Effect of Milk Preacidification on Low Fat Mozzarella Cheese: III. Post-Melt Chewiness and Whiteness

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ABSTRACT

The effect of calcium reduction (as a result of milk preacidification) on post-melt chewiness and whiteness of low fat Mozzarella cheese was determined. Four vats (230 kg of milk per vat) of cheese were made in 1 d using no preacidification (control), preacidification pH 6.0 and pH 5.8 with acetic acid, and preacidification to pH 5.8 with citric acid. Cheese manufacture was repeated on four different days using a randomized complete block design. The total calcium content and the water-insoluble calcium content of the cheese were lower in the cheeses made from preacidified milks. The amount of water-soluble and water-insoluble calcium changed during refrigerated storage, as did pH. The post-melt chewiness and whiteness of low fat Mozzarella cheese were affected by milk preacidification. The largest level of calcium reduction and modification in post-melt chewiness and whiteness occurred in the pH 5.8 citric treatment. Multiple regression analysis of post-melt chewiness and cheese whiteness at 38°C after heating and cooling indicated that both water-insoluble calcium and proteolysis were strongly associated with changes in the post-melt chewiness and whiteness of low fat Mozzarella cheese. High levels of proteolysis and low levels of water-insoluble calcium were associated with decreased post-melt chewiness and whiteness of low fat Mozzarella cheese.

(Key words: low fat, Mozzarella cheese, preacidification, post-melt chewiness and whiteness)

INTRODUCTION

In milk, calcium can exist as free ionic calcium or caseinate calcium (bound to casein) or it can be complexed with phosphate in the form of calcium phosphate ion clusters or microgranules. Additionally, the complexed calcium phosphate may also be bound to casein (micellar calcium phosphate) or in the serum phase (colloidal calcium phosphate) (Holt, 1992). Micellar calcium phosphate is distributed throughout the casein matrix in milk (Koop et al., 1973, 1979) and is believed to interact with the phosphoserine residues of casein and act as a crosslinking agent within the casein micelle in milk (Aoki et al., 1987). Aoki et al. (1991) also demonstrated that submicelle structure and ordered structure of casein were not required for crosslinking of casein by colloidal calcium phosphate and that a minimum of three phosphate groups were required for crosslinking of casein by calcium phosphate (Aoki et al., 1992). As a result of this crosslinking function, colloidal calcium phosphate and caseinate calcium would play a role in the structural characteristics of the casein matrix in cheese.

The total amount of calcium in cheese has been shown to influence cheese texture (Solorza and Bell, 1995; Yun et al., 1995) and can be controlled with a variety of manufacturing parameters including: preacidification, pH at whey draining, cooking rate, and final cooling temperature (Keller et al., 1974; Lawrence et al., 1984; Lucey and Fox, 1993). Our laboratory has shown that preacidification to pH 5.8 with citric acid resulted in a large reduction (i.e., 40%) in cheese calcium and improvement in the unmelted (i.e., hardness) and melted (i.e., apparent viscosity) functional properties of low fat Mozzarella cheese (Metzger et al., 2001). In addition to unmelted and melted cheese functional properties, the post-melt properties (after heating and cooling, i.e., pizza bake) are also important and will have an effect.

