Influence of Homogenization Conditions of Buffalo Milk on the Recovery of Milk Constituents and Yield of Mozzarella Cheese

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Abstract: Mozzarella cheese making involves losses of milk constituents, especially during plasticizing stage of cheese curd. Buffalo milk is considered more suitable than cow milk for Mozzarella cheese making, especially in terms of colour, yield and stretch property of resultant product. Homogenization of milk reduces the losses of milk constituents, increases its whiteness and is expected to render superior flavor to cheese. The fat globule size for buffalo milk is larger and the cheese tends to be firmer and chewy as compared to cow milk counterpart. Homogenization of buffalo milk is of significance in this regard since it can improve the color, recovery of milk constituents culminating in higher cheese yield, a mellower product with lower tendency to oil-off during baking applications. Since the conditions of homogenization affects the recovery of milk constituents, it was decided to study temperature and pressure of homogenization on such aspect including cheese yield.

Homogenization of standardized buffalo milk at 55 or 65°C and 4.90 MPa (P_2) pressure is found beneficial with regard to recovery of milk fat, while use of lower pressure i.e. 2.45 MPa (P_1) at above temperatures is found beneficial for protein and TS recoveries. P_2 pressure is more beneficial than P_1 pressure in improving the fat recovery in buffalo milk Mozzarella cheese. There is an improvement in the yield of Mozzarella cheese with an increase in homogenization pressure. The yield of Mozzarella cheese prepared using buffalo milk homogenized at P_2 and P_1 pressure (at 65°C) was 17.00% and 16.10% respectively. The recoveries of milk fat, protein and TS and per cent yield for control cheese was 83.68%, 84.10%, 56.74% and 14.53% respectively.

Keywords: Mozzarella cheese, yield, recovery of milk solids, loss in whey, loss in moulding water.

INTRODUCTION

Homogenization of milk is one of the pretreatments employed selectively in the manufacture of certain varieties of cheese to obtain specific effects. Homogenization has been employed in the manufacture of Mozzarella cheese from cow milk to reduce milk solids losses in whey and moulding water [1,2] and to reduce excessive fat leakage when used on pizzas [1]. There is scanty literature on the use of homogenization of milk for Mozzarella cheese manufacture. Mozzarella cheesemaking involves considerable losses of milk constituents, especially during plasticizing stage of cheese curd in hot water. Buffalo milk is considered to be more suitable than cow milk for Mozzarella cheese making, especially in terms of the yield, color and stretch property of resultant product [3]. Homogenization of milk is reported to reduce the losses of milk constituents (especially milk fat), increase the whiteness of product and is expected to yield cheese having superior flavor [1,2]. The fat globules of buffalo milk are of larger size and the tendency of cheese obtained from buffalo milk tends to be firmer and chewy as compared to cow milk counterpart. Hence, homogenization of buffalo milk is of greater significance, since such treatment can improve the color, recovery of milk constituents in cheese culminating in higher yield, a mellower product with lower tendency to oil-off during baking applications. Attaining higher yield from unit quantity of cheese milk enables the cheese maker to earn better returns. Since the conditions of homogenization (viz. temperature and pressure) affect the recovery of milk constituents, it was decided to study these relevant factors on the recovery of milk constituents and its impact on cheese yield.

MATERIALS AND METHODS

Homogenization of Buffalo Milk

The buffalo milk standardized to casein/fat ratio of 0.7 (i.e. 4.4% milk fat) was homogenized in a single stage ‘Rannie-make’ homogenizer, after pasteurization. For homogenization of milk, three temperatures viz., 45 (T_1), 55 (T_2) and 65°C (T_3) and two pressures viz. 2.45 MPa (i.e. 25 kg/cm^2) (P_1) and 4.90 MPa (i.e. 50 kg/cm^2) (P_2) were employed. Low pressure homogenization was specifically adopted in order to reduce the adverse effects of treatment on curd forming characteristics of milk and on baking properties (shred, melt, stretch) of cheese.

