# Effect of Fat Reduction on Chemical Composition, Proteolysis, Functionality, and Yield of Mozzarella Cheese<sup>1</sup>

ABSTRACT

Mozzarella cheese was made from skim milk standardized with cream (unhomogenized, 40% milk fat) to achieve four different target fat percentages in the cheese (ca. 5, 10, 15, and 25%). No statistically significant differences were detected for cheese manufacturing time, stretching time, concentration of salt in the moisture phase, pH, or calcium as a percentage of the protein in the cheese between treatments. As the fat percentage was reduced, there was an increase in the moisture and protein content of the cheese. However, because the moisture did not replace the fat on an equal basis, there was a significant decrease in the moisture in the nonfat substance in the cheese as the fat percentage was reduced. This decrease in total filler volume (fat plus moisture) was associated with an increase in the hardness of the unmelted cheese. Whiteness and opacity of the unmelted cheese decreased as the fat content decreased. Pizza baking performance, meltability, and free oil release significantly decreased as the fat percentage decreased. The minimum amount of free oil release necessary to obtain proper functionality during pizza baking was between 0.22 and 2.52 g of fat/100 g of cheese. Actual cheese yield was about 30% lower for cheese containing 5% fat than for cheese with 25% fat. Maximizing fat recovery in the cheese becomes less important to maintain high cheese yield, and moisture control and the retention of solids in the water phase become more important as the fat content of the cheese is reduced.

MICHAEL A. RUDAN, DAVID M. BARBANO,

J. JOSEPH YUN,<sup>2</sup> and PAUL S. KINDSTEDT<sup>3</sup> Northeast Dairy Foods Research Center, Department of Food Science, Cornell University, Ithaca, NY 14853

(**Key words**: fat reduction, Mozzarella cheese, composition, functionality)

**Abbreviation key**: **AV** = apparent viscosity, **FO** = free oil, **LMPS** = low moisture part skim, **MNFS** = moisture in the nonfat substance, **TPA** = texture profile analysis.

#### INTRODUCTION

Reduction of fat intake in the American diet is recommended by the US Department of Agriculture (32). Fat reduction in the diet is important based on the scientific evidence linking diets high in fat to coronary heart disease and certain types of cancer (36). To help consumers achieve healthier eating, the food industry has responded by developing reduced fat foods. In 1994, the cheese industry introduced 247 new cheese products, many of which were reduced fat cheeses (8, 22).

Driven by the popularity of pizza, Mozzarella cheese sales continue to grow. Low moisture partskim (**LMPS**) Mozzarella cheese is commonly used for pizza because of its desirable functionality (13). However, LMPS Mozzarella cheese, by definition, contains 14 to 22% fat, depending on its moisture content (9). Given the healthier eating goals of consumers and the continued demand for pizza, there has been an interest in developing a lower fat Mozzarella cheese (10, 20, 29, 30, 31).

Fat performs many important functions within a food. For cheese, fat contributes to the taste, texture, functionality, and appearance. Because about 75% of all Mozzarella cheese is used as an ingredient for pizza, proper melt (shred fusion) and appearance (browning and blistering upon heating) are important characteristics (1, 12). When fat is removed from cheese, the overall quality of the cheese decreases (10, 20, 29, 30, 31).

Tunick et al. (31) used homogenization of milk and a lower cooking temperature as a means to enhance the functionality of reduced fat Mozzarella cheese. Those researchers found that homogenization resulted in a reduced fat Mozzarella cheese (ca. 10%

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 $<sup>^2</sup> Upstate$  Farms Cooperative, Inc., 196 Scott St., Buffalo, NY 14204.

<sup>&</sup>lt;sup>3</sup>Department of Animal Science and Food Science, University of Vermont, Burlington 05405.

fat) that was harder and less meltable than Mozzarella cheese at the same fat content that was made from unhomogenized milk. Use of a lower cook temperature, in combination with homogenization, increased the moisture content of the cheese. This increase in moisture decreased the hardness and increased the meltability of the reduced fat Mozzarella cheese (31). In a later study, Tunick et al. (30) stored a reduced fat Mozzarella cheese (7% fat) for 10 wk, made only minor modifications to the manufacturing procedure, and produced a cheese with functionality during pizza baking that was similar to LMPS Mozzarella cheese. However, excessive browning of the 10-wk-old reduced fat cheese occurred when it was cooked at temperatures above  $175^{\circ}C$  (30).

Merrill et al. (20) used a modified manufacturing procedure to retain more moisture in the reduced fat Mozzarella cheese (10 to 15% fat). This method produced a cheese with the same apparent viscosity (**AV**), meltability, and browning as LMPS Mozzarella cheese (20). Fife et al. (10) used the procedure of Merrill et al. (20) and further reduced the fat content to meet the FDA requirement for low fat Mozzarella cheese (less than 6% fat). Overall, increased moisture contents improved some of the qualities of low fat Mozzarella cheese. However, those researchers (10) concluded that more studies were needed on improved functionality during cooking.

The objective of this study was to identify the important changes in composition and proteolysis and their effect on cheese functionality when progressive amounts of fat were removed from Mozzarella cheese. The current study differs from previous research on lower fat Mozzarella cheese as follows: 1) to help understand changes in cheese composition and functionality given changes in fat content, no changes were made to the manufacturing procedure to manipulate cheese composition; 2) to better simulate large-scale Mozzarella cheese manufacturing conditions, the cheese curd was stretched with a twinscrew pilot-scale Mozzarella mixer (model 640; Stainless Steel Fabricating, Columbus, WI) and not manually; and 3) to gain an understanding of the important economic aspects of cheese manufacture, the effect of fat reduction on cheese yield was determined.

