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Effects of reconstitution rate and hydration time on the yield of fresh cheese from reconstituted milk

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ABSTRACT: Powdered milk, due to its versatility, extended shelf life, and ease of transport, serves as a viable alternative for cheese production, especially in regions with limited access to fresh milk. However, the cheese yield from reconstituted milk may vary depending on factors such as the concentration of milk powder in the solution and the degree of casein hydration. This study aimed to evaluate the effects of reconstitution rate and hydration time of powdered milk on cheese yield. A 3 × 3 factorial design with three replicates per treatment was employed, testing reconstitution rates of 10%, 20%, and 30%, along with hydration times of 0, 1, and 2 hours. Cheeses were prepared by reconstituting whole milk powder in heated water, followed by the addition of calcium chloride and Ha-la[®] chymosin. The mixture was incubated at 37 °C for coagulation, then the curd was cut and stirred, concluding with hot-water syneresis and moulding. The resulting cheeses were evaluated based on yield parameters, including weight yield, adjusted yield, protein losses in whey, and protein retention in cheese. Results showed that higher concentrations of milk powder significantly increased yield, rising from 17.5% at 10% concentration to 22.6% at 30%, whereas hydration time had no notable effect. This may be due to the rapid solubilization of casein or the possibility that hydration periods longer than 2 hours are necessary to influence protein interactions. Protein retention in cheese improved from 58.4% to 78.5%, while protein losses in whey decreased from 41.6% to 21.5% with increasing concentration. Production costs remained consistent across treatments; however, the 30% concentration yielded more cheese per batch, thus enhancing the cost-benefit ratio for processors. These findings support the use of reconstituted milk for cheese production in areas where fresh milk is scarce, offering a practical and economical option for the dairy industry. In conclusion, increasing the concentration of milk powder in reconstituted milk enhances cheese yield, while hydration time does not significantly influence these parameters.

Keywords: cheese yield; dairy processing; milk powder; milk technology; protein retention.



Efeitos da taxa de reconstituição e do tempo de hidratação no rendimento de queijo fresco a partir de leite reconstituído

RESUMO: O leite em pó, devido à sua versatilidade, ao maior prazo de validade e à facilidade de transporte, serve como uma alternativa viável para a produção de queijo, especialmente em regiões com acesso limitado a leite fresco. No entanto, o rendimento do queijo a partir do leite reconstituído pode variar dependendo de fatores como a concentração de leite em pó na solução e o grau de hidratação da caseína. Este estudo teve como objetivo avaliar os efeitos da taxa de reconstituição e do tempo de hidratação do leite em pó no rendimento do queijo. Foi empregado um delineamento fatorial 3×3 com três repetições por tratamento, testando taxas de reconstituição de 10%, 20% e 30%, juntamente com tempos de hidratação de 0, 1 e 2 horas. Os queijos foram preparados pela reconstituição do leite em pó integral em água aquecida, seguida pela adição de cloreto de cálcio e quimosina Ha-la®. A mistura foi incubada a 37 °C para coagulação, então a coalhada foi cortada e agitada, finalizando-se o processo com sinérese em água quente e moldagem. Os queijos resultantes foram avaliados com base em parâmetros de rendimento, incluindo rendimento em peso, rendimento ajustado, perdas de proteína no soro e retenção de proteína no queijo. Os resultados mostraram que concentrações mais altas de leite em pó aumentaram significativamente o rendimento, passando de 17,5% na concentração de 10% para 22,6% na concentração de 30%, enquanto o tempo de hidratação não teve efeito notável. Isso pode ser devido à rápida solubilização da caseína ou à possibilidade de que períodos de hidratação maiores que 2 horas sejam necessários para influenciar as interações proteicas. Com o aumento da concentração, a retenção de proteína no queijo melhorou de 58,4% para 78,5%, enquanto as perdas de proteína no soro diminuíram de 41,6% para 21,5%. Os custos de produção permaneceram consistentes entre os tratamentos; no entanto, a concentração de 30% rendeu mais queijo por lote, melhorando a relação custo-benefício para os processadores. Essas descobertas apoiam o uso de leite reconstituído para a produção de queijo em áreas onde o leite fresco é escasso, oferecendo uma opção prática e econômica para a indústria de laticínios. Conclui-se que o aumento da concentração de leite em pó no leite reconstituído melhora o rendimento do queijo, enquanto o tempo de hidratação não influencia significativamente os parâmetros testados.

Palavras-chave: leite em pó; processamento de laticínios; rendimento do queijo; retenção de proteína; tecnologia do leite.