Manufacture of Mozzarella Cheese

Control cheese (C) from unhomogenized buffalo milk, standardized to casein/fat (C/F) of 0.7 was made

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as per the method of Sundar and Upadhyay (1990) employing 'starter culture technique' [4]. Experimental Mozzarella cheese were prepared from homogenized buffalo milk, standardized to same C/F ratio as for control as per the procedure of Jana and Upadhyay (1993) [5]. Cheesemaking from homogenized buffalo milk necessitated certain modifications to obtain a product having satisfactory stretching qualities. These modifications were (a) addition of 0.01% (w/w) calcium chloride to milk, (b) draining whey at higher acidity viz., 0.43 and 0.45% LA when adopting P_1 and P_2 pressure respectively, (c) using lower molding water temperature of 85-90^{o}C, and (d) keeping less contact period of curd with mould water viz., 2-3 min and 1-2 min. respectively when employing P_1 and P_2 pressures. For cheesemaking from control milk (unhomogenized), calcium salt was avoided, draining of whey was done at 0.41% LA, moulding water temperature of 95^{o}C was used and the contact period of cheese curd with moulding water was 4-5 min.

Analyses

The milk and Mozzarella cheese were analyzed for fat by Gerber method and protein by micro-Kjeldahl method [6]. The total solids (TS) of milk and Mozzarella cheese was determined by the Mojonnier method [7]. The titratable acidity (TA) of milk was determined using BIS method [8]. The Mozzarella cheese was analyzed for ash by BIS method [9]. The pH and TA of cheese were determined using the procedures outlined by Patel *et al.* [10].

Statistical Analysis

The statistical analysis of the data was carried out using factorial experiment in a Completely Randomized Design [11]. The values provided for homogenized milk cheeses are mean of six treatments, replicated thrice (i.e. 3 temperatures and 2 pressures).

RESULTS AND DISCUSSION

Composition of Cheese

The average values of proximate composition of Mozzarella cheese obtained from homogenized buffalo milks (irrespective of pressure and temperature of homogenization) has been depicted in Table **1** along with the values for control cheese (C). Homogenization of milk had a significant ($P<0.05$) effect on the composition of Mozzarella cheese through a higher moisture and fat retention; the homogenized milk cheeses had significantly higher moisture and fat on dry matter (FDM) content when compared to C (Table **1**). Higher acidity of whey at draining yielded homogenized milk cheeses having lower pH and lower ash content compared to C. Higher homogenization pressure had a more marked effect on the composition of the resultant cheeses than did higher temperature of homogenization.

An increase in the proportion of homogenized milk blended with unhomogenized milk yielded Mozzarella cheese having higher moisture content; a corresponding decrease in the protein content of cheese was noted. There was a significant increase in the FDM content of cheese as a result of milk homogenization [12].

Yield of Mozzarella Cheese

The per cent yield of homogenized milk cheeses was higher by 5.85 to 17.00 % over control cheese

Table 1: Composition of Mozzarella Cheese from Unhomogenized and Homogenized Buffalo Milks

Parameters	Mean (Per cent)	
	Control cheese*	Homogenized milk cheese#
Moisture (%)	46.07^x	50.73^y
Fat (%)	25.75	26.13
Fat on dry matter (%)	47.78^x	53.09^y
Protein (%)	21.46^x	19.75^y
Protein at constant (46%) moisture (%)	21.50	21.64
Salt (%)	0.92	0.93
Ash (%)	2.32^x	2.01^y
Acidity (% LA)	0.60^x	0.65^y
pH	5.11^x	5.00^y

* - Mean of 6 lots, # - Mean of 18 lots, ^{xy}Values with different superscripts within the same row differed significantly; $P < 0.05$.