#### MATERIALS AND METHODS

# Fat Standardization of Cheese Milk

Raw skim milk and cream were obtained from the Cornell University dairy plant. The fat content of the raw skim milk [(19); method number 15.8.B.] and cream was determined by Babcock [(2); method number 33.3.18, 995.18] as a guide for standardization. Four batches of milk (ca. 250 kg each) were standardized by the addition of cream (ca. 40% fat) to skim milk to 0.4, 0.8, 1.6, and 3.2% fat (wt/wt) to achieve four target fat percentages in the cheese of 5, 10, 15, and 25%, respectively. The batches of standardized milk were pasteurized (Model Universal Pilot Plant; PMS Processing Machinery and Supply Co., Philadelphia, PA) at 72°C for 16 s, cooled to 4°C, and stored overnight at 4°C until used for cheese manufacture the next day.

#### **Cheese Manufacturing Method**

To produce a homogeneous chemical composition, cheese was made using the no brine, stirred-curd method as previously described (6). Eight to 10 cylinders (7.5 cm in diameter  $\times$  30 cm long) containing about 1.4 kg of cheese per cylinder were made per vat and were stored at 4°C.

#### **Chemical Analysis**

Compositional analyses of the milk, whey, stretching water, and cheese were done as previously described (27). As a measure of proteolysis, the nitrogen that was soluble in 12% TCA and the nitrogen that was soluble in pH 4.6 acetate buffer extracts of the cheese were determined at 5, 20, 33, and 49 d of refrigerated storage as described by Bynum and Barbano (7). Both soluble protein values were expressed as a percentage of the total protein content of the cheese. Proteolysis of  $\alpha_{s1}$ -CN plus  $\alpha_{s2}$ -CN and  $\beta$ -CN during refrigerated storage was determined using SDS-PAGE as previously described (34, 37). Because of the extreme toughness of the low fat cheese, an electrically powered homogenizer (model 17105, Omni-Mixer Homogenizer, Omni International, Waterbury, CT), instead of a hand-operated tissue homogenizer, was used to homogenize the cheese in the sample buffer. Samples were prepared after 5, 20, and 49 d of refrigerated storage.

#### **Cheese Functional Properties**

Texture profile analysis (**TPA**), as defined by Bourne (3), was conducted on unmelted cheese cylinders (2 cm in diameter  $\times$  2 cm high) at 10°C, 50% compression, and a cross-head speed of 12.7 cm/ min in quadruplicate for each treatment after 5 d of refrigerated storage. A modified Schreiber test (16) was used to quantify cheese meltability. Procedures for the TPA and melt test were as previously described (38). The AV was determined using helical viscometry (14), and free oil (**FO**) was determined using a centrifugation method (15). Meltability, AV, and FO tests were performed after 5, 20, 36 (33 for meltability), and 49 d of storage at  $4^{\circ}$ C.

A pizza baking test (25) was used to evaluate the functionality of the cheese when used as a topping for pizza. Photographs (35 mm, 200 speed film, natural light) were taken after baking to record the data. The pizza baking test was conducted for each treatment after 30 d of storage at  $4^{\circ}$ C.

# Recoveries and Yield Calculations

The actual percentages of fat and N recoveries in the cheese, whey, and stretching water and the actual cheese yield were calculated as previously described (17). Theoretical yield was calculated using the Van Slyke and Price formula as modified by Barbano and a new general method cheese yield formula (4). The original formula of Van Slyke and Price (33) for Cheddar cheese yield was modified to reflect Mozzarella cheese yield. Modifications included changing the assumed fat recovery value from 0.93 to 0.85, changing the constant factor from 1.09 to 1.13 (4), and changing the desired cheese moisture to the actual cheese moisture for a given vat. A new general method (Barbano formula) for calculating Mozzarella cheese yield was also used (4). The Barbano formula includes the SNF content of the separated whey to estimate the amount of whey solids retained in the water phase of the cheese. Values of 0.85 and 1.092 were used in the Barbano formula (4) for the retention factors for fat and calcium phosphate, respectively. The solute exclusion factor in the Barbano formula (4) was calculated for each fat level using the data from this study; factors were 0.8534, 0.8246, 0.7640, and 0.5420 for the 5, 10, 15, and 25% target cheese fat percentages, respectively. The actual percentages of moisture and salt in the cheese were also used in the calculation of theoretical yield with this formula.

# Experimental Design and Statistical Analysis

A 4  $\times$  4 randomized complete block design was used. On each day of cheese manufacturing, four different fat percentages of cheese were made; this procedure was repeated four times. Changes in proteolysis and functional properties (meltability, AV, and FO) during refrigerated storage were monitored using a split-plot design in which the whole-plot factor (fat level) was replicated in a 4  $\times$  4 randomized complete block design. For the whole-plot factor, fat level was analyzed as a classification variable, and day of cheese making was blocked. For the subplot factor, age and the quadratic form of age (i.e.,  $age^2$ ) were analyzed as quantitative variables. The degrees of freedom in the statistical model were the same for both proteolysis and functional properties. The PROC GLM of SAS (28) was used for all data analyses.

### **RESULTS AND DISCUSSION**

### Composition of Milk, Whey, Stretching Water, and Cheese

Chemical composition of the milk, whey, stretching water, and cheese is shown in Table 1. The fat content of the standardized milks ranged from 3.21 to 0.35% to achieve the desired reductions in the fat content of the cheeses (Table 1). As expected, the observed total protein and casein contents of the milks gradually decreased as the fat content of the milk increased. The casein to fat ratio increased (P < 0.05) from 0.73 to 6.89 as the fat content in the milk decreased. As expected, there was no statistically significant difference in the calculated SNF content of the skim portion of the four milks (data not shown).