1 Introduction

Powdered milk is widely recognized as one of the most efficient and practical forms of milk preservation and commercialization, primarily due to its high versatility and extended shelf life. Through the dehydration process, most of the water content is removed, resulting in a product that can be stored safely at room temperature

for long periods (Ryabova; Semipyatny; Galstyan, 2023). This eliminates the immediate need for refrigeration and enables stable storage and handling of milk over extended timeframes, which is particularly advantageous for distribution in remote regions or areas lacking adequate infrastructure for the transportation and preservation of fresh milk (Karaman *et al.*, 2015; Li *et al.*, 2019). Furthermore, by significantly reducing both the volume and weight of liquid milk, the dehydration process optimizes logistic efficiency, thereby reducing transportation and storage costs. This cost-effective aspect plays a critical role in supplying dairy products to markets located in regions with insufficient milk production capacity or geographical barriers (Phosanam *et al.*, 2021). In this context, understanding how to maximize the yield of cheeses produced from reconstituted milk is particularly relevant for dairy plants operating in regions where cold-chain logistics are limited, as the identification of optimal treatments can support the production of larger cheese volumes per batch at reduced cost.

Historically, the use of reconstituted milk in cheese production dates back to the early 20th century. According to Gilles *et al.* (1979), it gained strategic importance during and after World War II, a period marked by severe shortages of fresh milk. In this context, reconstituted milk emerged as a reliable alternative, enabling continued cheese production despite logistical constraints. Since then, its application in cheesemaking has consolidated as a relevant technological solution, particularly in regions where milk supply is seasonally inconsistent or where dairy plants are located far from milk-producing zones (Bihola *et al.*, 2025). In addition to overcoming supply limitations, the deliberate inclusion of dairy-derived solids, such as powdered milk, condensed milk, milk protein concentrates, and ultrafiltered milk fractions, serves to achieve important technological goals in cheese manufacture. Among these are the enhancement of casein concentration within the milk matrix and the maximization of protein recovery during the cheesemaking process, both of which contribute to higher yields and improved product consistency (Faion *et al.*, 2019; Giménez *et al.*, 2023).

Nevertheless, cheese production from reconstituted milk poses particular technological challenges, especially regarding the reconstitution rate and hydration time. The reconstitution rate refers to the proportion of powdered milk added to a specific volume of water to reproduce the characteristics of whole milk, while hydration time corresponds to the interval during which the milk powder is allowed to interact and dissolve thoroughly in the aqueous medium before undergoing further processing steps. Syneresis, defined as the expulsion of whey from the curd during and after coagulation, and curd firmness, understood as the structural strength and resistance of the coagulum to breaking or crumbling, are among the key parameters influenced by these factors. These parameters are crucial because they directly influence the chemical composition, colloidal stability, and rheological behavior of reconstituted milk, thereby affecting critical aspects of cheesemaking, including coagulation behavior, curd firmness, syneresis control, and moisture retention capacity (Al-Bedrani *et al.*, 2021; Katz *et al.*, 2016).

During reconstitution, complete and homogeneous hydration of milk proteins is fundamental to ensuring the structural integrity and functionality of the milk matrix used in cheesemaking. Inadequate reconstitution ratios or insufficient hydration periods may impair protein rehydration, leading to incomplete network formation in the curd and suboptimal moisture incorporation, which can ultimately compromise cheese yield and textural properties (Avsar *et al.*, 2010). According to Bihola *et al.* (2025), a hydration period of approximately two hours is sufficient to promote both protein and lipid stabilization, resulting in a more uniform and emulsified reconstituted milk system suitable for cheese production. This step is

therefore essential to achieving desirable quality and yield in cheeses derived from reconstituted milk bases.

Moreover, variations in reconstitution conditions, including both the concentration of milk solids and hydration time, can influence the distribution and retention of solids in the coagulated mass, which are decisive factors in determining cheese yield and overall processing efficiency (Kayihura, 2024; Sharma; Jana; Chavan, 2012). Specifically, the concentration of milk powder during reconstitution has a direct impact not only on cheese yield but also on the loss of casein fractions in the whey. Studies indicate that changes in reconstitution concentration affect the partitioning of casein micelles between the curd and the whey, thereby influencing the efficiency of curd formation and the retention of valuable milk proteins (Kayihura, 2023).