Table 2: Yield of Mozzarella Cheese as Affected by Homogenization Conditions

Temperature	Yield (kg cheese/100 kg milk)		
	Pressure		Average for Temperature
	P1	P2	
T1	15.38	16.18	15.78[a]
T2	15.83	16.47	16.15[ab]
T3	16.10	17.00	16.55[b]
Average for Pressure	15.77[x]	16.55[y]	
Source	S.Em.	C.D.(0.05)	C.V.%
P	0.149	0.459	2.76
T	0.182	0.562	
PXT	0.258	NS	

P_1 and P_2 – homogenization pressure of 2.45 and 4.90 MPa, T_1, T_2, T_3 – homogenization temperature of 45, 55 and 65°C; [xy]Values with different superscripts within the same row differed significantly; $P < 0.05$; [ab]Values with different superscripts within the same column differed significantly; $P < 0.05$.

(Table **2**). The cheese yield showed an increasing trend with an increase in either the temperature or pressure of milk homogenization. Such effects of homogenization pressure and temperature were found to be statistically significant (P<0.05). Cheeses made using P_2 pressure had significantly (P<0.05) higher yield compared to its counterpart made using P_1 pressure. Likewise, cheeses made using T_3 temperature had significantly (P<0.05) higher yield as compared to cheese made from milk homogenized at T_1 temperature. However, the yield was at par for cheeses made from milks homogenized at T_1 and T_2, as well as T_2 and T_3 temperatures. The interaction of homogenization pressure and temperature (P X T) failed to produce any significant effect on the cheese yield (Table **2**).

It is evident from Table **2** that the cheese yield, expressed as kg cheese/100 kg milk, increased significantly (P<0.05) on homogenization (i.e. increase in yield of 8.53 % and 13.90 % over control was noted when adopting P_1 and P_2 pressures respectively). The fat and protein recoveries, as depicted in Table **3**, were found to be significantly (P<0.05) higher in case of homogenized milk cheeses (i.e. 13.00 % and 2.38 % over control respectively) owing to reduced losses of such constituents in whey and moulding water (Appendix-I). However, this did not reflect in the same proportion on cheese yield, possibly due to simultaneous increased losses of other milk constituents (i.e. minerals and lactose) at draining and plasticizing stages.

These findings are in agreement with those obtained by Quarne *et al.* [2] and Schafer and Olson [13] who observed significantly higher yield of cow milk Mozzarella cheese compared to control; the cheeses were prepared using starter culture and direct acidification methods respectively by these workers. An increase in the yield of cheese with an increase in homogenization pressures has also been reported by Rao *et al.* [14] and Kwak *et al.* [15]. The difference in the per cent yield between control and homogenized milk cheeses could be attributed to an increase in the overall recovery of milk solids and higher moisture content in homogenized milk cheeses. The increase in the yield of homogenized milk cheeses with an increase in pressure and temperature of homogenization could be ascribed to the further improvement in the recovery of milk fat (Table **4**) and owing to greater increase in the moisture content of resultant cheeses (Table **1**).

The average value of per cent yield for control cheese (14.53 %) obtained in the present study is higher than that reported by Kwak *et al.* [15] (i.e. 10.54 %) for cow milk Mozzarella, but lower than those reported by Patel *et al.* [10] (i.e. 15.13-19.20 %) and Sundar and Upadhyay [4] (i.e. 14.76-15.46 %) for Mozzarella cheese made from buffalo milk. The observed differences may be ascribed to the differences in the content of protein in milk, cooking temperature employed, TS recovery in cheese and moisture content of resultant cheese.