As the fat content of the milk decreased, there was a significant decrease in the fat and total solids contents of the whey and in the fat content of the stretching water. The protein contents of the whey and stretching water differed only slightly or not at all among treatments (Table 1). The significance of the milk solids in the whey is discussed in the "Fat and Nitrogen Recoveries" section of the present study.

The chemical composition of the cheeses is shown in Table 1. As fat content decreased, moisture, protein, and calcium contents increased significantly. Because the moisture did not replace the fat on an equal basis, there was a significant decrease in the moisture in the nonfat substance (MNFS) and in the moisture to protein ratio in the cheese as the fat percentage was reduced. This decrease in total filler volume (fat plus moisture) was expected to affect the texture of the cheese, and these results are discussed later. Total cheese manufacturing time (results not shown), pH, salt, and calcium as a percentage of protein were not affected by variations in fat content. The mean cheese manufacturing time from rennet addition to the end of direct salting prior to stretching was 105 min.

#### **Proteolysis**

The soluble nitrogen content of the cheese expressed as a percentage of the cheese weight (results

not shown) and expressed as a percentage of the total nitrogen of the cheese (Table 2) differed significantly between cheeses of different fat contents. As the fat content of the cheese decreased, both the pH 4.6-soluble nitrogen and 12% TCA-soluble nitrogen as a percentage of total nitrogen decreased (Figures 1 and 2, respectively). Age had a large impact on the soluble nitrogen content of cheese (Table 2). During refrigerated storage, both the pH 4.6-soluble nitrogen and 12% TCA-soluble nitrogen significantly increased (Figures 1 and 2). Furthermore, there was a significant interaction of fat level and age (Table 2), indicating that the differences in the rates of increase in soluble nitrogen were significantly different between cheeses of differing fat percentages (Figures 1 and 2).

As the fat content of the cheese was reduced, the protein and moisture content significantly increased, but, in general, both the moisture to protein ratio and the MNFS decreased (Table 1). Because residual coagulant (chymosin) is primarily responsible for the initial hydrolysis of caseins occurring in Mozzarella cheese during refrigerated storage (5), the increase in the moisture (which contains soluble chymosin) content of the cheese with decreasing fat content might have been expected to increase the amount of proteolysis. However, the amount of pH 4.6-soluble nitrogen and 12% TCA-soluble nitrogen significantly decreased (Table 2; Figures 1 and 2) as the protein and moisture contents of Mozzarella cheese increased. The trends in proteolysis (Figures 1 and 2) appear to be directly related to the ratio of moisture to protein in the cheese. That is, the two higher fat cheeses contained more moisture per unit of protein (and had

		Targ				
Component	5%	10%	15%	25%	SEM	LSD <sup>2</sup>
Milk						
pН	6.65 <sup>a</sup>	6.62 <sup>b</sup>	6.62 <sup>b</sup>	6.62 <sup>b</sup>	0.01	0.02
Fat, %	0.35 <sup>d</sup>	0.81 <sup>c</sup>	1.59 <sup>b</sup>	3.21 <sup>a</sup>	0.01	0.05
Protein, %	3.13 <sup>a</sup>	3.11 <sup>a</sup>	3.10 <sup>a</sup>	3.03 <sup>b</sup>	0.01	0.04
Casein, %	2.41 <sup>a</sup>	2.39 <sup>a</sup>	2.39 <sup>a</sup>	2.34 <sup>b</sup>	0.01	0.03
NPN, %	0.21 <sup>b</sup>	$0.22^{\mathrm{a}}$	$0.22^{\mathrm{a}}$	0.21 <sup>b</sup>	0.00	0.01
C:F <sup>3</sup>	6.89 <sup>a</sup>	$2.95^{b}$	1.50 <sup>c</sup>	0.73 <sup>d</sup>	0.08	0.24
Whey						
Fat, %	0.07 <sup>d</sup>	0.11 <sup>c</sup>	0.21 <sup>b</sup>	0.53 <sup>a</sup>	0.01	0.03
Protein, %	0.91	0.89	0.89	0.88	0.02	0.05
Total solids, %	6.59 <sup>c</sup>	6.64 <sup>c</sup>	6.77 <sup>b</sup>	7.02 <sup>a</sup>	0.03	0.09
Stretching water						
Fat, %	0.06 <sup>c</sup>	0.16 <sup>c</sup>	0.43 <sup>b</sup>	1.18 <sup>a</sup>	0.06	0.18
Protein, %	0.08 <sup>ab</sup>	0.07 <sup>b</sup>	0.09 <sup>ab</sup>	0.11 <sup>a</sup>	0.01	0.03
Cheese						
pН	5.12	5.12	5.09	5.09	0.02	0.05
Moisture, %	53.19 <sup>a</sup>	51.01 <sup>b</sup>	48.34 <sup>c</sup>	43.19 <sup>d</sup>	0.33	1.05
Fat, %	4.10 <sup>d</sup>	9.41 <sup>c</sup>	16.48 <sup>b</sup>	26.73 <sup>a</sup>	0.17	0.55
FDB <sup>4</sup> , %	8.77 <sup>d</sup>	19.20 <sup>c</sup>	31.90 <sup>b</sup>	47.06 <sup>a</sup>	0.20	0.65
Protein, %	35.54 <sup>a</sup>	32.85 <sup>b</sup>	29.02 <sup>c</sup>	24.92 <sup>d</sup>	0.25	0.81
M:P <sup>5</sup>	1.50 <sup>b</sup>	1.55 <sup>b</sup>	1.67 <sup>a</sup>	1.73 <sup>a</sup>	0.03	0.08
MNFS <sup>6</sup> , %	55.47 <sup>c</sup>	56.31 <sup>c</sup>	57.88 <sup>b</sup>	58.94 <sup>a</sup>	0.31	1.00
Salt, %	1.69	1.70	1.60	1.56	0.06	0.18
S:M <sup>7</sup> , %	3.18 <sup>b</sup>	3.34 <sup>b</sup>	3.31 <sup>b</sup>	3.62 <sup>a</sup>	0.12	0.39
Ca %	1.00 <sup>a</sup>	0.90 <sup>b</sup>	0.82 <sup>c</sup>	0.67 <sup>d</sup>	0.02	0.07
Ca (% of P) <sup>8</sup>	2.80	2.75	2.84	2.68	0.06	0.20