Despite the recognized importance of these parameters, few studies have simultaneously evaluated different reconstitution rates and hydration times to determine their combined effects on the yield and protein retention of fresh cheeses. This gap in the literature is the focus of the present research. Based on previous findings, the working hypothesis is that higher reconstitution rates and longer hydration times enhance cheese yield and reduce protein losses in whey, thereby contributing to a more economically viable production process. Considering these technological factors and their implications for dairy processing, it is essential to investigate how both the hydration period and the powdered milk concentration affect cheese yield. Thus, the present study was designed to evaluate the influence of these two critical parameters, hydration time and reconstitution rate, on the production performance of fresh cheese manufactured from reconstituted milk.

Further in this paper, section 2 describes the procedures adopted for cheese production, considering the different treatments applied, yield analyses, protein quantification, and statistical analysis of the data. Section 3 presents, interprets, and compares the obtained data with reference studies addressing the same topic, and finally, section 4 answers the study's objective based on the findings.

2 Materials and methods

This section describes the primary materials and methods used for cheese production, as well as the procedures applied for yield analysis and statistical evaluation.

2.1 Cheese preparation

To evaluate the effect of whole milk powder reconstitution rate and hydration time on the yield of fresh cheeses, three reconstitution rates (w/v) were established: 10%, 20%, and 30%. The 10% rate represents the approximate standard reconstitution level for whole milk powders, while the higher rates were chosen to assess the effect of increased concentration on cheese yield and protein retention. Three hydration times were also tested: 0 hours, 1 hour, and 2 hours, selected to reflect practical industrial conditions by minimizing production downtime while allowing sufficient protein hydration. Each treatment was performed in triplicate using different batches of whole milk powder from distinct brands.

For reconstitution, distilled water heated to 40 °C was used. The milk powder was manually dissolved by stirring with a polished solid glass rod until complete dissolution, requiring approximately 20 minutes. This duration was determined from preliminary observations, which indicated that it was sufficient to achieve complete visible solubilization and uniform dispersion of the powder. Visual confirmation of the absence of undissolved particles was adopted as the practical criterion, consistent with industrial reconstitution practices described in technical references for recombined milk (Bylund, 2003). The whole milk powder used in this study was a commercial product that complied with the standards of the Ministry of Agriculture, Livestock, and Food Supply (Brasil, 2018). According to these regulations, whole milk powder must contain a maximum of 5.0% (w/w) moisture, at least 26.0% (w/w) fat, and a minimum of 24.0% (w/w) protein in the non-fat dry extract.

The methodology proposed by Presente, Fraga and Schmidt (2016) was adapted for fresh cheese production. The reconstituted milk was transferred to 1 L beakers and heated in a water bath until reaching 37 °C. Subsequently, 0.8 mL of 50% calcium chloride and 0.8 mL of Ha-la® chymosin were added. After homogenization, the milk was incubated at 37 °C for 40 minutes to allow coagulation.

The curd obtained was cut into grains of approximately 1 cm and subjected to slow stirring for 5 minutes to promote whey drainage, followed by a 15-minute resting period and an additional 5 minutes of rapid stirring. The whey was then drained, and 250 mL of water at 60 °C was added to the curd to facilitate syneresis and obtain a firmer mass. The curd was molded, turned, and unmolded, thus completing the cheese production process. Salt was intentionally omitted to avoid osmotic effects that could interfere with protein hydration capacity, since sodium chloride promotes whey expulsion and alters moisture retention in the curd through osmotic pressure. This step ensured that cheese yield reflected solely the effects of milk powder concentration and hydration time, isolating these variables from the known syneresis-enhancing influence of salting commonly applied in industrial cheesemaking (Fox *et al.*, 2017).

2.2 Determination of moisture content in cheeses

The percentage moisture content of the cheeses was determined using the gravimetric method (IDF, 1987). Five grams of each sample were weighed in pre-dried crucibles previously dried in an oven at 102 ± 2 °C for at least one hour.

The crucibles containing the samples were then placed in the oven at 102 ± 2 °C for two hours. After removal, the crucibles were cooled in a desiccator for 10 minutes and weighed. The heating process was repeated for one hour until the mass difference between successive weighing did not exceed 0.001 g.

Moisture percentage was calculated using the following equation:

$$\text{Moisture (\%)} = \frac{[(m_0 + m_1) - m_2]}{m_1} \times 100 \quad (1)$$

where: m₀ = mass of the empty crucible; m₁ = mass of the sample; m₂ = mass of the crucible with the sample after drying to constant weight.

