

Effect of Fat Replacer (Salatrim®) on Chemical Composition, Proteolysis, Functionality, Appearance, and Yield of Reduced Fat Mozzarella Cheese¹

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ABSTRACT

A no brine, stirred-curd procedure was used to manufacture reduced fat (9% fat wet basis) Mozzarella cheese. Skim milk was standardized to 0.8% fat with unhomogenized milk fat (control), an equal blend of fat replacer Salatrim® types 1 and 3 (Pfizer, Inc., Milwaukee, WI) (solid at room temperature), and 100% Salatrim® type 3 (liquid at room temperature). A stable dispersion (20% fat, wt/wt) was made by homogenizing Salatrim® in skim milk. Cheese making was repeated on each of 3 d using a randomized complete block design. All cheeses had similar pH, salt contents, and calcium contents; cheese made with Salatrim® had higher moisture and fat, but lower protein contents. Nitrogen that was soluble at pH 4.6 was higher for Salatrim® cheeses and increased for all cheeses during refrigerated storage. The meltability and apparent viscosity of all cheeses were similar, but the control had a significantly higher score for hardness and more free oil release, and cheese shreds scorched less during pizza baking. Hunter L, a, and b values of the unmelted cheese indicated that the Salatrim® cheeses were whiter and less yellow than the control, and all cheese decreased in whiteness over time. Salatrim® cheeses had significantly lower fat losses in the whey and stretching water and had higher actual and moisture-adjusted yields. Homogenization was probably responsible for the differences between the control and Salatrim® cheeses in chemical composition, proteolysis, functionality, appearance, and yield. Despite the large differences in fat properties, Salatrim® was probably

responsible only for the lack of yellowness in cheese color.

(**Key words:** reduced fat, Mozzarella cheese, fat replacer, functionality)

Abbreviation key: AV = apparent viscosity, FO = free oil, M50 = equal blend of Salatrim® 1 and 3, M100 = Salatrim 3, MNFS = moisture in the nonfat substance, TPA = texture profile analysis, VMD = volume mean diameter.

INTRODUCTION

Previous work (21) has indicated that homogenization of milk or addition of separately homogenized cream to skim milk significantly increased the whiteness of unmelted, reduced fat Mozzarella cheese. Homogenization of milk or cream did not affect parameters of texture profile analysis (TPA), meltability as measured by the Schreiber melt test, or apparent viscosity (AV), but decreased the amount of free oil (FO) release, reduced melting, and increased scorching of the reduced fat cheese (ca. 9%) during pizza baking (21). Therefore, more work is needed to improve the functionality of reduced fat Mozzarella cheese, especially during pizza baking. Fat replacers have been used by investigators to replace some of the functionality lost because of the removal of fat in low fat foods, including cheese (18).

Water-soluble fat replacers based on microparticulated carbohydrates and microparticulated protein (fat mimetics) have been used to manufacture low fat Mozzarella cheese (18). McMahon et al. (18) investigated the effects of commercially available fat mimetics that are based on carbohydrates (Stellar™ and Novagel™) and proteins (Simplese® and Dairy-Lo®) on the composition and functionality of low fat Mozzarella cheese (4 to 5% fat). All of the fat mimetics used by McMahon et al. (18), significantly increased the moisture content of the cheese; however, there was no difference in functionality compared with that of the control (no fat replacer added) after 28 d of refrigerated storage at 4°C. Based upon that study

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(18), fat mimetics based on carbohydrate or protein appear to have limited or no effect on improving the functionality of low fat Mozzarella cheese. Limitations are due to their molecular structure and characteristics. Under various conditions, especially heating, materials based on carbohydrate and protein do not behave like materials based on fat. This difference in behavior could be a major drawback for fat mimetics because Mozzarella cheese is consumed mainly in a heated, melted state on pizza.

According to Giese (7), fat substitutes are triglycerides with tailored fatty acid composition and stereospecific configurations (e.g., Salatrim[®]) or synthetic molecules that have chemical structures similar to those of triglycerides (e.g., Olestra[®]). Salatrim[®] (an acronym derived from short- and long-chain acyltriglyceride molecules) is an example of a tailored fatty acid triglyceride used as a fat substitute. Salatrim[®] achieves a calorie reduction based on two principles: 1) short-chain fatty acids (e.g., butyric) provide fewer calories per unit of weight than do longer chain fatty acids, and 2) stearic acid (the primary long-chain fatty acid of Salatrim[®]) is only partially absorbed by the body. The net result is a triglyceride that has all of the physical properties of fat, but that contains only 5 cal/g instead of 9 cal/g for naturally occurring fat (14). The FDA has proposed to amend its food labeling regulation such that the total amount of fat declared on the label for a product containing Salatrim[®] as the only fat source would be 5/9 of the total amount of fat of a traditionally made product (6).

Salatrim[®] was designed to replace fat in a wide range of applications, such as confectionery, peanut spreads, and dairy products, including cheese (14). However, there have been no published reports on the use of Salatrim[®] in reduced fat Mozzarella cheese. Therefore, the objective of this study was to determine the effect of replacing milk fat with two Salatrim[®] products (with melting properties different from each other and from milk fat) on the chemical composition, proteolysis, functional properties, appearance, and yield of reduced fat Mozzarella cheese.

MATERIALS AND METHODS

Fat Standardization and Cheese Manufacture

Raw skim milk and raw unhomogenized cream (40% milk fat) were obtained from the Cornell University dairy plant (Ithaca, NY). The fat contents of the skim milk [(17); method number 15.8.B.] and cream were determined [(1); method number 33.3.18, 995.18]. The control milk (ca. 250 kg) was standardized to about 0.8% fat by the addition of the raw,

unhomogenized cream to skim milk; the milk was then HTST pasteurized (Model Universal Pilot Plant; Processing Machinery and Supply Co., Philadelphia, PA) at 72°C for 16 s.