TABLE 1. Mean chemical composition of milk, whey, stretching water, and cheese.

a.b.c.dMeans within same row with no common superscript differ (P < 0.05). Rows with no superscripts indicated no differences (P > 0.05) between means.

<sup>1</sup>Target cheese fat on a cheese weight basis (n = 4).

<sup>2</sup>Least significant difference (P = 0.05).

<sup>3</sup>Casein to fat ratio.

 ${}^{4}\text{Fat}$  content on a dry weight basis.

<sup>5</sup>Ratio of moisture to protein.

<sup>6</sup>Moisture in the nonfat substance of the cheese.

<sup>7</sup>Percentage of salt in moisture in the cheese.

<sup>8</sup>Calcium as a percentage of protein content of the cheese.

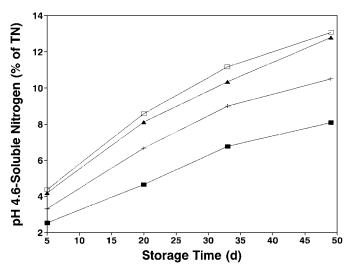
Factors	df	pH 4.6- Soluble nitrogen	12% TCA- Soluble nitrogen	$\alpha_{s1}$ -CN Plus $\alpha_{s2}$ -CN <sup>1</sup>	β-CN <sup>1</sup>
Whole plot					
Fat level (F)	3	23.53* ( <i>P</i> < 0.01)	4.93* ( <i>P</i> < 0.01)	$25.95^*$ ( $P = 0.02$ )	1.45 ( <i>P</i> = 0.23)
Day of cheese making	3	2.10 ( $P = 0.25$ )	0.55 ( $P = 0.28$ )	21.97* ( <i>P</i> = 0.04)	3.18 ( $P = 0.05$ )
Error	9	1.29	0.37	4.91	0.83
Subplot					
Age (A)	1	491.36* ( <i>P</i> < 0.01)	$115.72^*$ ( $P < 0.01$ )	$5143.51^*$ ( $P < 0.01$ )	1.32 ( <i>P</i> = 0.31)
$(\mathbf{A} \times \mathbf{A})$	1	12.94* ( <i>P</i> < 0.01)	2.12 ( <i>P</i> < 0.01)	362.18* ( <i>P</i> < 0.01)	0.05 ( $P = 0.85$ )
Interaction $(F \times A)$	3	4.12* ( <i>P</i> < 0.01)	1.77* ( <i>P</i> < 0.01)	1.15* ( <i>P</i> < 0.01)	1.05 ( $P = 0.48$ )
$\begin{array}{l} \text{Interaction} \\ \text{F} \times (\text{A} \times \text{A}) \end{array}$	3	0.45 ( $P = 0.10$ )	0.08 ( $P = 0.41$ )	1.97* ( <i>P</i> < 0.01)	0.43 ( $P = 0.79$ )
Error	40	0.21	0.08	1.65	1.22
R <sup>2</sup>		0.988	0.979	0.993	0.493

TABLE 2. Mean squares, probabilities, and degrees of freedom for indices of proteolytic changes of Mozzarella cheese during 49 d storage at 4°C.

 $^{1}As$  determined by Verdi et al. (34).

\*Statistically significant (P < 0.05).

more proteolysis), and the two lower fat cheeses contained less moisture per unit of protein (and had less proteolysis). Tunick et al. (31) also found that MNFS (a measure of the ratio of moisture to protein) influenced the amount of proteolysis occurring in Mozzarella cheese. In addition, Fife et al. (10) found that caseins in low fat Mozzarella cheese (65 to 66.2% MNFS; 2.2 to 5% fat) underwent proteolysis similar to that of LMPS Mozzarella cheese (64.5% MNFS; 19.3% fat). Unfortunately, because of an analytical problem in the study by Fife et al. (10) the moisture content had to be estimated; therefore, the actual moisture was not known, which makes their results difficult to interpret. Based on the present study and



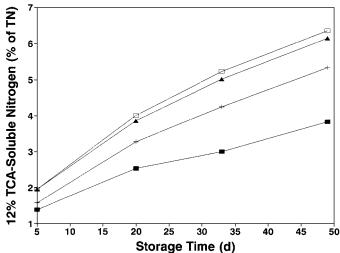


Figure 1. The effect of fat percentage on pH 4.6-soluble nitrogen in the cheese, as a percentage of the total nitrogen (TN), during storage at 4°C for target levels of 5% fat ( $\blacksquare$ ), 10% fat (+), 15% fat ( $\blacktriangle$ ), and 25% fat ( $\Box$ ). SEM = 0.23%.

Figure 2. The effect of fat percentage on 12% TCA-soluble nitrogen in the cheese, as a percentage of total nitrogen (TN), during storage at 4°C for target levels of 5% fat ( $\blacksquare$ ), 10% fat (+), 15% fat ( $\blacktriangle$ ), and 25% fat ( $\Box$ ). SEM = 0.14%.