Recovery of Milk Constituents in Mozzarella Cheese

Fat Recovery in Cheese

The data presented in Table **3** show that the average values of fat recovery in Mozzarella cheese ranged from 83.68 % for control to 98.15 % for sample

Table 3: Comparison of Yield and Recovery of Milk Constituents in Mozzarella Cheese Obtained from Unhomogenized and Homogenized Milks

Treatment	Yield (kg cheese/100 kg milk)	Recovery of milk constituents (%)		
		Fat	Protein	Total Solids
C	14.53^{a}	83.68^{a}	84.10^{a}	56.74^{a}
P1T1	15.38^{b}	87.98^{b}	85.62^{b}	57.32^{a}
P1T2	15.83^{b}	93.08^{c}	88.54^{c}	58.46^{a}
P1T3	16.10^{bc}	96.72^{d}	89.65^{c}	58.98^{a}
P2T1	16.18^{c}	94.72^{e}	82.37^{d}	56.43^{a}
P2T2	16.47^{c}	96.73^{df}	84.41^{ae}	57.19^{a}
P2T3	17.00^{cd}	98.15^{g}	86.00^{ab}	57.70^{a}
Control vs. Homogenized				
S.Em.	0.261	0.346	0.652	0.723
C.D.(0.05)	0.561	0.742	1.40	NS
C.V.%	2.63	0.597	1.22	2.01

C – control, P_1 and P_2 – homogenization pressure of 2.45 and 4.90 MPa, T_1, T_2, T_3 – homogenization temperature of 45, 55 and 65°C; ^{abcdefg}Values with different superscripts within the same column differed significantly; $P < 0.05$.

P_2T_3. The tabulated values indicates that homogenized milk cheeses had significantly ($P<0.05$) higher fat recovery compared to control. The recovery of fat in homogenized milk cheeses as compared to control was higher by 5.14% for cheese P_1T_1 to 17.29 % for sample P_2T_3. Other cheeses had intermediate values.

The values depicted in Table **4** reveals that fat recovery in cheese showed an increasing trend with an increase in the pressure and temperature of homogenization. The effects of homogenization pressure and temperature per se and their interaction (P X T) were found to be statistically significant. The highest fat recovery was associated with cheese sample P_2T_3 which was significantly ($P<0.05$) different from the fat recovery associated with the rest of the homogenized milk cheeses.

The differences in the values of fat recovery between control and homogenized milk cheeses as well as amongst homogenized cheese samples could be attributed to the lower fat losses in whey and mouldling water (Appendix-I) as a consequence of milk homogenization. These effects were marked with an increase in the homogenization pressure or temperature as a result of increased efficiency of milk homogenization. These findings are in agreement with those reported for Mozzarella [1,2] and Kachkaval [16] cheeses made from homogenized cow milk; Kachkaval is also a plasticized cheese variety. Quarne *et al.* [2] reported greater fat recovery (i.e. 92.59 %) in case of homogenized (3.45 MPa pressure) cow milk Mozzarella cheese made by starter culture method as compared to its unhomogenized counterpart (i.e. 91.98 %).

Table 4: Recovery of Milk Constituents in Mozzarella Cheese as Affected by Homogenization Conditions

Temp.	Fat recovery			Protein recovery			TS recovery		
	Pressure		Av. for Temp.	Pressure		Av. for Temp.	Pressure		Av. for Temp.
	P1	P2		P1	P2		P1	P2	
T1	87.98	94.72	91.35^{a}	85.62	82.37	83.99^{a}	57.32	56.43	56.88^{a}
T2	93.08	96.73	94.91^{b}	88.54	84.41	86.47^{b}	58.46	57.19	57.83^{a}
T3	96.72	98.15	97.44^{c}	89.65	86.00	87.82^{b}	58.98	57.70	58.34^{a}
Av. for Pressure	92.59^{x}	96.53^{y}		87.93^{x}	84.26^{y}		58.25^{x}	57.11^{x}	
Source	S.Em.	C.D.(0.05)	C.V.%	S.Em.	C.D.(0.05)	C.V.%	S.Em.	C.D.(0.05)	C.V.%
P	0.170	0.525	0.54	0.368	1.135	1.28	0.368	1.134	1.91
T	0.209	0.643		0.451	1.390		0.451	NS	
PXT	0.295	0.909		0.638	NS		0.637	NS	

P_1 and P_2 – homogenization pressure of 2.45 and 4.90 MPa, T_1, T_2, T_3 – homogenization temperature of 45, 55 and 65°C; ^{xy}Values with different superscripts within the same row differed significantly; $P < 0.05$; ^{abc}Values with different superscripts within the same column differed significantly; $P < 0.05$.