Three fats were compared: milk fat, a blend of 50% Salatrim[®] 1 and Salatrim[®] 3 (**M50**), and 100% Salatrim[®] 3 (**M100**). Salatrim[®] 1 (a blend of monobutyric and dibutyric acyltriglycerides in combination with stearic acid; solid at room temperature) and Salatrim[®] 3 (predominantly dibutyric, monostearic acyltriglyceride; liquid at room temperature) were obtained from Pfizer, Inc. (Milwaukee, WI). Penetrometry indicated that at 10°C the M50 and milk fat had similar hardness, and the M100 was much softer than the M50 or milk fat. Differential scanning calorimetry and dilatometry indicated that all fats were completely melted at temperatures above 40°C. The Salatrim[®] fats were virtually colorless, and the milk fat was yellow in appearance.

To prepare the Salatrim[®] fats for cheese manufacture, two 20% fat (wt/wt) Salatrim[®] creams were prepared. Either M50 or the M100 Salatrim[®] fat blend was mixed with raw skim milk, batch pasteurized (65.5°C for 30 min), and then homogenized at 63°C using first- and second-stage pressures of 13.8 MPa (2000 psi) and 3.45 MPa (500 psi), respectively, to obtain a stable dispersion. The Salatrim[®] creams were batch pasteurized prior to homogenization to prevent lipid hydrolysis. Following standardization of skim milk to about 0.8% fat by the addition of M50 and M100 creams, the M50 and M100 milks (ca. 250 kg) were HTST pasteurized as just described. All milks were cooled to 4°C and stored overnight at 4°C until used for cheese manufacture the next day. The two Salatrim[®] treated mixtures were homogenized separately as a 20% fat cream, instead of homogenizing the entire standardized milk, to minimize the amount of milk protein adsorbed on the surface of fat globules, which reduces curd shattering during cheese manufacture (21).

To produce a homogeneous chemical composition, cheese was made using the no brine, stirred-curd method (3).

Chemical Analyses

During cheese manufacture, the titratable acidity and pH of the milk and whey and the pH of the cheese were determined as previously described (29). The fat contents of the milk, whey, and stretching water were determined by ether extraction [(1); method number 33.2.26, 989.05]. The fat content of the cheese was determined by Babcock [(17); method number 15.8.2.d]. All samples were tested for total nitrogen by Kjeldahl [(1); method number 33.2.11, 991.20]. Non-casein nitrogen of the milk was determined by the

International Dairy Federation procedure (8) and nonprotein nitrogen by Kjeldahl [(1); method number 33.2.12, 991.21]. Total nitrogen and noncasein nitrogen for milk were assayed in triplicate, cheese moisture and fat in quadruplicate, and all others in duplicate.

Whey solids were determined gravimetrically by drying approximately 3 g of whey at 100°C in a forced-air oven (model OV-490A-2; Blue M, Blue Island, IL) for 4 h. Cheese moisture, salt and calcium concentration (11), and pH were determined as previously described (29).

Soluble protein (pH 4.6 and 12% TCA) was determined after 2, 9, 16, 30, and 44 d of storage at 4°C, as previously described (5).

Functional Properties of Unmelted and Melted Cheese

The TPA, as described by Bourne (4), was performed on the unmelted cheese using an Instron Universal Testing Machine (model TM; Instron Corp., Canton, MA) after 30 d of storage at 4°C. A modified Schreiber test (13) was used to quantify cheese meltability and was determined after 2, 9, 16, 30, and 44 d of storage at 4°C. The AV was determined on the melted cheese using helical viscometry as described by Kindstedt and Kiely (10), and FO was determined on the melted cheese using a centrifugation method described by Kindstedt and Rippe (12). Both AV and FO were determined after 6, 16, 30, and 44 d of storage at 4°C. The pizza bake test was used to evaluate the functionality (shred melting and browning) of the cheese as a pizza topping after 30 d of storage at 4°C (20). All tests were performed using the same procedures as previously described (30).

Milk Fat Particle Size and Cheese Appearance

Milk fat globule size was determined using a Malvern Mastersizer E particle size analyzer (model E; Malvern Instruments, Worcestershire, UK) as previously described (24). The volume mean diameter (**VMD**), the mean volume to surface diameter (Sauter mean), and the mean diameter below which 90% of all fat volume is contained ($d_{0.9}$) were calculated.

The appearance of the milk and cheese was quantitatively determined using a Macbeth Color-Eye Spectrophotometer (model 2020; Kollmorgen Instruments Corp., Newburgh, NY) as previously described (21). The color of the cheese was measured in quadruplicate after 2, 9, 16, 30, and 44 d of storage at 4°C.

Recoveries and Yield Calculations

The percentages of actual fat and N recoveries in the cheese, whey, and stretching water and actual cheese yield were calculated as previously described (15). Yield adjusted for moisture and salt was calculated using values of 53 and 1.6% for the moisture and salt contents, respectively, as described by Lau et al. (15). Theoretical yield was calculated using a modification of the formula for cheese yield of Van Slyke; the original formula, based on Cheddar cheese yield, was modified to reflect Mozzarella cheese yield (2). Modifications included changing the assumed fat recovery value from 0.93 to 0.85, changing the constant factor from 1.09 to 1.13 (2), and using 53% for the desired cheese moisture. A new method for calculating Mozzarella cheese yield was also used (2). This new formula also included the SNF content of the separated whey to estimate the amount of whey solids retained in the water phase of the cheese. In addition, values of 0.85, 1.092, and 53% were used in the new formula (2) for the fat retention, calcium phosphate retention, and desired cheese moisture content, respectively. The actual value for the salt content of the cheese and a solute exclusion factor value of 0.9306 were used as terms in the new formula.

Experimental Design and Statistical Analysis

On each of 3 d of cheese manufacture, the control, M50, and M100 cheeses were made. Changes in proteolysis, functional properties (melt, AV, and FO), and the appearance of unmelted cheese during refrigerated storage were analyzed using a split-plot design in which the whole plot factor (fat type) was replicated in a 3 × 3 randomized complete block design. For the whole-plot factor, treatment was analyzed as a classification variable, and day of cheese manufacture was blocked. For the subplot factor, age and interaction between ages were analyzed as quantitative variables. The degrees of freedom in the statistical model were the same for the soluble N, melt, and appearance. The degrees of freedom of the error term for the subplot factor error for the AV and FO results was 21 instead of 30 because four aging times were used instead of 5. The PROC GLM procedure of SAS (23) was used for all data analyses.