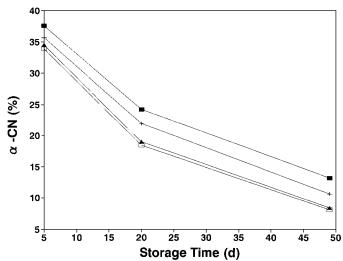


Figure 3. The effect of fat percentage on  $\alpha_{s1}$ -CN plus  $\alpha_{s2}$ -CN in the cheese during storage at 4°C for target levels of 5% fat (**■**), 10% fat (+), 15% fat (**▲**), and 25% fat (**□**). SEM = 0.64%.

the work of Tunick et al. (31), the ratio of moisture to protein or the MNFS appear to influence the amount of proteolysis that occurs in Mozzarella cheese.

At the different fat levels, the  $\alpha_s$ -CN ( $\alpha_{s1}$ -CN plus  $\alpha_{s2}$ -CN) were subject to hydrolysis, but  $\beta$ -CN remained unchanged during refrigerated storage (Table 2; Figure 3). This result is consistent with the observations of Tunick et al. (31) and Fife et al. (10). The amount of proteolysis occurring in Mozzarella cheese is important because it may have an effect on the functional properties of the unmelted and melted cheeses.

### Unmelted Cheese Functional Properties

**TPA.** After 5 d of refrigerated storage, fat percentage significantly affected all TPA parameters measured. As the fat content of the cheese decreased, the TPA hardness 1, cohesiveness, and springiness increased (Table 3).

The filled gel composite model has been used (18, 26, 35) to help understand the changes in the rheological characteristics of unmelted cheese as a result of fat reduction. By electron microscopy, Mozzarella cheese has been determined to be a composite material made up of irregular fat and whey columns surrounded and supported by a fibrous threedimensional protein matrix (21, 24, 29). With respect to the filled gel composite model, the fat and whey (mostly moisture) represent the filler within the casein network (gel). If the fat has no interaction (molecular bonding, colloidal forces, or friction) with the matrix, then, as its volume fraction is decreased, there is more matrix to deform per unit volume, and, consequently, the composite should get harder and more difficult to melt (35).

Fat reduction in the cheese increased the moisture and protein contents (Table 1). Consistent with the predictions of the filled gel composite theory (35) and the work of Tunick et al. (31), the TPA hardness 1 of the unmelted cheese increased as the fat content and total filler volume decreased and the amount of matrix (protein) increased (35). Although the moisture content increased, it did not completely offset the decrease in fat as indicated by the steady increase in protein content as the fat content was reduced (Table 1). Thus, the total filler volume (fat plus moisture) of the Mozzarella cheese also decreased progressively from 69.92, 64.82, 60.42, to 57.29% as the fat content decreased.

Springiness and cohesiveness of the unmelted cheese increased as the fat content decreased (Table 3). Tunick et al. (31) also found that the springiness was higher for low fat Mozzarella cheese than for full fat Mozzarella cheese. Those researchers (31) thought that the absence of fat resulted in a more flexible protein network. Also, because more protein and more intact protein were present in the lower fat

TABLE 3. Texture profile analysis (TPA) parameters of Mozzarella cheeses made with four different fat percentages after 5 d of storage at  $4^{\circ}$ C.

ТРА	Target fat <sup>1</sup>					
Parameter	5%	10%	15%	25%	SEM	LSD <sup>2</sup>
Hardness, N Cohesiveness Springiness, mm	219.50ª 0.71ª 7.57ª	169.92 <sup>b</sup> 0.61 <sup>b</sup> 7.35 <sup>a</sup>	94.02 <sup>c</sup> 0.60 <sup>b</sup> 6.79 <sup>b</sup>	64.21 <sup>d</sup> 0.47 <sup>c</sup> 5.22 <sup>c</sup>	9.21 0.02 0.09	29.4 0.06 0.30

a,b,c,dMeans within same row with no common superscript differ (P < 0.05).

<sup>1</sup>Target cheese fat on a cheese weight basis (n = 4).

<sup>2</sup>Least significant difference (P = 0.05).

TABLE 4. Mean squares and probabilities of the meltability, apparent viscosity (AV), and free oil (FO) release of Mozzarella cheese during 49 d storage at  $4^{\circ}$ C.

		AV	FO
Factors	Meltability	(×10 <sup>6</sup> )	FO
Whole plot			
Fat level (F)	106.76*	0.836	2173.69*
	(P < 0.01)	(P = 0.07)	(P < 0.01)
Day of	4.73	1.105*	16.76
cheese making	(P = 0.44)	(P = 0.04)	(P = 0.40)
Error	4.82	0.254	15.12
Subplot			
Age (A)	660.18*	203.145*	24.02*
	(P < 0.01)	(P < 0.01)	(P = 0.03)
$\mathbf{A} \times \mathbf{A}$	22.85*	58.852*	59.23*
	(P < 0.01)	(P < 0.01)	(P < 0.01)
Interaction	10.05*	0.913*	6.50
$(F \times A)$	(P < 0.01)	(P = 0.03)	(P = 0.29)
Interaction	0.95	0.061	7.29
$\mathbf{F} \times (\mathbf{A} \times \mathbf{A})$	(P = 0.65)	(P = 0.89)	(P = 0.24)
Error	1.76	0.284	5.00
R <sup>2</sup>	0.955	0.961	0.988

\*Statistically significant (P < 0.05).

cheeses, more matrix was available per unit of volume to restore the cheese to its original shape after a 50% compression. The change in cohesiveness could have implications during shredding as cheese clumping can cause problems during commercial shredding operations (13).