The average value of per cent fat recovery for control cheese (83.68 %) obtained in the present study is similar to that reported by Patel *et al.* [10], but somewhat higher than that reported by Sundar and Upadhyay [4] for buffalo milk Mozzarella cheese made from 3.0 % fat milk and C/F of 0.7 adjusted milk respectively. The observations on the recovery of milk fat in the present study are supported by the findings of Breene *et al.* [1], Maxcy *et al.* [17] and Quarne *et al.* [18].

Protein Recovery in Cheese

The data presented in Table **4** show that the average values of protein recovery in cheeses ranged from 82.37 % for sample P_2T_1 to 89.65 % for sample P_1T_3. The tabulated values indicates that control sample had significantly (P<0.05) higher protein recovery when compared with homogenized cheese sample P_2T_1. On the other hand, control cheese had significantly (P<0.05) lower protein recovery when compared to rest of the homogenized milk cheeses, except for sample P_2T_2. The recovery of protein in homogenized milk cheeses as compared to control was higher by 0.37 % for sample P_2T_2 to 6.60 % for sample P_1T_3. Other homogenized milk cheeses, barring sample P_2T_1 had intermediate protein recovery values.

The values presented in Table **4** reveals that the protein recovery in cheeses showed an increasing and decreasing trend with an increase in the temperature and pressure of homogenization respectively. Such effects of homogenization pressure and temperature *per se* were found to be statistically significant. However, the protein recoveries of cheese made from milk homogenized at T_2 and T_3 temperature were at par with each other, but were significantly (P<0.05) higher than that of cheeses made from milk homogenized at T_1 temperature. The interaction of homogenization pressure and temperature (P X T) failed to produce any significant effect on the protein recovery of cheese.

The differences in the values of protein recovery between control and homogenized milk cheeses as well as amongst homogenized milk cheeses could be attributed to the overall lower losses of protein in whey as well as moulding water (Appendix-I), when the milk intended for cheese making was homogenized. The above mentioned effect was pronounced with increasing homogenization temperature only. The observed lower value of protein recovery for homogenized cheese P_2T_1 compared to control could be ascribed to higher losses of protein in whey as well as in moulding water for the former cheese.

The observed increasing trend in the recovery of protein in cheese with increase in homogenization temperature maybe ascribed to the improved homogenization efficiency. This in turn, might have resulted in increased adsorption of casein on the fat globules giving rise to more fat-protein complex [19], leading to reduced protein losses in whey as well as in moulding water. The higher recovery of protein in homogenized milk cheese is in accordance with the explanation of Mabbitt and Cheeseman [20]. The observed decline in the recovery of protein with an increase in homogenization pressure is in agreement with the findings of Breene *et al.* [1] for cow milk Mozzarella cheese.

The average value of protein recovery for control cheese (i.e. 84.10 %) obtained in the present study is within the range reported by Sundar and Upadhyay [4] but slightly higher than that reported by Patel *et al.* [10] for buffalo milk Mozzarella cheese. Zedan *et al.* [21] reported lower protein recovery of 68.30 % for Mozzarella cheese made from buffalo milk standardized to 3.0 % milk fat using starter culture method [21]. The differences in the values of protein recovery between the present study and that reported by Patel *et al.* [10] might have been due to variation in the losses of protein in whey as well as in moulding water in the two studies.

Total Solids Recovery in Cheese

The average values of TS recovery in homogenized milk cheeses ranged from 56.43 % for sample P_2T_1 to 58.98 % for sample P_1T_3. Except for cheese sample P_2T_1, which had TS recovery lower than that of control cheese by 0.55%, the per cent increase in TS recovery of homogenized milk cheeses over that of control ranged from 0.79 % for P_2T_2 to 3.95 % for sample P_1T_3 (Table **3**). The TS recovery in case of homogenized milk cheeses was somewhat higher (though non-significant difference) than that of control cheese.