RESULTS

Composition of Milk, Cheese, Whey, and Stretching Water

The composition of the milks used for cheese manufacture is shown in Table 1. The pH and protein

contents of the standardized milks were not different. The fat contents of the M50 and M100 milks were slightly higher than that of the control, which was probably due to the evaporative concentration during the batch pasteurization of the cream. As expected, homogenization of the 20% fat Salatrim[®] creams significantly reduced the particle size of the fat globules. The VMD, Sauter mean, and d0.9 were significantly larger for the unhomogenized control than for the homogenized Salatrim[®] M50 and M100 milks. The particle size for the M50 and M100 milks was not as small as would be expected for homogenized milk because the homogenization of cream produces clusters (21). Clustering was also observed in a previous study (17) when 20% cream made with milk fat was homogenized. The particle size results are based on an analysis without a dissociating agent to disrupt the clusters prior to particle size measurement.

As expected, homogenization of the Salatrim[®] cream caused the whiteness (i.e., Hunter L value) of the M50 and M100 milks to be greater than that of the unhomogenized control milk (Table 1). There were small differences in the Hunter a and b values among the control, M50, and M100 treatments. The control was slightly greener (greater negative Hunter a value) than the M50 and M100 milks, and the yellowness was almost the same for all three milks (Table 1).

The composition of the cheeses is shown in Table 2; the M50 and M100 cheeses were very similar. Because the fat contents of the M50 and M100

Salatrim[®] milks were slightly higher than that of the control (Table 1), the fat content of the Salatrim[®] cheeses tended to be slightly higher than that of the control, although only the M50 cheese was significantly higher in fat than the control. Compared with the control cheese, the cheeses made with Salatrim[®] had significantly higher moisture contents, moisture in the nonfat substance (MNFS), ratio of moisture to protein, and fat on a dry basis and significantly lower protein contents. The pH, salt, salt in the moisture phase, calcium, and calcium as a percentage of protein contents of all of the cheeses did not differ significantly.

The composition of the whey and stretching water is shown in Table 3. No difference was detected in the protein content of the whey and stretching water for any cheese. The control had a significantly higher concentration of fat in the whey and stretching water than did the cheeses made with Salatrim[®].

Proteolysis

The effects of treatment and time of refrigerated storage on indices of proteolysis based on pH 4.6-soluble N or 12% TCA-soluble N are shown in Table 4. Treatment had a significant effect on the pH 4.6-soluble N content but not on the 12% TCA-soluble N content, even though the same trends in the data were apparent (Figures 1 and 2). Age, as expected, also had an effect on both the pH 4.6-soluble N and 12% TCA-soluble N contents of the cheeses. The Salatrim[®] treatments contained more pH 4.6-soluble N, and the difference, compared with the

TABLE 1. Mean (n = 3) composition, fat particle size, and appearance of milk made from skim milk standardized with unhomogenized milk fat (control), skim milk standardized with a 50:50 blend of Salatrim[®] types 1 and 3 triglycerides (M50), and skim milk standardized with 100% Salatrim[®] type 3 triglycerides (M100).

Component	Treatment			SEM	LSD ²
	Control	M50	M100		
pH	6.64	6.63	6.64	0.01	0.05
Fat, %	0.82 ^b	0.83 ^{ab}	0.85 ^a	0.01	0.02
Protein, %	3.07	3.08	3.08	0.01	0.03
Casein, %	2.34	2.36	2.37	0.01	0.03
d43, ³ μm	3.12 ^a	1.70 ^b	1.77 ^b	0.11	0.44
d32, ⁴ μm	0.80 ^a	0.59 ^b	0.61 ^b	0.01	0.02
d0.9, ⁵ μm	6.05 ^a	3.69 ^b	3.42 ^b	0.14	0.53
Hunter L value ⁶	77.15 ^b	78.40 ^a	78.20 ^a	0.09	0.37
Hunter a value	-5.65 ^c	-5.25 ^a	-5.35 ^b	0.02	0.09
Hunter b value	1.66 ^b	1.77 ^a	1.72 ^{ab}	0.02	0.08

a,b,c Means within the same row without a common superscript differ ($P < 0.05$).

¹Pfizer, Inc. (Milwaukee, WI).

² $P = 0.05$.

³Volume mean diameter of fat particles.

⁴Sauter mean diameter of fat particles.

⁵Fat particle diameter below which 90% of fat volume is contained.

⁶Hunter values: L = whiteness, a = greenness, and b = yellowness.

TABLE 2. Mean (n = 3) values for the initial composition of reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control), skim milk standardized with a 50:50 blend of Salatrim®¹ types 1 and 3 triglycerides (M50), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100) at 1 d of storage at 4°C.

Component	Treatment			SEM	LSD ²
	Control	M50	M100		
pH	5.20	5.19	5.22	0.02	0.07
Moisture, %	52.81 ^b	54.22 ^a	54.35 ^a	0.25	0.98
Fat, %	9.22 ^b	9.61 ^a	9.42 ^{ab}	0.06	0.25
FDB, ³ %	19.55 ^b	21.00 ^a	20.63 ^a	0.25	0.96
Protein, %	31.10 ^a	29.46 ^b	29.39 ^b	0.27	1.05
M:P ⁴	1.70 ^b	1.84 ^a	1.85 ^a	0.03	0.10
MNFS, ⁵ %	58.18 ^b	59.99 ^a	60.00 ^a	0.31	1.23
Salt, %	1.54	1.57	1.52	0.07	0.29
S in M, ⁶ %	2.92	2.89	2.79	0.13	0.50
Ca, %	0.93	0.89	0.90	0.01	0.06
Ca, % of Protein	3.00	3.02	3.06	0.03	0.10

^{a,b}Means within the same row without a common superscript differ ($P < 0.05$).

¹Pfizer, Inc. (Milwaukee, WI).