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on the consumer acceptance of low fat Mozzarella cheese. Low fat Mozzarella has an excessively chewy post-melt texture compared with low-moisture, part skim (LMPS) Mozzarella (Metzger and Barbano, 1999). In addition, substantial differences in cheese whiteness during heating and cooling have been observed when low fat Mozzarella is compared with LMPS Mozzarella (Metzger et al., 2000b). Immediately after baking, low fat Mozzarella has an acceptable appearance on a pizza if a hydrophobic surface coating is added before baking (Metzger et al., 2000b, Rudan and Barbano, 1998). However, during cooling, the cheese can become translucent and its appearance is no longer acceptable (Metzger et al., 2000b). The reversible whiteness changes in low fat Mozzarella cheese during heating and cooling are believed to be caused, in part, by hydrophobic protein-protein interactions in the serum phase of cheese (Metzger et al., 2000b). The reduction in calcium content caused by milk pre-acidification may have an effect on the post-melt chewiness and whiteness of low fat Mozzarella cheese since both post-melt chewiness and whiteness are influenced by protein-protein interactions. However, as described previously, calcium exits in different forms (ionic, micellar, and colloidal or micellar calcium phosphate) and the status of the calcium in cheese may be more important than the total calcium content. The portion of the calcium that is bound to casein (micellar calcium and micellar calcium phosphate) is probably critical, and any factors influencing the portion of calcium bound to protein may impact cheese functionality. Sinha et al. (1979) demonstrated that an equilibrium between water-soluble and water-insoluble calcium exists in cottage cheese. It is hypothesized that the water-insoluble calcium in Mozzarella cheese may be bound to protein or held as an insoluble mineral complex (micellar calcium and micellar calcium phosphate). As a result, the amount of water-insoluble calcium may be more important than total calcium with respect to unmelted cheese hardness and post-melt chewiness. The objective of this study was to determine whether reducing total calcium content (caused by milk preacidification) and the water-soluble and water-insoluble calcium content of the cheese influence the post-melt chewiness and whiteness of low fat Mozzarella cheese.

MATERIALS AND METHODS

Experimental Design

A cheese-making trial was conducted using a 4 × 4 randomized complete block design. Four treatments were employed and included: control (no preacidification), pH 6.0 acetic (preacidification to pH 6.0 with acetic acid), pH 5.8 acetic (preacidification to pH 5.8 with acetic acid), and pH 5.8 citric (preacidification to pH 5.8 with citric acid). Cheese manufacture was repeated on four different days, and the four treatments (control, pH 6.0 acetic, pH 5.8 acetic, and pH 5.8 citric) were made on each day of cheese manufacture. The details of milk standardization, preacidification, and cheese manufacturing were described previously (Metzger et al., 2000a, 2001).

Calcium

Total calcium was determined by atomic adsorption spectroscopy as described previously (Metzger et al., 2000a). In addition to the total calcium content, the water-soluble calcium (WSC) content was determined at 2, 15, 30, 60, and 90 d of refrigerated storage. Water-soluble calcium was determined by blending 5 g of cheese and 50 g of distilled water at 60°C for 30 s with an Omni mixer-homogenizer (model 17105, Omni International, Waterbury, CT). The cheese plus water slurry was then filtered (Whatman #1; Whatman International Ltd., Maidstone, England). The calcium content of the filtrate was determined by atomic absorption spectroscopy, as described previously for milk, except that 1 g of filtrate was used instead of 0.75 g of milk (Metzger et al., 2000a). The grams of calcium extracted with this procedure per 100 g of cheese is referred to as WSC in this study. Water-insoluble calcium was determined by subtracting WSC from the total calcium and WSC as a percentage the total calcium (%WSC) was determined by dividing the WSC (g/100 g of cheese) by the total calcium (g/100 g of cheese), multiplied by 100. Previous research has measured soluble calcium concentration in the water phase of the cheese by extraction of the cheese serum with centrifugation (Guo and Kindstedt, 1995) or pressing (Morris et al. 1988).

Post-Melt Whiteness and Chewiness

The L-value of the cheese during heating and cooling was determined with a Macbeth Color-Eye Spectrophotometer as described previously (Metzger et al., 2000b). The L-value was determined at 7, 24, 38, 49, and 60°C during heating and at 49, 38, 24, and 7°C during subsequent cooling at 30 d of refrigerated storage and at 7, 24, 38, 49, 60, and 71°C during heating and at 60, 49, 38, 24, and 7°C during subsequent cooling at 60 and 90 d of refrigerated storage. The maximum temperature during heating was increased from 60 to 71°C after 30 d of refrigerated storage to ensure that the maximum L-value during heating was attained. At 30 d of storage, the cheese reached a maximum L-value at 60°C, but the temperature required for maximum L-value increased as the cheese got older.
The post-melt chewiness was determined using a previously described objective test (Metzger et al., 1999) at 30, 60, and 90 d of refrigerated storage. In this method, the cheese is melted in a rectangular form, cut into 13- × 13-mm pieces, placed in a stomacher bag with 37°C water (chewed in stomacher for 20 s). The contents of the bag are collected on sieves, and the size distribution of the cheese particles was correlated with chewiness. The results are expressed as a percentage of cheese solids caught on each sieve. A high percentage of cheese solids caught on the sieve with the largest openings corresponds to a cheese that is chewy, and a low percentage of cheese solids caught on the same sieve corresponds to cheese that is not chewy.