**Cheese appearance.** Fat content significantly affected the appearance of the unmelted cheese, both unshredded and shredded. Fat reduction made the cheese less white and more translucent. Merrill et al. (20) also observed a color change in Mozzarella cheese as the fat content was lowered from 19.3 to 10.3%. By visual inspection, they reported that fat reduction increased the translucency and produced a greenish tint in the unmelted cheese. Mozzarella cheese appearance is an important attribute, and more work is needed in this area first, to quantify the effect of fat reduction on cheese appearance and, second, to improve (i.e., increase whiteness and opacity) the appearance of unmelted low fat Mozzarella cheese.

# Melted Cheese Functional Properties

Differences in melted cheese functionality (meltability, AV, FO, and pizza baking) were primarily due to differences in the original composition of the cheese (fat, protein, calcium, and moisture), to differences in proteolysis of the cheese as a result of these compositional differences, or both.

**Melt.** Fat percentage significantly affected the meltability of the cheese measured using the modified Schreiber test (Table 4). As the fat content of the cheese decreased, the melted diameter of the cheese disc decreased (Figure 4). Tunick et al. (31) also found that Mozzarella cheese meltability decreased as the fat content decreased. Fife et al. (10) and Merrill et al. (20), using a modified cheese manufacture procedure to elevate moisture content, found that fat percentage had no influence on the meltability of Mozzarella cheese. However, the test tube-type melt test used by Fife et al. (10) and Merrill et al. was different from the modified Schreiber (16) disc-type melt test used in our study and that of Tunick et al. (31).

Age had a large impact on the meltability of the cheese (Table 4). During refrigerated storage, the meltability of the cheese significantly increased at all fat percentages. After 49 d of refrigerated storage, the two higher fat cheeses (15 and 25% fat) had similar meltability (Figure 4). Furthermore, after 49 d of refrigerated storage, the two lower fat cheeses (5 and 10% fat) melted about the same as the two higher fat cheeses did initially at d 5 (Figure 3). An increase in the meltability of cheese during storage is consistent with the observations of other researchers (10, 20, 31). As the matrix was degraded by proteolysis, the ability of the cheese to maintain its structure during heating decreased (31). Thus, for a given amount of

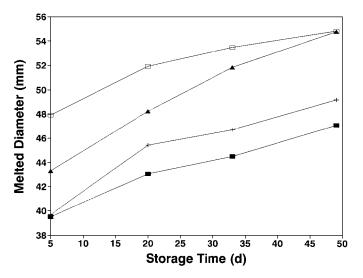


Figure 4. The effect of fat percentage on meltability during storage at 4°C for target levels of 5% fat ( $\blacksquare$ ), 10% fat (+), 15% fat ( $\blacktriangle$ ), and 25% fat ( $\Box$ ). SEM = 0.66 mm.

heat, the melted diameter of the cheese disc increased as the amount of proteolysis increased during storage.

AV. Fat percentage had a small interaction effect that decreased with age, but age had a large impact on the AV of the cheese (Table 4). During refrigerated storage, the AV of the cheese significantly decreased at all fat percentages (Figure 5), but the difference in AV between cheeses with different fat percentages decreased as indicated by the significant fat by age interaction (Table 4). Fife et al. (10) and Merrill et al. (20) also found that AV of low fat Mozzarella cheese decreased with time of refrigerated storage.

**FO.** Fat percentage had a large effect on the FO released by the cheese (Table 4; Figure 6). As the fat content of the cheese increased, the amount of FO release, expressed as a percentage of the fat in the cheese, significantly increased. The two lower fat cheeses (5 and 10% fat) expressed virtually no FO, but the highest fat cheese (25% fat) released about 40% of its total fat content (Figure 6).

The FO test is important because an excess of FO on the surface of a pizza after baking is a major quality defect for Mozzarella cheese manufacturers (15). However, Rudan and Barbano (25) demonstrated that lack of FO release is the critical event that limits shred melting and allows scorching of fatfree and lower fat Mozzarella cheeses. The importance of FO release during pizza baking is further discussed.

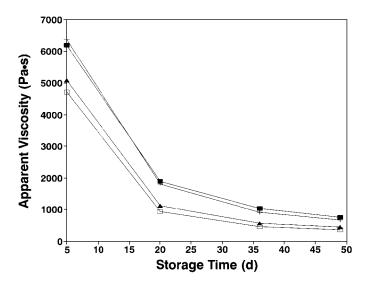


Figure 5. The effect of fat percentage on apparent viscosity during storage at 4°C for target levels of 5% fat ( $\blacksquare$ ), 10% fat (+), 15% fat ( $\blacktriangle$ ), and 25% fat ( $\square$ ). SEM = 230 Pa·s.

Journal of Dairy Science Vol. 82, No. 4, 1999

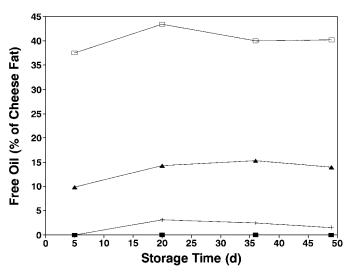


Figure 6. The effect of fat percentage on free oil release, as a percentage of cheese fat, during storage at 4°C for target levels of 5% fat ( $\blacksquare$ ), 10% fat (+), 15% fat ( $\blacktriangle$ ), and 25% fat ( $\square$ ). SEM = 1.1%.