² $P = 0.05$.

³Fat content on a dry weight basis.

⁴Ratio of moisture to protein.

⁵Moisture in the nonfat substance of the cheese.

⁶Salt in the moisture phase of the cheese.

control, increased during refrigerated storage (Figure 1). Although the cheeses did not differ significantly in content of 12% TCA-soluble N (Table 4), the cheese containing Salatrim® tended ($P = 0.10$) to contain more 12% TCA-soluble N than did the control, and this relationship was maintained over 44 d of storage at 4°C (Figure 2).

Functional Properties

Unmelted cheese. The TPA hardness of the M50 and M100 cheeses was significantly less than that of the control (Table 5). The TPA springiness tended to be lower for the M50 and M100 cheeses; however, no

significant differences in the TPA springiness or cohesiveness among cheeses were detected.

Melted cheese. There was a significant interaction of treatment and age (Table 6), indicating that meltability (measured using the Schreiber melt test) was influenced by fat type, but the mean square for this interaction was a small portion of the total mean square for the model. Any trend in the data is difficult to determine (Figure 3). Furthermore, factors such as differences in moisture contents could also have had an influence on cheese meltability. The use of Salatrim® did not have a detectable influence on AV, but age had a large impact on the AV (Table 6). The AV significantly decreased from about 5000 Pa·s at 6

TABLE 3. Mean (n = 3) composition of whey and stretching water from reduced fat Mozzarella cheeses made with skim milk standardized with unhomogenized milk fat (control), skim milk standardized with a 50:50 blend of Salatrim®¹ types 1 and 3 triglycerides (M50), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100).

Component	Treatment			SEM	LSD ²
	Control	M50	M100		
Whey					
Fat, %	0.09 ^a	0.05 ^b	0.05 ^b	0.00	0.01
Protein, %	0.88	0.87	0.87	0.00	0.01
Total solids, %	6.63	6.60	6.64	0.02	0.09
Stretching water					
Fat, %	0.18 ^a	0.10 ^b	0.08 ^b	0.02	0.06
Protein, %	0.08	0.10	0.09	0.00	0.02

^{a,b}Means within the same row without a common superscript differ ($P < 0.05$).

¹Pfizer, Inc. (Milwaukee, WI).

² $P = 0.05$.

TABLE 4. Mean squares, probabilities (in parentheses), and degrees of freedom for indices of proteolytic changes (as a percentage of total N) of reduced fat Mozzarella cheese during 44 d of storage at 4°C.

Factors	df	pH 4.6-Soluble N	12% TCA-Soluble N
Whole plot			
Treatment (T)	2	11.82* (<0.01)	0.884 (0.10)
Day of cheese manufacture (blocked)	2	2.60 (0.05)	0.237 (0.40)
Error	4	0.38	0.20
Subplot			
Age (A)	1	397.57* (<0.01)	107.81* (<0.01)
A × A	1	10.47* (<0.01)	0.12 (0.25)
T × A	2	1.59* (0.01)	0.13 (0.25)
T × (A × A)	2	1.73* (0.01)	0.16 (0.18)
Error	30	0.30	0.09
R ²		0.98	0.98

* $P < 0.05$.

d to about 500 Pa·s at 44 d, but there was no detectable effect of treatment on AV. The control cheese released significantly more FO as a percentage of fat in the cheese than did the M50 and M100 cheeses (Table 6). Overall, the total amount of FO released for all treatments increased as the time of storage at 4°C increased (Figure 4).

Functionality of the cheeses during the pizza bake test after 30 d of refrigerated storage at 4°C is shown

in Figure 5. All cheeses performed poorly, as indicated by the low degree of shred melt and fusion and by the high degree of browning and scorching. These defects were more severe for the two Salatrim® treatments than for the control treatment.

Appearance

The effect of Salatrim® on the appearance of the unmelted cheese (4°C) over 44 d of refrigerated storage at 4°C are shown in Table 7. Treatment had a significant effect on the Hunter L and b values; age had a significant effect on all values. The M50 and M100 cheeses were significantly whiter than the controls; the whiteness of all cheeses decreased during storage (Figure 6). The M50 and M100 cheeses lost less of their whiteness during storage than did the control (Figure 6; Table 7, interaction of treatment and age). The mean Hunter a value over 44 d of refrigerated storage at 4°C was 0.33, 0.00, and -0.18 for the control, M50, and M100 treatments, respectively, indicating that the color of these cheeses was neither green nor red. The Hunter b value for the control cheese was greater than that for the M50 and M100 cheeses, indicating that the control cheese was significantly yellower (Figure 7; Table 7). Visual observation confirmed these results, as the M50 and M100 cheeses appeared to be substantially whiter and less yellow than the control cheese (results not shown).

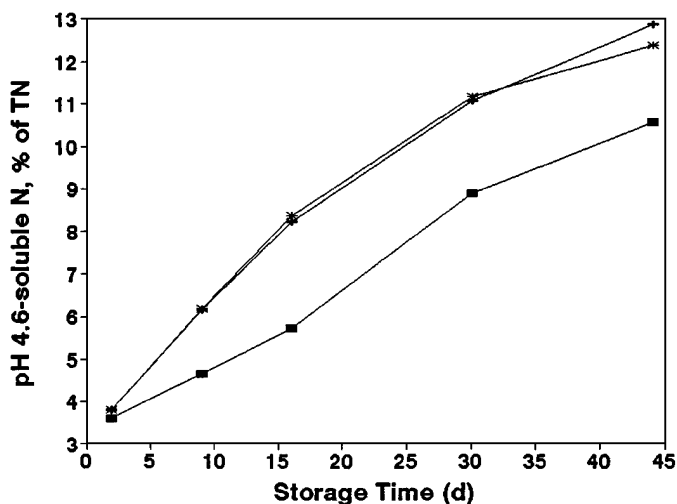


Figure 1. Effect of treatment on pH 4.6-soluble N, as a percentage of the total N (TN), in reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control; ■), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50; +), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100; *) during 44 d of storage at 4°C.