Statistical Analysis

Changes in %WSC and water-insoluble calcium were analyzed using a split plot design with treatment (pre-acidification level) as the whole plot factor. For the whole plot factor, treatment was analyzed as a class variable, and the day of cheese manufacture was blocked. For the subplot factor, age, and age × age were analyzed as quantitative variables.

Changes in post-melt chewiness during refrigerated storage were analyzed using two-way ANOVA with treatment and day of manufacture analyzed as class variables and age analyzed as a quantitative variable. Treatment was tested for significance using treatment × day of manufacture as an error term, and age was tested for significance using age × treatment as an error term. Additionally, the post-melt chewiness at each time (30, 60, and 90 d of refrigerated storage) was analyzed using two way ANOVA with treatment and day of manufacture analyzed as class variables. If the F-test for the model was significant (P < 0.05) the treatment means were compared using least significant difference test (P < 0.05).

The L-value of the cheeses at 7°C before heating, at the maximum temperature, and at 7°C after heating and cooling were compared at each time (30, 60, and 90 d of refrigerated storage) using ANOVA. Sources of variation included treatment, day of cheese manufacture, and temperature and were analyzed as class variables. In addition, the interaction of treatment × day of cheese manufacture, temperature × day of cheese manufacture, and treatment × temperature were also included in the model. Treatment was tested for significance using treatment × day of cheese manufacture as the error term, temperature was tested for significance using temperature × day of manufacture as the error term, and treatment × temperature interaction was tested for significance using the mean square error as the error term. The PROC GLM procedure of SAS was used for all analyses of changes in %WSC and water-insoluble calcium during refrigerated storage, post-melt chewiness data, and whiteness data (SAS, 1990).

Multiple regression analysis was performed to determine the correlation of composition, proteolysis, and the state of calcium with the post-melt chewiness of low fat Mozzarella cheese in the study. Initially the data were analyzed using a mixed model (PROC MIXED of SAS) since observations were not independent. However, when the total calcium content of the cheese was placed in the model, the treatment and day of cheese manufacture effect were accounted for and the mixed model was not necessary. This is not surprising since the level of calcium was the treatment effect in this study.

Initially the data were analyzed at each individual time (30, 60, and 90 d of refrigerated storage) using best subsets regression (PROC REG of SAS). Similar results were obtained at 60 and 90 d of refrigerated storage; however, the 30-d data had higher levels of unexplained variation. This probably was a result of the chewiest samples being off scale (i.e., too chewy) in the post-melt chewiness test at 30 d of refrigerated storage (Metzger et al., 1999). The 60 and 90 d data were then pooled, and best subsets regression was again performed on the pooled data. Multiple regression analysis (PROC REG of SAS) was performed on the pooled 60- and 90-d post-melt chewiness data and the pooled 30-, 60-, and 90-d post-melt whiteness data. The order of the terms in the model was manipulated to determine the relative importance of the influence of proteolysis and water-insoluble calcium on post-melt chewiness and whiteness.

RESULTS

Composition

The analyses reported in this paper were done on cheese described in an earlier study (Metzger et al., 2000a). All levels of milk preacidification resulted in a significant reduction in calcium content, and values of 0.976, 0.872, 0.754, and 0.578% of total calcium were obtained, respectively, for the control, pH 6.0 acetic, pH 5.8 acetic, and pH 5.8 citric treatments. There were no differences in moisture, salt, moisture to protein ratio, moisture in nonfat substance or salt to moisture ratio among treatments (Metzger et al., 2000a).

Calcium

%WSC. The %WSC was affected (P < 0.05) by preacidification treatment, age, age × age, and the interaction
Table 1. Mean squares, probabilities (in parentheses), and degrees of freedom (df) for factors that may influence %WSC\(^1\) and water insoluble calcium content of low fat Mozzarella cheese during storage at 4°C.