2.3 Determination of yield parameters

The economic yield of the cheeses was calculated by using the formula proposed by Katz *et al.* (2016):

$$\text{Yield (\%)} = \frac{\text{Liters of milk}}{\text{Weight of cheese}} \times 100 \quad (2)$$

Additionally, weight yield and adjusted yield were calculated according White and Ryan (1983):

$$\text{Weight yield (\%)} = \frac{\text{Weight of cheese}}{\text{Weight of milk}} \times 100 \quad (3)$$

$$\text{Adjusted yield (\%)} = \frac{\text{Weight yield}}{\text{Moisture content of cheeses}} \times 100 \quad (4)$$

For cost-benefit evaluation, the total cost of powdered milk (in BRL) used in each treatment was divided by the cheese mass obtained (Melilli *et al.*, 2002). This approach enabled internal comparison between treatments, indicating the milk powder proportion that yielded the lowest cost per kilogram of cheese.

2.4 Determination of protein content in reconstituted milk, whey and cheeses

The protein content of reconstituted milk and whey was determined using the formaldehyde method described by Zakari *et al.* (2022). After homogenization, 10 mL of each sample was pipetted into a beaker. To neutralize the protons present in milk, 1 mL of 1% phenolphthalein solution and 0.4 mL of 28% potassium oxalate solution were added.

Titration was performed with a 0.1 N sodium hydroxide (NaOH) solution until the equivalence point was reached. Then, 2 mL of 10% formalin was added, homogenized, and allowed to rest for 1 minute to enable the reaction. A second titration with 0.1 N NaOH was then performed. The protein content (%) was calculated from the volume of NaOH consumed in the second titration using the following formula:

$$\text{Protein (\%)} = V \times 1.747 \quad (5)$$

Where: V = volume (ml) of 0.1 N NaOH solution used in the second titration

The protein content of the cheeses was determined using the micro-Kjeldahl method (AOAC, 2023). Approximately 0.25 g of sample was weighed and placed into Kjeldahl microtubes with 2.0 g of catalytic mixture and 5 mL of pure sulfuric

acid. The tubes were heated in a digestion block at 350 °C for ~12 hours until the liquid turned clear with a bluish-green tint.

After cooling, 15 mL of 50% sodium hydroxide solution was added, producing a dark-grey solution. Distillation was carried out in an Erlenmeyer flask containing 25 mL of 4% boric acid until a final volume of ~100 mL was reached. After the addition of five drops of mixed indicator solution, titration was performed with 0.1 N hydrochloric acid until the endpoint was achieved.

Nitrogen and protein contents were calculated as follows:

$$\text{Nitrogen (\%)} = \frac{V \times N \times f \times 0,014}{m} \times 100 \quad (6)$$

$$\text{Protein (\%)} = \% \text{ nitrogen} \times 6.38 \quad (7)$$

where: V = volume of 0.1 N HCl used in titration; N = theoretical normality of 0.1 N HCl solution; f = correction factor for HCl solution; m = mass of the sample.

Two analytical approaches were employed, as the different matrices allow the use of distinct procedures to determine the same parameter. In formaldehyde titration, proteins are quantified through the release of amino groups that neutralize protons, allowing rapid and reliable determination in liquid systems such as milk and whey. In contrast, the micro-Kjeldahl method determines total nitrogen and converts it to protein content, which is the standard approach for solid dairy products, such as cheese. Although the chemical principles differ, the results are expressed as total protein (%) and can be directly compared to assess protein yield.

2.5 Statistical analyses

Each treatment combination was performed in three independent replicates. Statistical analyses were conducted to evaluate the effects of reconstitution rate and hydration time on the yield of fresh cheeses. Microsoft Excel Professional Plus 2019, JASP 0.18.0.0, and Sisvar 5.6 software were employed.

Initially, the Shapiro–Wilk test was used to assess data normality. For normally distributed data, Analysis of Variance (ANOVA) was applied, followed by Tukey’s multiple comparisons test. For non-normal data, the non-parametric Friedman test was applied, followed by the Conover test for multiple comparisons. For simple comparisons, the Kruskal-Wallis test with Dunn’s post-test was performed.

All analyses were conducted considering a 5% significance level.

3 Results and discussion

As shown in Table 1, cheeses produced with milk reconstituted at a concentration of 30% tended to yield more satisfactory results.