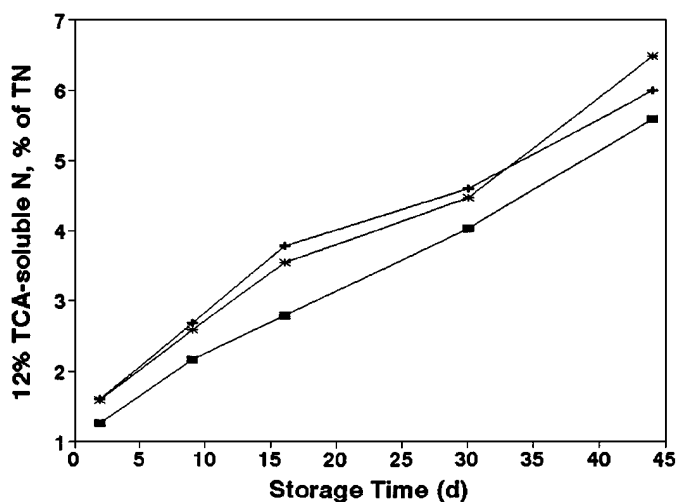


Figure 2. Effect of treatment on 12% TCA-soluble N, as a percentage of the total N (TN), in reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control; ■), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50; +), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100; *) during 44 d of storage at 4°C.

TABLE 5. Texture profile analysis (TPA) parameters measured at 10°C of reduced fat Mozzarella cheeses at 30 d of storage at 4°C.

TPA Parameter	Treatment ¹			SEM	LSD ²
	Control	M50	M100		
Hardness, N	76.51 ^a	49.51 ^b	56.84 ^b	3.24	10.37
Cohesiveness	0.63	0.52	0.59	0.06	0.19
Springiness, mm	5.65	4.14	4.14	0.55	1.76

^{a,b}Means (n = 3) within the same row without a common superscript differ ($P < 0.05$).

¹Reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100).

² $P = 0.05$.

Recoveries and Cheese Yield

The effects of treatment on fat, N recovery, and cheese yield are shown in Table 8. Because fat and protein are the major milk solids in Mozzarella cheese, it is important to account for their distribution in the cheese, whey, and stretching water. The mean values for actual total recoveries of fat were 96.4, 97.8, and 95.3% and N recovery was 100.4, 101.1, and 101.6% for the control, M50, and M100 treatments, respectively. The mean actual total fat recovery in this study was lower than that experienced by the investigators in previous studies. The measurement of the cheese fat (ca. 9% fat) for analysis of cream in Babcock bottles (graduated from 0 to 50% fat) may have led to a slight systematic underestimation of the fat content of the cheese in all of the treatments. There was no difference ($P > 0.05$) from treatment on the actual total recovery of fat or

N. Therefore, for each day of cheese manufacture, the fat and N recoveries were adjusted to the mean value for the actual total recovery for the three treatments. The adjusted values for fat recovery for the M50 and M100 cheeses were significantly higher than that of the control (Table 8). Consequently, the M50 and M100 treatments had significantly lower fat losses (about half) to the whey and stretching water than did the control. No significant differences were detected in adjusted values for N recovery between treatments for the cheese, whey, and stretching water.

The actual yields, yields adjusted for moisture and salt, Van Slyke theoretical yields, and Barbano theoretical yields for the M50 and M100 cheeses were significantly greater than those for the control cheese (Table 8). Because differences in cheese moisture

TABLE 6. Mean squares and probabilities (in parentheses) of the meltability, apparent viscosity, and free oil (as a percentage of fat) of reduced fat Mozzarella cheese during 44 d of storage at 4°C.

Factors	Meltability	Apparent viscosity ($\times 10^6$)	Free oil
Whole plot			
Treatment (T)	9.57 (0.18)	0.311 (0.48)	0.349* (<0.01)
Day of cheese manufacture (blocked)	1.26 (0.72)	0.65 (0.27)	0.00 (0.63)
Error	3.50	0.36	0.01
Subplot			
Age (A)	422.15* (<0.01)	97.18* (<0.01)	0.17* (0.01)
A \times A	5.54* (0.05)	29.70* (<0.01)	0.08 (0.07)
T \times A	11.25* (<0.01)	0.01 (0.97)	0.05 (0.15)
T \times (A \times A)	7.44* (0.01)	0.14 (0.67)	0.04 (0.17)
Error	1.38	0.34	0.02
R ²	0.92	0.95	0.76

* $P < 0.05$.

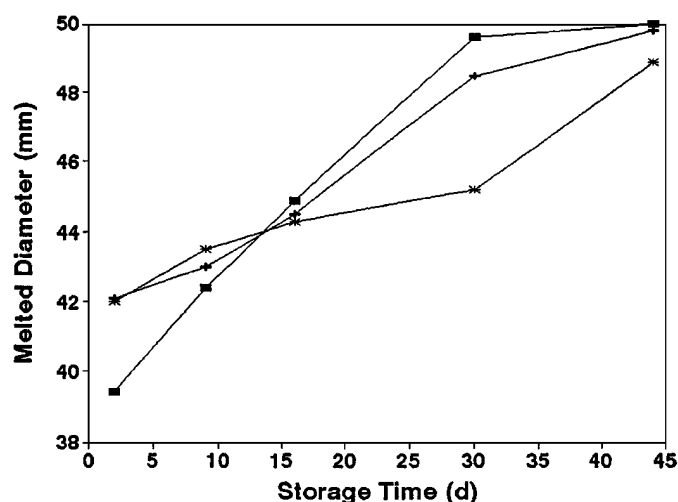


Figure 3. Effect of treatment on the meltability of reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control; ■), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50; +), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100; *) during 44 d of storage at 4°C.