<table>
<thead>
<tr>
<th>Factors</th>
<th>df</th>
<th>%WSC</th>
<th>Water insoluble calcium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole-plot</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Treatment(^2)</td>
<td>3</td>
<td>916.83(^*)</td>
<td>1888.01(^*)</td>
</tr>
<tr>
<td>Day of cheese</td>
<td>3</td>
<td>1089.66(^*)</td>
<td>875.59(^*)</td>
</tr>
<tr>
<td>Error</td>
<td>9</td>
<td>63.26</td>
<td>71.54</td>
</tr>
<tr>
<td>Subplot</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age(^3)</td>
<td>1</td>
<td>2628.42(^*)</td>
<td>13,342.20(^*)</td>
</tr>
<tr>
<td>Age × age</td>
<td>1</td>
<td>283.44(^*)</td>
<td>153.66(^*)</td>
</tr>
<tr>
<td>Interaction of</td>
<td>3</td>
<td>258.48(^*)</td>
<td>74.51(^*)</td>
</tr>
<tr>
<td>treatment × age</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Interaction of</td>
<td>3</td>
<td>28.80</td>
<td>12.86</td>
</tr>
<tr>
<td>treatment × age</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(age × age)</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Error</td>
<td>56</td>
<td>12.66</td>
<td>7.70</td>
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<tr>
<td>R(^2)</td>
<td></td>
<td>0.95</td>
<td>0.98</td>
</tr>
</tbody>
</table>

\(^1\)%WSC = water soluble calcium as a percentage of total calcium.

\(^2\)Treatments = control, pH 6.0 acetic, pH 5.8 acetic, and pH 5.8 citric.

\(^3\)Age = 2, 15, 30, 60, 90 d of refrigerated storage at 4°C.

*Statistically significant (P < 0.05).

of treatment × age (Table 1). Initially the %WSC ranged from 40 to 50% in all treatments (at 2 d of refrigerated storage), even though there were large differences in total calcium content among the preacidification treatments (Figure 1). The pH of all cheeses was similar at 2 d, as reported previously (Metzger et al., 2000a). However, during storage, the %WSC among treatments diverged and ranged from 40 to 75% at d 90 (Figure 1). The pH of the cheeses for different treatments also diverged with the pH of the control cheese increasing with time, while the pH of the cheese made from acetic acid preacidified milks remained the same and those made with citric acid decreased with time of storage (Metzger et al., 2000a). In the preacidified treatments, the %WSC increased, while in the control it remained stable. The change in %WSC during refrigerated storage indicated that changes in the cheese during refrigerated storage were affecting the equilibrium between WSC and water-insoluble calcium. The observed changes in %WSC during refrigerated storage might have been a result of changes in cheese pH during refrigerated storage. The equilibrium between micellar (bound) and nonmicellar (unbound) calcium is affected by pH in milk (van Hooydonk et al., 1986), and a similar situation may occur in cheese. To determine whether a relationship existed between %WSC and cheese pH, the %WSC was plotted as a function of cheese pH for all cheeses across time in this study (Figure 2), and a regression analysis was performed. A quadratic relationship was found between %WSC and cheese pH. Cheese pH was highly correlated (r = 0.84) with %WSC, and values ranged from 30% at pH 5.5 to 90% at pH 4.95.

**Water-insoluble calcium.** The water-insoluble calcium content of cheese was affected (P < 0.05) by preacidification treatment, age, age × age, and the interaction of treatment × age (Table 1). The preacidified treatments had less water-insoluble calcium than the control.
at all times (Figure 3), and this was correlated with lower total calcium content and lower cheese pH. As was the case with total calcium, water-insoluble calcium was lower for the pH 5.8 acetic treatment than for the pH 6.0 acetic treatment. Furthermore, the water-insoluble calcium was lower for the pH 5.8 citric treatment than the pH 5.8 acetic treatment. The observed difference in water-insoluble calcium between the preacidified treatments and the control was large, especially when the pH 5.8 citric treatment was compared with the control. In the control, the water-insoluble calcium was above 0.55 g/100 g at all times, while in the pH 5.8 citric treatment water-insoluble calcium was less than 0.30 g/100 g at all times. As was mentioned previously, the level of water-insoluble calcium may be more important for post-melt chewiness than total calcium because water-insoluble calcium represents the calcium that interacts with protein and influences cheese texture by cross-linking protein. At constant total calcium content, the proportion of water-insoluble calcium will be influenced by changes in pH during refrigerated storage.