**Pizza baking.** The pizza baking test is important for two reasons: 1) of the 1.02 billion kg (2.25 billion lb) of Mozzarella cheese produced in the US in 1996 (23), a high percentage is used for pizza and 2) the results of the modified Schreiber disc-type or test tube-type melt test do not seem to correlate well with the results of the pizza baking (26) or cooking (10) tests.

The cheese with a 5% fat content had limited melting and fusing of the shreds with a high degree of scorching of individual shreds and no blister formation, giving the pizza an atypical burnt appearance as seen in Figure 7a. The two higher fat cheeses (15 and 25% fat) displayed complete melting and fusing of the shreds and browning and blistering of the melted cheese as seen in Figure 7 (c and d, respectively). The 10% fat cheese (Figure 7b) had more melting and fusing of the shreds than did the 5% fat cheese but did not brown or blister as did the two highest fat cheeses. These results indicated that given the pizza baking conditions, cheese storage time, and manufacturing procedure used in the present study, it appeared that the minimum amount of fat required for Mozzarella cheese to function properly during pizza baking was between 10 and 15% fat. Furthermore, because of the importance of FO release for proper melting and browning of Mozzarella cheese (25), it appeared that the minimum amount necessary, given the pizza baking conditions, cheese storage time, and FO determination methodology (expressed as grams of FO released per 100 g of cheese) used in the

Figure 7. Pizza baking functionality after 30 d of storage at  $4^{\circ}$ C for target levels of 5% fat (a), 10% fat (b), 15% fat (c), and 25% fat (d).

present study, was between 0.22 and 2.52 g for the 10% fat cheese and 15% fat cheese, respectively. Not surprisingly, in an earlier study (25), we found that the amount of a hydrophobic surface coating (a simulated FO release) needed to achieve proper melting and browning of a fat-free cheese was in this range at 0.84 g of fat/100 g of cheese. Therefore, the results of the present study correlate well with the results from our earlier study (25).

# Fat and Nitrogen Recoveries

The mean total fat recoveries were 98.27, 99.66, 99.80, and 98.30%, and the mean total nitrogen recoveries were 101.57, 101.60, 101.62, and 102.66%, respectively, for the 5, 10, 15, and 25% target fat percentages. Because the total recoveries for fat and total nitrogen did not differ (P > 0.05) among treatments, the data were adjusted (27) to a mean value of 100% recovery as an average for all treatments. Fat content significantly affected the percentage of fat recovery, but not the percentage of nitrogen recovery, in the cheese and whey (Table 5). The lowest and highest fat cheeses (5 and 25% fat, respectively) retained a lower percentage of total fat in the cheese and, therefore, lost a higher percentage of total fat to whey than did the medium fat cheeses (10 and 15% fat). The percentage of total fat loss in the stretching water decreased as the fat content of the cheese decreased.

The target fat recovery in the cheese for both theoretical Mozzarella cheese yield formulas is 85%, which is lower than the 93% commonly used for Cheddar cheese. Higher fat loss during the manufacture of Mozzarella is expected for two reasons. First, the temperature of the milk at clotting and cutting is higher for Mozzarella (35 to  $38.3^{\circ}$ C) than for Cheddar (30 to  $31^{\circ}$ C) and favors greater fat loss in the whey at draining for Mozzarella cheese. Second, Mozzarella has a stretching step during which time significant amounts of fat can be lost in the stretching water.

Fat loss in the whey at draining appeared to be influenced by two separate factors. At a low ratio of casein to fat (i.e., high target cheese fat), the rennet curd matrix at cutting may reach a maximum fatholding capacity for the average curd size and temperature, possibly because of the interruption of the casein matrix by fat (11) above which the percentage of fat lost to the whey increases. At a high ratio of casein to fat (i.e., low target cheese fat), the rennet curd matrix at cutting and during cooking was more brittle because of the low fat content. During cheese manufacture, there was more curd shattering and fines as the ratio of casein to fat increased, which caused a higher percentage of fat loss to whey. Finally, although the percentage of fat recovery in the cheese for the lowest and highest fat cheeses did not differ (Table 5), the total weight of fat lost was less for the low fat cheese than for the higher fat cheese because the total amount of fat available in the milk was much lower for the low fat cheese. Therefore, direct comparisons of percentage fat recovery for cheeses with dramatically different fat concentrations should be done with caution. Control of fat recovery becomes more important as the fat content of the cheese increases.

Fat loss in the stretching water increased as the fat content of the cheese increased under the stretching conditions used in this study. The same screw speed (12 rpm) was used for all fat percentages. Experience gained in cheese manufacturing trials that were conducted after the present study indicated that the use of progressively higher screw speeds with higher fat cheeses (i.e., softer) would decrease fat loss in the stretching water and increase total fat recovery in the cheese.

# Yield

The actual and theoretical yields significantly decreased as the fat content in the cheese decreased.

TABLE 5. Mean adjusted fat and nitrogen recovery in the cheese, whey, and stretching water and actual and theoretical [Barbano (4) and Van Slyke and Price (33) as modified by Barbano (4)] cheese yields and efficiencies for cheeses made with four different fat percentages.