Table 1 ▶

Yield parameters and production cost of cheeses produced with reconstituted milk at concentrations of 10%, 20%, and 30%, with hydration times of 0, 1, and 2 hours.
Source: research data

	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Yield (L/kg) ¹	10	7.27 ± 1.86 Aa	–	–
	20	3.57 ± 0.12 ABa	–	–
	30	2.74 ± 0.20 Ba	–	–
	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Weight yield (%) ²	10	14.93 ± 3.40 Aa	17.48 ± 1.04 Aa	13.83 ± 1.50 Aa
	20	27.11 ± 2.25 Ba	29.30 ± 2.88 Ba	23.93 ± 4.52 Ba
	30	37.32 ± 5.03 Ca	37.95 ± 1.78 Ba	39.75 ± 4.08 Ca
	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Adjusted yield (%) ²	10	23.07 ± 5.63 Aa	29.10 ± 2.83 Aa	21.18 ± 3.30 Aa
	20	45.84 ± 2.66 Ba	48.19 ± 5.59 Ba	38.01 ± 7.10 Ba
	30	67.96 ± 8.20 Ca	66.40 ± 0.30 Ca	64.52 ± 6.81 Ca
	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Production cost (BRL/kg) ³	10	32.73 ± 8.36 Aa	26.52 ± 1.80 Aa	33.33 ± 4.14 Aa
	20	29.18 ± 4.79 Aa	30.69 ± 1.02 Aa	38.47 ± 7.16 Aa
	30	36.99 ± 2.72 Aa	35.53 ± 2.82 Aa	33.89 ± 2.83 Aa

¹ Means within a row with identical superscripts do not differ significantly according to the Friedman test followed by Conover's post-hoc test ($p > 0.05$). Means within a column with different superscripts differ significantly according to the Friedman test, followed by Conover's post-hoc test ($p < 0.05$).

² Means within a row with identical superscripts do not differ significantly according to ANOVA followed by Tukey's post-hoc test ($p > 0.05$). Means within a column with different superscripts differ significantly according to ANOVA followed by Tukey's post-hoc test ($p < 0.05$).

³ Means within a row and column with identical superscripts do not differ significantly according to ANOVA ($p > 0.05$).

It was observed that the hydration time of reconstituted milk did not significantly influence ($p > 0.05$) the yield of the cheeses produced for any of the evaluated parameters. By contrast, as the concentration of powdered milk in the solution increased, a significant upward trend in cheese yield was noted ($p < 0.05$). Among the yield parameters evaluated, adjusted yield (%) exhibited the greatest variation across milk powder concentrations, increasing from 23.07% at 10% to 67.96% at 30%, demonstrating high sensitivity to reconstitution rate.

Weight yield (%) also increased significantly with increasing milk powder concentration, ranging from 14.93% at 10% to 37.32% at 30%, although the relative variation was lower than that observed for adjusted yield. In contrast, production cost (BRL/kg) remained relatively stable across treatments, with no significant differences detected among hydration times or milk powder concentrations. These results indicate that yield-related parameters, particularly adjusted yield, are more responsive to variations in milk powder concentration than to changes in hydration time, highlighting the importance of optimizing reconstitution rates to maximize cheese production efficiency.

These findings contrast with those of Avsar (2010), who reported increased cheese yield from reconstituted milk with extended hydration times. This divergence can likely be attributed to differences in milk powder composition, total solids concentration, and cheese type. While Avsar used a cheese base containing approximately 40% total solids and 14% protein to produce white-brined cheeses, the present study used reconstituted whole milk powders with lower solid content to produce fresh cheeses without ripening.

Additionally, the hydration times evaluated here (0 to 2 hours) were substantially shorter than those reported (up to 24 h), which likely limited any effect on casein micelle expansion and protein network formation. Differences in mixing, homogenization, and renneting conditions between studies may also contribute to the observed variation, reinforcing that the impact of hydration time on yield is highly dependent on both powder characteristics and the specific cheesemaking process employed.

Cheese yield is a critical parameter in dairy technology, representing the volume of milk required to produce one kilogram of cheese on a natural basis, i.e., considering the intrinsic moisture content of the product. In the present study, it was observed that, at hydration times of 0 and 2 hours, the concentration of reconstituted milk significantly influenced ($p < 0.05$) the yield of fresh cheeses. This indicates that both the solids content of the milk and the time allowed for protein hydration can alter the efficiency of cheese production.

This behavior can be explained by the fact that higher milk solids concentrations, resulting from the reconstitution process, promote greater retention of proteins and other milk components within the curd matrix during coagulation and whey separation. These results corroborate the findings of Awad, Salama and Ragb (2015), who also identified higher yields when powdered milk was incorporated into cheesemaking. The authors suggested that this strategy can be advantageous for optimizing industrial output.