### DISCUSSION

#### Factors Affecting Post-Melt Chewiness

The lower water-insoluble calcium content in the preacidified treatment cheeses (Figure 3) was associated with a reduction in the post-melt chewiness of low fat Mozzarella cheese (Table 2). Reduced water-insoluble calcium indicates that less calcium is available for cross-linking among caseins (Geurts et al., 1972; Solarza and Bell, 1995). However, a reduction in post-

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**Figure 3.** Effect of milk preacidification (control: ◆, pH 6.0 acetic: □, pH 5.8 acetic: ■, pH 5.8 citric: ●) on water-insoluble calcium (SEM = 0.037 g/100 g) of low fat Mozzarella cheese during storage at 4°C (LSD = 0.06, 0.06, 0.08, 0.05, and 0.09, at d 2, 15, 30, 60, and 90, respectively).
melt chewiness was also observed for all treatments during refrigerated storage. Two factors were changing within the cheese during refrigerated storage: 1) the amount of water-insoluble calcium was decreasing (Figure 3) for each preacidified cheese, and 2) the extent of proteolysis was increasing in all cheeses, as reported previously (Metzger et al., 2000a). This indicates that decreasing water-insoluble calcium and increasing proteolysis during refrigerated storage could play important roles in changes in post-melt chewiness.

In an effort to determine the effect of water-insoluble calcium, proteolysis, moisture, protein, and fat on post-melt chewiness, multiple regression analysis was performed. As described in the materials and methods section of this study, the 60- and 90-d data were pooled, and best subsets regression was performed. The single factor model $R^2$ were 0.72, 0.71, 0.11, 0.10, and 0.02 for water-insoluble calcium, pH 4.6 soluble N, fat, moisture, and protein, respectively. These results indicated that water-insoluble calcium and proteolysis had a larger effect on the post-melt chewiness of low fat Mozzarella cheese in this study than variation in fat, moisture, and protein content. These results are not surprising since there was a wide range of water-insoluble calcium and proteolysis in this study, while there was a very small range in fat, moisture, and protein content of the cheese. These results do not mean that cheese moisture, fat, and protein contents have no impact on post-melt chewiness. However, the large changes in post-melt chewiness during refrigerated storage in this study were associated with differences in water-insoluble calcium and proteolysis.

To determine whether proteolysis and water-insoluble calcium were explaining the same variation in post-melt chewiness, multiple regression analysis was performed, and the order of the terms in the statistical model was manipulated and significance of the sequential sums of squares was determined. In the first model, the order was pH 4.6 soluble N and then water-insoluble calcium, and in the second model the order was water-insoluble calcium and then pH 4.6 soluble N (Table 4). In both models the second term explained ($P < 0.05$) additional variation. Therefore, both pH 4.6 soluble N and water-insoluble calcium are associated with post-melt chewiness. However, these two factors were collinear with respect to their influence on post-melt chewiness and the separate effect of each factor cannot be easily uncoupled in this dataset. A separate experiment would be needed to clearly separate the impact of these variables on post-melt chewiness.

Factors Affecting Post-Melt Whiteness

The whiteness of low fat Mozzarella cheese (L-value) increased during heating and decreased during cooling for all treatments at all times of refrigerated storage.
Figure 4. a) Effect of milk preacidification (control: ○, pH 6.0 acetic; □, pH 5.8 acetic; ■, pH 5.8 citric; ●) on changes in L-value (SEM = 0.529) during heating and cooling of low fat Mozzarella cheese after 30 d of storage at 4°C (LSD = 2.02, 1.69, and 5.05, at 7°C before heating, 60°C after heating, and 7°C after cooling, respectively); b) Effect of milk preacidification on changes in L-value (SEM = 0.944) during heating and cooling of low fat Mozzarella cheese after 60 days of storage at 4°C (LSD = 2.47, 4.05, and 9.26, at 7°C before heating, 71°C after heating, and 7°C after cooling, respectively); and c) Effect of milk preacidification on changes in L-value (SEM = 0.974) during heating and cooling of low Mozzarella cheese after 90 d of storage at 4°C (LSD = 2.38, 6.95, and 8.01, at 7°C before heating, 71°C after heating, and 7°C after cooling, respectively).