The TS recovery in Mozzarella cheeses showed an increasing and decreasing trend with an increase in the temperature and pressure of homogenization respectively (Table **4**). However, only homogenization pressure exerted significant effect on TS recovery of cheeses. The cheeses made from milk homogenized at P_1 pressure had significantly (P<0.05) higher TS recovery as compared to its counterpart made using P_2 pressure. The interaction of homogenization pressure and temperature (P X T) failed to have any significant

Appendix I: Losses of Milk Constituents in whey and Moulding Water as Influenced by Homogenization of Cheese Milk

Treatment	Losses in whey (%)		Losses in moulding water (%)	
	Fat	Protein	Fat	Protein
C	4.62	13.89	4.42	1.11
P_1T_1	3.92	12.38	1.32	1.04
P_1T_2	2.46	9.46	0.53	0.93
P_1T_3	1.68	8.35	0.29	0.88
P_2T_1	2.85	15.63	0.63	1.17
P_2T_2	1.83	13.59	0.43	1.03
P_2T_3	1.54	12.00	0.28	0.96

C – control, P_1 and P_2 – homogenization pressure of 2.45 and 4.90 MPa, T_1, T_2, T_3 – homogenization temperature of 45, 55 and 65°C.

effect on the TS recovery of Mozzarella cheeses (Table **4**).

The observed differences between control and homogenized milk cheeses with respect to TS recovery could be ascribed to the overall reduced losses of fat and protein in whey as well as in moulding water (Appendix-I) in the manufacture of homogenized milk cheeses. Moreover, the use of lower temperature of plasticizing water (i.e. 85-90°C vs. 95°C for C) and its contact period with cheese curd made from homogenized milks could have contributed to the observed difference in the TS recoveries. Such finding is in agreement with those reported for Mozzarella cheese [1,2]. Krcal and Durko [16] however did not find any significant difference in TS recovery of Kachkaval cheese made from unhomogenized or homogenized milks. Krcal and Herian [22] reported that the temperature of plasticizing solution partly affected the dry matter content of Kachkaval cheese.

The observed significant difference in the TS recovery of homogenized milk cheeses with an increase in the pressure of homogenization might be attributed to the higher losses of TS in whey and higher losses of protein in moulding water in the manufacture of cheeses made from milk homogenized at P_2 pressure as compared to P_1 pressure (Appendix-I). Such decrease in the TS recovery of homogenized milk cheese with an increase in homogenization pressure has also been reported for cow milk Mozzarella cheese [1].

The average value of TS recovery for control cheese obtained in the present study is well within the range reported by Patel *et al.* (1986), but higher than that reported by Sundar and Upadhyay [4] for Mozzarella cheese made from unhomogenized buffalo milks. Such differences in the findings could be attributed to the variation in the losses of TS in whey and moulding water in the respective studies. The observations on the recovery of TS in the present study are supported by the findings of Quarne *et al.* [2].

Bhattarai and Acharya [23] noted fat lost in whey and stretch water to be 9.84 % and 6.20 % respectively when Mozzarella cheese was prepared from unhomogenized buffalo milk. These workers noted fat and protein recovery in buffalo Mozzarella cheese to be 83.83 % and 76.53 % respectively; yield of cheese (at constant moisture) was 10.84%. According to report of Ahmed *et al.* [24], the losses of fat and total protein in whey was 7.06 % and 2.80 % respectively, while the losses of fat and total protein in kneading water was 9.34 % and 0.67 % respectively.

It can be concluded that use of low pressure homogenization (i.e. 2.45 or 4.90 MPa pressure) of standardized buffalo milk at 55 or 65°C enables significant improvement in the recoveries of milk solids, especially fat and protein leading to advantage in cheese yield. Low temperature homogenization (i.e. 45°C) is not advocated to reap the benefits of such milk pre-treatment on cheese yield.

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