Furthermore, other studies, such as those by Pires *et al.* (2021), emphasize that the addition of milk protein concentrates not only increases cheese yield but also enhances overall solids recovery and reduces whey volume, thereby contributing to greater efficiency and sustainability. Taken together, these findings highlight the relevance of adjusting milk composition through reconstitution and supplementation strategies to enhance yield, particularly in artisanal and semi-industrial production settings.

When analyzing weight yield, which considers the proportion of cheese weight relative to milk weight, and adjusted yield, which accounts for cheese moisture content, it was found that the concentration of powdered milk significantly affected ($p < 0.05$) these parameters, regardless of hydration time.

White and Ryan (1983) similarly observed that increasing solids concentration in reconstituted milk resulted in higher weight yield and adjusted yield in cottage cheese. Likewise, Tidona *et al.* (2021) found that mozzarella produced using a mixture of 40 g/100 g reconstituted skim milk powder and 60 g/100 g fresh milk achieved a weight yield comparable to cheeses made exclusively with fresh milk.

Al-Bedrani *et al.* (2021) also reported that adding whey protein concentrate increased cheese yield and improved rheological parameters, including elasticity and compressive strength. In agreement with the present findings, Alinovi *et al.* (2022) observed that incorporating 40% recombined milk in Crescenza-type cheese production did not significantly increase adjusted yield. While cheese produced exclusively with fresh milk showed an adjusted yield of 16.41%, that produced with 40% reconstituted milk presented a yield of 16.69%.

Sharma, Jana and Chavan (2012) reported that one of the most effective strategies to increase cheese yield from reconstituted powdered milk is the application of ultrafiltration techniques before production. This membrane-based process enables the selective removal of specific components, particularly whey proteins. However, while technically effective, ultrafiltration systems involve high equipment and operational costs, which may limit their applicability in artisanal or small-scale cheese production, where investment capacity and technical resources are constrained (Lima; Costa Júnior; Pinto, 2008).

The partial elimination of whey proteins before subjecting the milk to heat treatment is advantageous because it reduces the likelihood of thermally induced complexes forming between β -lactoglobulin and κ -casein, which are commonly observed in conventional powdered milk processing and negatively affect protein functionality and curd formation. By mitigating this interaction, ultrafiltration promotes greater protein recovery in the curd and, consequently, increases yield by up to 7.3%, making the performance of reconstituted milk comparable to that of raw milk in cheesemaking.

Giménez *et al.* (2023) investigated Cremoso cheese production and found that fortifying milk with 5.2% skim milk powder led to a statistically significant increase in yield, rising from 13.3% in the control group to 19.2% in the treated group. This represented an approximate 50% enhancement, reinforcing the role of increased total solids in curd retention.

The authors also reported superior recovery of total solids in fortified samples, attributed to improved retention of key macronutrients such as casein and lipids within the protein matrix. Collectively, these findings underscore the technological benefits of modulating milk composition, through concentration or selective protein removal, as a means to optimize cheesemaking performance, enhance efficiency, and potentially reduce production costs through improved raw material utilization.

Regarding cost analysis, which assessed the financial implications of powdered milk in each formulation, no statistically significant differences ($p > 0.05$) were observed among treatments. Although using 30% reconstituted milk led to a marked increase in cheese yield, this improvement in production efficiency did not result in a proportional rise in manufacturing costs. This indicates that, despite similar overall costs, higher-yield treatments, such as those with 30% milk powder concentration and 0 hour or 2 hours of hydration (Table 1), allow small- and medium-scale producers to obtain more cheese per batch, enhancing production efficiency without increasing the financial burden.

This outcome is considered favorable from an economic standpoint, as it suggests the possibility of enhancing yield without negatively affecting the overall cost structure. These findings are consistent with those of Awad, Salama and Ragb (2015), who emphasized that incorporating ingredients such as skim milk powder, milk protein concentrates, and stabilizers can reduce production costs by up to 40% per kilogram of cheese. This reduction is primarily attributed to significant yield improvements and the high consumer acceptance of the final product.

Thus, the strategic incorporation of such ingredients can contribute not only to technological advances in cheese manufacturing but also to the economic sustainability and competitiveness of dairy processing operations.

The protein content values in reconstituted milk, whey, and cheese are presented in Table 2.

Table 2 ▶

Protein content of milk, whey, and cheeses produced from reconstituted milk at concentrations of 10%, 20%, and 30%, with hydration times of 0, 1, and 2 hours.