Figure 5. a) Effect of water-insoluble calcium on the post-melt whiteness (L-value at 38°C after heating and cooling) of low fat Mozzarella cheese; b) Effect of pH 4.6 soluble N on the post-melt whiteness (L-value at 38°C after heating and cooling) of low fat Mozzarella cheese; and c) Effect of composite factor (water-soluble calcium/pH 4.6-soluble N) on the post-melt whiteness (L-value at 38°C after heating and cooling) of low fat Mozzarella cheese.
(Figure 4a, b, and c). However, whiteness at 71°C at 60 and 90 of refrigerated storage was lower in the preacidiﬁed treatments than the control and whiteness at 7°C after heating and cooling was lower in the preacidiﬁed cheeses than the control at all times of storage. Furthermore, the whiteness at 7°C after heating and cooling decreased during refrigerated storage for all treatments. The increase in whiteness of low fat Mozzarella during heating is associated with the formation of light scattering gel particles in the serum phase of cheese, and during cooling the decrease in whiteness is a result of solubilization of these light scattering protein gel particles in the serum phase of cheese (Metzger et al., 2000b). Hydrophobic interactions among casein and peptides derived from casein dissolved in the serum phase of cheese are thought to be responsible for the formation of these light scattering gel particles (Metzger et al., 2000b). The observed effects of preacidiﬁcation on whiteness during heating and cooling (lower L-value during heating and after heating cooling) may be a result of modiﬁcations in protein-protein interactions in the serum phase of cheese.

During baking on a pizza, a cheese temperature of 60 to 70°C is obtained under typical baking conditions (232°C for 5 min using an industrial pizza oven) immediately after baking (Kindstedt et al., 1989). After baking, the cheese begins to cool, and it is typically consumed at 40 to 50°C. As a result, the whiteness of low fat Mozzarella at 38°C after heating and cooling may be important and may be a factor in the consumer acceptability of low fat Mozzarella cheese on a pizza. To determine whether there was a relationship between L-value at 38°C (after heating and cooling) and water-insoluble calcium or proteolysis, the L-values of all samples at all times of storage (30, 60, and 90 d) were plotted as a function of water-insoluble calcium (Figure 5a) and pH 4.6 soluble N (Figure 5b). At low levels of pH 4.6, soluble N (<8%) or high levels of water-insoluble calcium (>0.50%) the L-value at 38°C after heating and cooling was high (>85). Thus, the L-value at 38°C after heating and cooling may be associated with water-insoluble calcium and proteolysis.

To determine whether proteolysis and water-insoluble calcium explained the variation in post-melt whiteness, multiple regression analysis was performed, and the order of the terms in the statistical model was manipulated and signiﬁcance of the sequential sums of squares was determined. In the ﬁrst model, the order was pH 4.6 soluble N and then water-insoluble calcium, and in the second model the order was water-insoluble calcium and then pH 4.6 soluble N (Table 5). In both models, the second term explained (P < 0.05) additional variation. Thus, in this study both proteolysis and water-insoluble calcium were associated with changes in post-melt whiteness.

A composite factor was calculated by dividing the amount of water-insoluble calcium by the pH 4.6 soluble N. The L-value at 38°C after heating and cooling for each cheese at all times (30, 60, and 90 d) was plotted as a function of the composite factor (Figure 5c). If the composite factor was less than 0.05, the post-melt L-value of the cheese at 38°C was low. Thus, at high levels of proteolysis or low levels of water-insoluble calcium, the whiteness of low fat Mozzarella cheese on a pizza after baking will be low, which may adversely affect consumer acceptance of low fat Mozzarella cheese on a pizza.