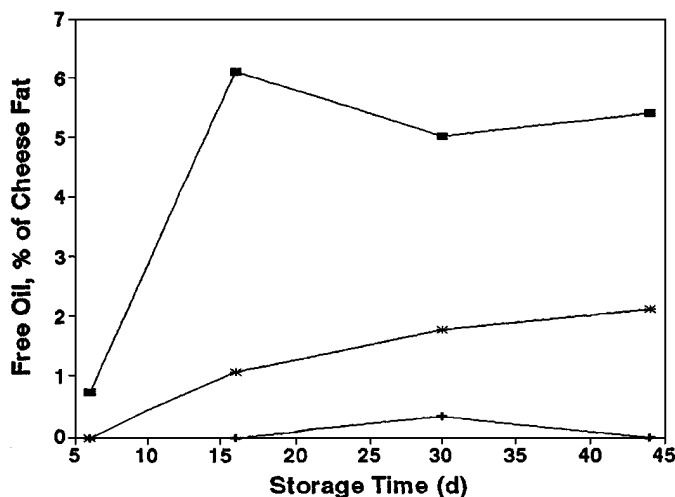


Figure 4. Effect of treatment on the release of free oil from reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control; ■), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50; +), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100; *) during 44 d of storage at 4°C.

and salt contents can distort the evaluation of the recovery of milk solids, yields that were adjusted for moisture and salt contents are calculated to compare yields among treatments. Theoretical cheese yields are calculated to give the cheese manufacturer an idea of the efficiency (adjusted yield divided by theoretical multiplied by 100) of their operation. The theoretical yields using modified equation of Van Slyke and Barbano consistently underestimated the

TABLE 7. Mean squares and probabilities (in parentheses) for Hunter¹ indices of color changes of reduced fat Mozzarella cheese during 44 d of storage at 4°C.

Factor	L Value	a Value	b Value
Whole plot			
Treatment (T)	152.38* (<0.01)	0.348 (0.08)	4.15* (<0.01)
Day of cheese manufacture (blocked)	3.44 (0.35)	2.23* (<0.01)	0.51 (0.18)
Error	2.50	0.07	0.19
Subplot			
Age (A)	132.37* (<0.01)	0.96* (<0.01)	1.17* (<0.01)
A × A	60.25* (<0.01)	0.21* (0.01)	0.34* (<0.01)
T × A	6.23* (<0.01)	0.01 (0.68)	0.15* (0.03)
T × (A × A)	1.82 (0.14)	0.00 (0.97)	0.02 (0.53)
Error	0.87	0.03	0.04
R ²	0.97	0.91	0.97

¹Hunter values: L = whiteness, a = greenness, and b = yellowness.

**P* < 0.05.



Figure 5. The appearance of a pizza topped with reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control; left), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50; lower right), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100; upper right) at 30 d of storage at 4°C.

adjusted yield, which resulted in efficiencies (adjusted yield divided by theoretical yield multiplied by 100) of greater than 100 for all cheeses, except for the control cheese, for which the Barbano formula accurately predicted the actual yield (efficiency equal to 100). The overprediction by the Barbano formula for the M50 and M100 cheeses was because the fat recovery exceeded the default value of 85% used in the formula.

DISCUSSION

Salatrim® is a product that is available as a solid or liquid fat at room temperature (ca. 23°C), depending on the fatty acid composition. It is not possible to obtain a stable dispersion of this fat in skim milk without homogenization. In the current study, the control was milk fat in its natural dispersion when unhomogenized. Thus, some differences in the characteristics of the cheeses were due to homogenization, and some may have been due to the differences between Salatrim® and milk fat. A previous study (21) documented the influence of homogenization on the characteristics of reduced fat Mozzarella cheese. Therefore, any influences of Salatrim®, separate from the effect of homogenization, should be detected when the results of the current study are compared with the results of the earlier study (21).

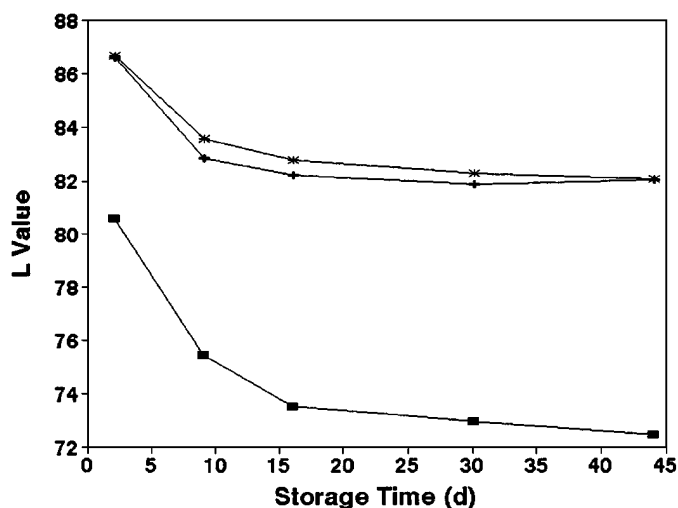


Figure 6. Effect of treatment on Hunter L value (whiteness) of reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control; ■), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50; +), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100; *) during 44 d of storage at 4°C.

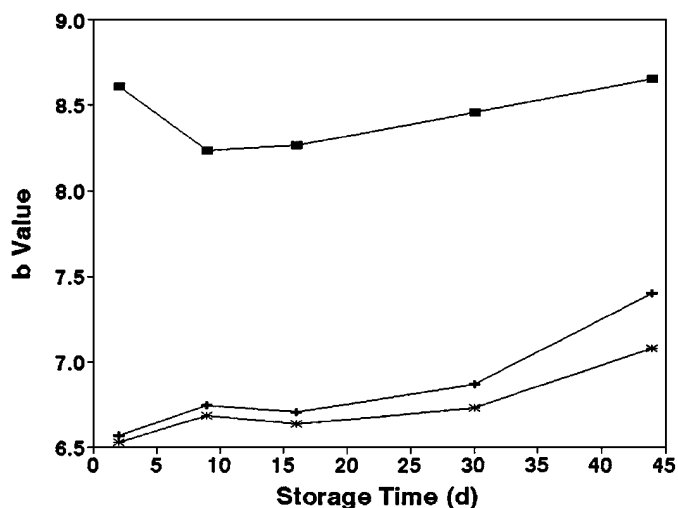


Figure 7. Effect of treatment on Hunter b value (yellowness) of the reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control; ■), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50; +), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100; *) during 44 d of storage at 4°C.