Source: research data

	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Reconstituted milk (%)¹	10	1.33 ± 0.20 A	–	–
	20	2.94 ± 0.25 AB	–	–
	30	5.03 ± 0.35 B	–	–
	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Whey (%)²	10	0.45 ± 0.21 Aa	0.24 ± 0.03 Aa	0.24 ± 0.15 Aa
	20	0.70 ± 0.12 Aa	0.52 ± 0.08 ABa	0.93 ± 0.06 Aa
	30	1.64 ± 1.17 Aa	0.93 ± 0.35 Ba	1.49 ± 0.76 Aa
	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Whey/milk protein ratio (%)³	10	33.91 ± 15.87 Aa	18.26 ± 2.26 Aa	17.83 ± 10.94 Aa
	20	23.76 ± 4.16 Aa	17.62 ± 2.68 Aa	31.60 ± 2.18 Aa
	30	32.64 ± 23.21 Aa	18.52 ± 7.01 Aa	29.52 ± 15.10 Aa
	Milk powder concentration (%)	Hydration time (h)		
		0	1	2
Cheese (%)²	10	17.18 ± 0.55 Aa	15.07 ± 1.34 Aa	17.21 ± 1.09 Aa
	20	17.11 ± 1.29 Aa	14.63 ± 4.98 Aa	9.84 ± 2.01 Aa
	30	16.46 ± 0.77 Aa	14.45 ± 5.56 Aa	9.50 ± 5.40 Aa

¹ Means within a column with different superscripts differ significantly according to the Kruskal-Wallis test followed by Dunn's post hoc test ($p < 0.05$).

² Means within a row with identical superscripts do not differ significantly according to the Friedman test followed by Conover's post hoc test ($p > 0.05$). Means within a column with different superscripts differ significantly according to the Friedman test followed by Conover's post hoc test ($p < 0.05$).

³ Means within a row and column with identical superscripts do not differ significantly according to ANOVA ($p > 0.05$).

A significant increase ($p < 0.05$) in the protein content of reconstituted milk was observed as the concentration of dissolved milk powder increased. This outcome was expected, as higher solution concentrations proportionally increase the solid components, including proteins.

The protein content in whey corresponds primarily to soluble proteins, such as α -lactalbumin and β -lactoglobulin, that are not incorporated into the curd during coagulation and are consequently removed along with the whey fraction during cheese production (Yadav *et al.*, 2015). The extent to which these proteins are lost to the whey is governed by several physicochemical parameters, two of which are particularly influential. First, the presence and availability of calcium ions play a crucial role in the structural organization of casein micelles, as they form ionic bridges that stabilize the micellar network and enhance the retention of caseins within the curd matrix. Second, the milk pH during processing strongly affects protein behavior, as acidification alters protein solubility and net charge, potentially increasing their dispersion into the whey phase (Kayihura, 2023).

These complex and interdependent interactions underscore the importance of tightly regulated processing conditions, as even slight deviations can lead to suboptimal protein recovery, reduced cheese yield, and variability in final product quality. Consequently, understanding and managing the molecular dynamics of protein distribution between curd and whey is essential for ensuring both technological efficiency and product standardization in cheese manufacturing.

Additionally, challenges in hydrating caseins in reconstituted dairy products can lead to significant protein losses during syneresis. Syneresis, the expulsion of whey from the curd, can be particularly problematic when casein hydration is insufficient, since it results in incomplete network formation and, consequently, higher proportions of soluble proteins being lost to the whey. This not only reduces overall cheese yield but also increases whey protein concentration, potentially affecting both nutritional quality and process efficiency (Avsar, 2010).

A promising approach to mitigate this issue involves optimizing the solubility and hydration of milk proteins by dissolving milk powder under controlled temperature conditions during reconstitution. Current evidence suggests that reconstitution at approximately 50 °C significantly improves hydration and solubility, thereby reducing protein loss during cheese processing. However, excessively high temperatures can be detrimental, as they promote protein denaturation, compromising functionality and negatively affecting the quality of the final product (Bihola *et al.*, 2025).

In this context, it was observed that only reconstituted milk hydrated for one hour showed a significant increase ($p < 0.05$) in protein loss to the whey. This specific response at 1 h may be due to a transient stage in protein hydration, during which interactions between milk proteins and casein micelles temporarily favor protein solubilization into the whey, a phenomenon not observed at shorter (0 h) or longer (2 h) hydration times (Fameau *et al.*, 2022). However, a more detailed analysis of the relationship between whey protein content and reconstituted milk concentrations revealed no significant differences ($p > 0.05$) in protein loss across treatments. These findings are consistent with those of Gazi and Huppertz (2015), who reported that increasing the concentration of milk proteins in reconstituted solutions could destabilize the casein micelle suspension, thereby increasing protein loss during whey drainage.