### Strategies to Improve the Quality of Low Fat Mozzarella Cheese

To be acceptable, the unmelted and melted functional properties and post-melt properties of low fat Mozzarella need to be similar to LMPS Mozzarella. Previous research in our laboratory has demonstrated that the melting and browning characteristics of low fat Mozzarella cheese during baking can be controlled with a hydrophobic surface coating (Rudan and Barbano, 1998). In addition, the unmelted and melted functional properties (e.g., TPA hardness and apparent viscosity) of low fat Mozzarella immediately after manufacture are similar to LMPS Mozzarella after 30 d of refrigerated storage if milk preacidification to pH 5.8 with citric acid is used (Metzger et al., 2000a).

### Table 4. Multiple regression analysis of post-melt chewiness versus water insoluble calcium and pH 4.6 soluble N.

<table>
<thead>
<tr>
<th>Model</th>
<th>Source of variation</th>
<th>Sequential SS</th>
<th>F-test</th>
<th>P-value</th>
</tr>
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<tr>
<td>1</td>
<td>pH 4.6 soluble N</td>
<td>17,820</td>
<td>97.38</td>
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</tr>
<tr>
<td></td>
<td>Water insoluble calcium</td>
<td>2003</td>
<td>10.94</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>2</td>
<td>Water insoluble calcium</td>
<td>18,036</td>
<td>98.56</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td></td>
<td>pH 4.6 soluble N</td>
<td>1786</td>
<td>9.76</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>

*Post-melt chewiness = pH 4.6 soluble N, water insoluble calcium.
*Post-melt chewiness = water insoluble calcium, pH 4.6 soluble N.
The results of the current study indicate that the post-melt chewiness of low fat Mozzarella cheese can be reduced with high levels of proteolysis or low levels of water-insoluble calcium or a combination of both factors. However, high levels of proteolysis and low levels of water-insoluble calcium decrease the post-melt whiteness of low fat Mozzarella cheese. Low post-melt whiteness may adversely affect the consumer acceptability of low fat Mozzarella cheese. It has been demonstrated previously (Rudan and Barbano, 1998) that the addition of homogenized cream to skim milk used to manufacture reduced fat Mozzarella cheese prevented a translucent post-bake color after cooling, even when substantial proteolysis had occurred. The effect of reducing fat particle size on the post-melt whiteness of prec acidified low fat Mozzarella cheese needs to be determined.

CONCLUSIONS

The post-melt chewiness and whiteness of low fat Mozzarella cheese decreased with increasing level of milk preacidification. Milk preacidification resulted in a reduction in both total cheese calcium and water-insoluble calcium. Changes in %WSC were correlated with changes in cheese pH during refrigerated storage. As a result, total calcium content and changes in cheese pH during storage affected the amount of water-insoluble calcium in cheese. The post-melt chewiness of low fat Mozzarella decreased as proteolysis increased and water-insoluble calcium content of the cheese decreased. Given the experimental design in the present study, it was not possible to determine which of the two factors has the largest impact on post-melt chewiness. In addition, the changes in post-melt whiteness of low fat Mozzarella cheese were associated with changes in proteolysis and water-insoluble calcium. High levels of proteolysis and low levels of water-insoluble calcium were correlated with a decrease in the post-melt whiteness of low fat Mozzarella cheese.

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REFERENCES


Table 5. Multiple regression analysis of post-melt whiteness versus water insoluble calcium and pH 4.6 soluble N.

<table>
<thead>
<tr>
<th>Model</th>
<th>Source of variation</th>
<th>Sequential SS</th>
<th>F-test</th>
<th>P-value</th>
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</thead>
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<tr>
<td>1</td>
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<td>1827.25</td>
<td>40.97</td>
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<td>Water insoluble calcium</td>
<td>205.85</td>
<td>4.62</td>
<td>0.04</td>
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<tr>
<td>2</td>
<td>Water insoluble calcium</td>
<td>1617.91</td>
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<td>pH 4.6 soluble N</td>
<td>415.19</td>
<td>9.31</td>
<td>&lt;0.01</td>
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</tbody>
</table>

1Post-melt whiteness = pH 4.6 soluble N, water insoluble calcium.
2Post-melt whiteness = water insoluble calcium, pH 4.6 soluble N.