Cheese Composition

Fat type seemed to have an impact on the initial composition of the cheese (Table 2). However, this effect was probably due to homogenization rather than to fat type. Homogenization of the Salatrim® creams (20% fat) increased the moisture content, which decreased the protein content of the cheeses (Table 2) and increased fat retention (Table 8), which resulted in a higher FDB for the cheeses made using Salatrim®. Other researchers (9, 19, 27) have also reported an increase in cheese moisture contents from homogenization. These initial differences in chemical composition would be expected to influence the degree of proteolysis and the functional properties of the cheese.

The results of Tunick et al. (27) showed that the amount of proteolysis occurring in cheese might be directly related to MNFS levels. Because the ratio of moisture to protein and the MNFS of the Salatrim® cheeses were greater than those of the control cheese, more proteolysis would be expected to occur in the Salatrim® cheeses. Overall, this result occurred; the pH 4.6-soluble N content, as a percentage of total N, of the Salatrim® cheeses was significantly higher than that of the control cheese, and the 12% TCA-soluble N also tended to be higher (Figures 1 and 2).

Functional Properties of Unmelted Cheese

The TPA characteristics of the unmelted cheese were measured at 30 d of storage at 4°C. Thus, the

combined influence of higher moisture and fat contents (i.e., more filler), more proteolysis, smaller size of fat particle, possibly greater interaction between the fat and casein matrix, and the lower hardness value of the M100 versus milk fat and the M50 fat produces a net effect on TPA parameters. In a previous study (21), homogenization of milk or cream, which decreased the size of the fat particles and possibly increased the interactions of casein and fat, had no effect or very little effect on TPA parameters of reduced fat Mozzarella cheeses made from milk fat. As a result, Rudan et al. (21) concluded that cheese composition (i.e., total filler volume) and proteolysis appear to be more important than smaller fat particle size and possible interactions of fat and casein at 10°C.

The control cheese had significantly higher TPA hardness (measured at 10°C) than did either of the cheeses made using Salatrim®. The larger filler volume and greater proteolysis provided by the Salatrim® could have contributed to this result. This scenario is consistent with the model for the filled gel composite, which predicts that an increase in the filler volume fraction (moisture and fat in this case for the Salatrim® cheeses) results in a decrease in the amount of matrix (protein) to deform per unit volume; thus, less force is required for a given deformation, and the composite becomes softer (28). In addition, the greater proteolysis in the Salatrim® cheeses would be expected to decrease the amount of intact protein capable of contributing to the matrix,

thereby decreasing the force required to obtain a given deformation (28). Finally, the M50 Salatrim® fat had the same hardness as the milk fat at 10°C, yet the TPA hardness of the cheese made with M50 was significantly lower than the milk fat control. Furthermore, the M50 fat is significantly harder than the M100 fat at 10°C, but the M50 and M100 cheese had similar TPA hardness. These results would suggest that the fat type has no effect on the TPA hardness of cheese at 10°C. Thus, the effect of total filler volume and proteolysis on the TPA hardness of the cheese at 10°C is more important than fat hardness. Generally, unmelted low fat Mozzarella cheese is too hard and springy (22). The changes in cheese that were produced by homogenization lowered the TPA hardness and, thus, seemed to improve the characteristics of the unmelted cheese.

Melted Cheese Functional Properties

For functionality of the melted cheese, there was little if any effect of Salatrim® on meltability (as measured by the Schreiber melt test) or AV (measured by helical viscometry). Given the higher moisture content, lower protein content, and increased proteolysis, it is unclear why the M50 and M100 cheeses did not have significantly greater melt,

as determined by the Schreiber melt test, or lower AV than the control cheese.

Homogenization of the Salatrim® cream produced lower FO release than did the control. The decrease in fat particle size and the new fat globule membrane formed as a result of homogenizing the Salatrim® cream resulted in a cheese that released significantly less FO upon heating than did the cheese made from unhomogenized fat. This result is consistent with previous reports (9, 21, 26). Tunick (26) demonstrated that homogenization of the milk used to make Mozzarella cheese (ca. 25% fat) significantly decreased the amount of FO. The results of Tunick (26) suggested that the reduction in fat particle size and the subsequent changes in the cheese structure, not the exposure of casein to homogenization, were responsible for the reduction in FO release (26).

The differences in triglyceride composition between the Salatrim® fats and milk fat result in differences in the ratio of solid to liquid fat at various temperatures among fat types, which could have influenced the FO release. The fat with the lowest melting point (i.e., M100) would be expected to release the most FO. The cheese made with the fat with the lowest melting point, however, released virtually no FO, indicating that, of all the possible factors, homogenization seemed to have the largest impact on the release of FO.

TABLE 8. Fat and N recoveries adjusted to the mean value of the actual total recovery of the cheese, whey, and stretching water and the actual yield adjusted for moisture and salt, theoretical cheese yields, and cheese yield efficiencies.

Recovery, yield, and efficiency	Treatment ¹			SEM	LSD ²
	Control	M50	M100		
Fat recovery, %					
Cheese	85.34 ^b	93.46 ^a	91.01 ^a	1.16	3.71
Whey	10.43 ^a	5.75 ^b	5.93 ^b	0.23	0.75
Stretching water	4.12 ^a	2.17 ^b	1.79 ^b	0.26	0.84
N Recovery, %					
Cheese	73.09	74.14	74.73	0.41	NS
Whey	25.74	25.40	25.30	0.10	NS
Stretching water	0.50	0.56	0.54	0.02	NS
Yield, kg/100 kg					
Actual	7.30 ^b	7.82 ^a	7.91 ^a	0.07	0.23
Adjusted ³	7.34 ^b	7.61 ^a	7.69 ^a	0.05	0.17
Van Slyke	7.06 ^c	7.14 ^b	7.19 ^a	0.01	0.04
Barbano	7.38 ^c	7.46 ^b	7.52 ^a	0.02	0.05
Efficiency ⁴					
Van Slyke	104	107	107	0.62	NS
Barbano	99.4	102	102	0.64	NS

a,b,c Means (n = 3) within the same row without a common superscript differ ($P < 0.05$).