Recovery and yield	Target fat <sup>1</sup>					
	5%	10%	15%	25%	SEM	LSD <sup>2</sup>
Fat recovery, %						
Cheese	78.59 <sup>b</sup>	84.13 <sup>a</sup>	84.06 <sup>a</sup>	77.55 <sup>b</sup>	1.07	3.43
Whey	17.62 <sup>a</sup>	13.13 <sup>bc</sup>	12.00 <sup>c</sup>	14.83 <sup>b</sup>	0.62	1.99
Stretching water	3.13 <sup>c</sup>	3.52 <sup>bc</sup>	4.82 <sup>b</sup>	6.62 <sup>a</sup>	0.44	1.42
Nitrogen recovery, %						
Cheese	73.79	73.97	73.92	74.24	0.41	1.31
Whey	25.62	25.48	25.45	25.62	0.13	0.42
Stretching water	0.40 <sup>b</sup>	0.40 <sup>b</sup>	0.49 <sup>ab</sup>	0.66 <sup>a</sup>	0.05	0.16
Yield, kg/100 kg						
Actual	6.59 <sup>d</sup>	7.13 <sup>c</sup>	8.03 <sup>b</sup>	9.20 <sup>a</sup>	0.02	0.25
Van Slyke <sup>3</sup>	6.28 <sup>d</sup>	6.87 <sup>c</sup>	7.96 <sup>b</sup>	<b>9.87</b> <sup>a</sup>	0.04	0.14
Barbano <sup>4</sup>	6.59 <sup>d</sup>	7.13 <sup>c</sup>	8.03 <sup>b</sup>	9.20 <sup>a</sup>	0.03	0.27
Efficiency, %						
Van Slyke	105.0 <sup>a</sup>	103.9 <sup>a</sup>	101.0 <sup>b</sup>	93.2 <sup>c</sup>	1.40	1.9
Barbano	100.0	100.0	100.0	100.1	0.46	1.1

<sup>a,b,c,d</sup>Means within same row with no common superscript differ (P < 0.05). Rows without superscripts indicate no differences (P > 0.05) between means.

<sup>1</sup>Target cheese fat on a cheese weight basis (n = 4).

<sup>2</sup>Least significant difference (P = 0.05).

<sup>3</sup>Van Slyke and Price as modified by Barbano theoretical yield formula: yield (kilograms/100 kg of milk = [(( $0.85 \times milk$  fat percentage) + (milk casein percentage - 0.1)) × 1.13]/1 - (cheese moisture/100).

<sup>4</sup>Barbano theoretical yield formula: yield (kilograms/100 kg of milk) = (A + B + C)/[1 - ((cheese moisture + salt)/100)] where A = percentage of fat recovery in cheese × percentage of milk fat, B = (percentage of milk CN - 0.1) × calcium phosphate retention factor, and C = [((A + B)/(1 - cheese moisture/100)) - (A + B)) × (percentage of separated whey solids/100)] × solute exclusion factor.

The actual yields exceeded the modified Van Slyke and Price formula theoretical cheese yield (efficiencies greater than 100%) for the 5, 10, and 15% fat target levels. At the highest fat content, the actual yield was lower than the estimate by the modified Van Slyke and Price formula (efficiency equal to 93.2%). The modified Van Slyke and Price formula underestimates yield potential by an increasing amount as fat content decreases and moisture content increases, because it does not account for the increasing retention of nonfat, noncasein, milk solids retained in the cheese. As expected, the Barbano theoretical yield formula, using the coefficients defined previously, accurately predicted (efficiencies equal or close to 100) the actual cheese yield at all fat percentages in the cheese. The use of the actual moisture and salt content of the cheese and a solute exclusion factor optimized (from the data in the present study) for the moisture content of cheese allowed the Barbano theoretical yield formula to provide accurate predictions over a wide range of cheese fat and moisture percentages. When making lower fat cheese, differences in fat recovery become less important, but differences in both the amount and the concentration of solutes in the aqueous phase of the cheese become more important because they have a larger impact on cheese yield.

Milk fat, one of the major components in milk, is trapped in the casein matrix during cheese making. Therefore, it was not surprising that the actual yield significantly decreased as the fat content of the cheese was reduced, and moisture was not increased enough to maintain a constant total filler volume. This decrease in yield has significant economic implications. The cheese made with a target of 5% fat had approximately 30% less cheese per 100 kg of milk in the vat. Thus, fixed costs and costs that are a direct function of the weight of milk in the vat and independent of the amount of cheese produced would be substantially higher per kilogram of low fat cheese. Even accounting for the value of the fat removed from the milk prior to cheese manufacture, the economics of low fat cheese might not be as favorable as that of high fat cheese unless the product can be sold at a premium price. A possible strategy to avoid the increase in fixed manufacturing costs per unit weight of cheese would be to increase the casein content of the

milk by fortification (to increase the yield per unit volume) but to maintain the casein to fat ratio to produce a cheese with a fat content of 5%.

#### CONCLUSIONS

When the fat content of Mozzarella cheese was reduced below 15%, the MNFS, the ratio of moisture to protein, and the amount of proteolysis during refrigerated storage all decreased. The hardness of unmelted cheese increased significantly as the fat content and total filler volume decreased. The characteristics of unmelted cheese were consistent with those predicted by the filled gel model. Whiteness of the unmelted cheese decreased as fat content decreased. With respect to baking characteristics, the minimum amount of FO release necessary to obtain proper functionality during pizza baking was between 0.22 and 2.52 g of fat/100 g of cheese. As a guide to the cheese manufacturer, these results suggest that they may enhance pizza baking performance by manipulating the cheese manufacturing process or cheese product formulation to increase the FO release of their lower fat Mozzarella cheese. Cheese yield for low fat Mozzarella was about 30% lower for cheese with a 5% fat content than for cheese containing 25% fat. Theoretical cheese yield predictions using the Barbano theoretical yield formula predicted yield over a wide range of cheese compositions, but the modified formula of Van Slyke and Price was limited in its ability to predict yield over a wide range of cheese composition. Maximizing fat recovery in cheese becomes less important for maintaining high cheese yield efficiency as fat content of the cheese is reduced as the moisture control and the retention of solids in the water phase of the cheese become more important.

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