On the other hand, Baig *et al.* (2022) observed that higher protein concentrations in milk reduced the loss of solid components, particularly fat and casein, during cheese production. Kayihura (2023) further demonstrated that increasing milk solids contributed to a proportional decrease in the loss of specific casein fractions, particularly α_2 -caseins and κ -caseins, during syneresis. Interestingly, in cheeses made with reconstituted milk concentrations of up to 25%, no significant losses of α_2 -caseins or β -caseins were observed in the whey, underscoring the complex relationship between milk composition and protein retention.

The absence of significant protein losses in whey ($p > 0.05$) corresponded to the lack of significant differences ($p > 0.05$) in the protein content of cheeses. Thus, it was observed that protein levels in the cheeses remained unaffected, regardless of reconstituted milk concentration or hydration time. Awad, Salama, and Ragb (2015) reported that incorporating protein concentrates into formulations can enhance protein retention in the final product, reducing losses to whey. Similarly, Tidona *et al.* (2021) found that cheeses produced with mixtures containing 40% skimmed milk powder and liquid milk yielded whey with 0.93% protein and cheeses with 16.5% protein.

The protein levels of the analyzed cheeses ranged from 9.50% to 17.21%, consistent with values typically reported for fresh cheeses. Protein content in fresh cheeses

generally ranges between 12.81% and 25.91%, suggesting that the use of milk powder in cheesemaking does not compromise the nutritional composition of the final product. This difference may also be attributed to higher moisture retention in the cheeses, partial protein loss in the whey during processing, and the use of reconstituted whole milk powder with variable protein content, which can all contribute to lower final protein concentrations. These findings are supported by Magenis *et al.* (2014) and Silva *et al.* (2020), who reported that milk powder preserves protein levels within expected ranges.

However, it is important to note that studies have shown that whole milk powder (WMP) may result in lower protein recovery compared with skim milk powder (SMP). This difference can be attributed to the higher fat content in WMP, which, when subjected to homogenization, interferes with curd formation. Homogenization of milk fat alters rheological properties, extending coagulation time and producing a curd of lower firmness. Fat globules disrupt the alignment of casein micelles, preventing efficient aggregation and reducing protein yield in the curd (Bihola *et al.*, 2025).

In contrast, Giménez *et al.* (2023) reported that supplementing with 5.2% skim milk powder significantly increased whey protein retention, particularly α -lactalbumin and β -lactoglobulin (variants A and B), within the cheese matrix. This improvement was attributed to the dual effect of SMP supplementation, which both increases casein concentration and raises the proportion of soluble whey proteins in the aqueous phase. The higher concentration of these proteins facilitates their incorporation into the curd during cheesemaking. This process is further enhanced by incomplete whey drainage, which results in greater whey protein retention in the curd. Consequently, this not only increases yield but also improves the protein profile of the final product.

4 Conclusion

Hydration times of up to 2 hours did not significantly affect cheese yield or protein content in either the whey or the final product. By contrast, increasing the concentration of milk powder improved cheese yield by up to 29.2% without increasing production costs. These results highlight the significant impact of solid concentration on cheese yield and emphasize that higher milk powder levels can enhance protein retention, reduce whey losses, and maintain the nutritional composition of the final product.

The incorporation of cream and whey protein concentrate (WPC) enhanced both fat and protein recovery, presenting viable strategies for improving the quality and efficiency of fresh cheese manufactured from reconstituted milk. Furthermore, these findings demonstrate that the use of reconstituted milk can be economically advantageous, offering practical benefits for both industrial and artisanal cheese production while promoting greater sustainability by reducing protein losses and improving process efficiency.

For future research, it is recommended to compare the performance of reconstituted milk with conventional fluid milk, evaluate formulations prepared with skim milk powder to isolate the role of fat in hydration and curd formation, and investigate the effects of extended hydration times beyond 2 hours to assess potential improvements in protein functionality and yield. Furthermore, texture evaluation should be incorporated to better understand the influence of formulation and processing variables on the sensory quality of the final product. Additionally, assessing the impact of processing variables on syneresis and texture will provide further insights into technological optimization and the sensory quality of fresh cheeses produced under different reconstitution conditions.

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Article contributions

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