¹Reduced fat Mozzarella cheeses made from skim milk standardized with unhomogenized milk fat (control), skim milk standardized with a 50:50 blend of Salatrim® (Pfizer, Inc., Milwaukee, WI) types 1 and 3 triglycerides (M50), and skim milk standardized with 100% Salatrim® type 3 triglycerides (M100).

² $P = 0.05$.

³Moisture (53%) and salt (1.6%) adjusted yield.

⁴Adjusted/theoretical yield.

The homogenized Salatrim® cheeses had less shred melt and fusion and more browning and scorching than did the control cheese during pizza baking (Figure 5). This result is consistent with those from an earlier study in which reduced fat Mozzarella cheese made with homogenized milk fat had less shred melt and fusion and more excessive browning and scorching during pizza baking than did reduced fat Mozzarella cheese made from unhomogenized milk (21). Therefore, homogenization of the fat, not the type of fat in this study, apparently decreases pizza bake functionality. The reason for this result is unclear, and more work is needed to develop a reduced fat Mozzarella cheese that performs similar to regular fat Mozzarella cheese (low moisture part-skim) during pizza baking. Furthermore, because no differences caused by homogenization were detected in meltability by the Schreiber melt test and in AV (as measured by helical viscometry) among the cheeses, yet pizza bake meltability was different, the usefulness of these tests for reduced fat Mozzarella cheese is in question. These tests do not appear to reflect the meltability of the cheese on pizza.

Appearance

The largest effect of fat type was on the appearance of the milk and cheese. However, appearance was not only a result of fat type, but also of homogenization. Homogenization of the cream significantly changed the fat globule size in the milk used for cheese manufacture (Table 1), which resulted in a change in the fat dispersion of the cheese, as shown in a previous study (21). This change in structure manifested itself as a change in the appearance of the unmelted cheese. A reduction in the size of the fat particles (with a subsequent increase in number) made the milk and unmelted cheese whiter. These results are consistent with those of Lemay et al. (16), who hypothesized that decreasing the milk fat globule size and increasing the number of globules increased the light-scattering properties of the cheese, leading to an increase in whiteness. Because Hunter b values for cheeses made from homogenized cream and un-homogenized milk fat in a previous study (21) were similar (Hunter b values between 8 and 9 over 44 d of storage), the lower Hunter b values of the Salatrim® cheeses was probably caused by the difference in yellowness between Salatrim® and milk fat. Furthermore, this result may explain the slight increase in Hunter L value of the Salatrim® cheeses compared with that of the cheese made from homogenized milk fat in a previous study (21). Unfortunately, as in the earlier study (21), because the browning of melted cheese during pizza baking was so severe for the cheeses made with homogenized Salatrim® cream

(Figure 5), it is not clear whether the melted cheese was also whiter. Ultimately, increasing the whiteness of unmelted and melted reduced fat Mozzarella cheese would be desirable, and more work is needed in this area.

Recoveries and Yield

Homogenized Salatrim® cheeses had higher fat recoveries in the curd and, consequently, lower fat losses to the whey and stretching water, than did the control cheese. Metzger and Mistry (19) also observed decreased fat loss to the whey for reduced fat Cheddar cheese as a result of the separate homogenization of the cream, probably as a result of the change in the structure of the rennet curd matrix caused by homogenization. Furthermore, possible cross-linking of the fat membrane formed because of homogenization of the rennet curd with the casein matrix (27) may also be responsible for the lower fat loss to the whey. Fat can also be lost during stretching of the curd. Electron micrographs have shown that homogenization significantly reduces particle size and more evenly distributes the fat particles throughout the casein matrix of the curd, but unhomogenized milk fat globules tend to form relatively large clusters in the casein matrix during stretching (21). The smaller, more evenly dispersed particles of fat in the cheese made from homogenized cream may be retained in the cheese to a greater degree than the large fat clusters, accounting for the lower fat losses to the stretching water of the cheeses made from homogenized fat.

The increase in cheese yield for the homogenized Salatrim® treatments corresponded to the lower fat and protein losses to the whey. Although the moisture contents of homogenized Salatrim® cheeses was higher, the moisture-adjusted yields and theoretical yields (Van Slyke and Barbano, Table 8) still indicated a significant increase in cheese yield. Because even small differences in cheese yield can be very important economically, homogenization offers an advantage to the cheese manufacturer.

CONCLUSIONS

Reduced fat Mozzarella cheese was successfully made using Salatrim® in place of milk fat. Homogenization of the Salatrim® cream, rather than the actual properties of the Salatrim® fat, probably had the greatest influence on the composition, proteolysis, functional properties, and yield of the cheese. Chemical composition (i.e., total filler volume), proteolysis, or both seemed to have an impact on the functionality of the unmelted cheese. The largest effect of homogenization on the functionality of the melted

cheese seemed to be the inhibition of FO release. A previous study (22) in which Mozzarella cheeses were made at four different fat contents also showed a progressive decrease in FO release as the fat content decreased, but we assumed that differences in FO release had no influence on baking properties. However, decreased FO release now appears to be accompanied by a decrease in meltability and an increase in scorching of the cheese during pizza baking. Therefore, the melting and browning behaviors of Mozzarella cheese may be related to factors such as FO release. Furthermore, because melting is a dynamic process occurring over a large temperature range and time, specific events occurring at specific times and temperatures may also be important to achieve desirable melting and browning properties of reduced fat Mozzarella cheese on pizza. More work is needed in this area.

Although the improvements in whiteness and yield are probably the result of homogenization rather than of fat type, Salatrim® provides certain advantages for nutritional labeling. For a given amount of fat, Salatrim® contributes only 5/9 of the calories of milk fat, and, thus, only 5/9 of the current total fat content might need to be claimed (FDA proposal). If this interpretation of the label declaration is approved by the FDA, the M50 and M100 cheeses in this study, but not the control cheese, could be labeled as low fat.

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