three-pronged spindle. Three new melted cheese stretch parameters were defined from the resulting force-time data set. Melt strength ($F_M$) was defined as the maximum value obtained as the spindle transits the cheese reservoir. The load at 5 cm and 20 cm of extensibility was defined as stretch quality and designated as $SQ_5$ and $SQ_{20}$, respectively. A fourth stretch parameter stretch length ($SL$), the distance the cheese strands remained attached to the spindle (or the distance at which the load on the spindle was $\leq$ to 5 g) was described (this parameter was not new). The entire force-time data set was referred to as the stretch profile of the cheese. Using the new terms, $F_M$, $SQ_5$, and $SQ_{20}$, along with the more traditional term $SL$, provides a way to more objectively test and describe the performance of melted cheese.
VITA

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