



## مجلة العلوم الشاملة

عدد خاص بالمؤتمر الدولي الثالث للعلوم والتقنية المجلد (10)، العدد (37)، (نوفمبر 2025) ردمد: 3014-6266

Volume (10), Issue (37), (NOV. 2025) SICST2025, www.sicst.ly

Comprehensive Journal of Science

ISSN: 3014-6266

# Production of Probiotic Soft White Cheese Using a Mixture of Fresh Whole Cow's Milk and Powdered Milk

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إنتاج الجبن الأبيض الطري المدعم حيوياً باستخدام خليط من حليب البقر الطازج كامل الدسم والحليب المجفف

أمينة بلعيد الزير  $^1$ ، نجاة يوسف جونطو $^1$ ، سهيلة محمد أبوشعالة $^1$ ، أنسوار خيري أبوعيسى $^1$ ، خديجة بلعيد الرمالى $^1$ ، نسدير عبد المجيد عمسر $^1$ ، فاطمة خليفة دعباج $^1$ \*

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Received: 30-09-2025; Revised: 10-10-2025; Accepted: 31-10-2025; Published: 25-11-2025

#### **Abstract:**

Soft white cheese is a very popular traditional dairy product in Libya and is made from fresh or pasteurized milk. There are currently few documented reports on the production of fresh Libyan white cheese using Lactic acid bacteria. Thus, this study proposed a new protocol for producing soft white cheese using a mixture of whole cow's milk and whole milk powder, with the addition of lactic acid bacteria strains to obtain a probiotic-enriched cheese and reduce the proportion of whey as a by-product. Different ratios of whole milk powder (0, 10, 20, and 30%) were added to whole cow's milk during cheese production. The results showed that cheese yield increased to 74.43% when 30% milk powder was used, compared to 12.23, 26.70, and 55.10% for cheeses made with 0, 10, and 20% milk powder, respectively. Sensory evaluation revealed that using 10–20% milk powder combined with lactic acid bacteria starter had no significant effect on the sensory properties of the soft cheese compared to traditional white cheese. However, excessive addition of milk powder reduced sensory acceptance among panelists. To improve yield and texture quality without negatively affecting sensory characteristics, a balanced combination of the two milk sources is recommended to produce a high-quality product that meets consumer preferences.

**Keywords:** Fresh White cheese; Probiotic soft White Cheese; High-Yield White Cheese.

مجلة العلوم الشاملة المجلد (10)، العدد (37)، (نوفمبر 2025) ردمد: 3014-6266 ISSN: 3014-6266

## الملخص:

يُعد الجبن الأبيض الطري من المنتجات اللبنية واسعة الاستهلاك في ليبيا. سصنع من الحليب الطازج أو المبستر في الوقت الحالي لا توجد دراسات حول تدعيم الجبن الأبيض الطري بإستخدام بادئات بكتيرية والحليب المدفف. تناولت هذه الدراسة بروتوكولاً جديداً لتصنيع الجبن الأبيض الطري (الجبنة المعصورة اللبيبية) باستخدام مزيج من حليب البقر كامل الدسم والحليب كامل الدسم المجفف مع إضافة سلالات من بكتيريا حمض اللاكتيك لإنتاج الجبن مدعماً حيوياً مع نسبة أقل لفصل مصل الحليب كمنتج ثانوي، حيث تم إضافة نسب مختلفة من الحليب كامل الدسم المجفف (0، 10، 20، و30%) إلى حليب الأبقار في صناعة الجبن الأبيض الطري. أظهرت النتائج أن إنتاج الجبن ارتفع إلى 74.43% في الجبن المصنوع باستخدام 30% من الحليب المجفف، مقارنة بقيم إنتاجية بلغت 12.23، 26.70% و55.00% عند إضافة 0، 10، و20% من الحليب المجفف، على التوالي. كما بينت نتائج التحليل الإحصائي للخواص الحسية للجبن الطري مقارنة بالجبن الأبيض التقليدي. الحليب المجفف مع بادئ بكتيريا حمض اللاكتيك لم يؤثر في الخواص الحسية للجبن الطري مقارنة بالجبن الأبيض التقليدي. لكن الإفراط في الحليب المجفف قد يضعف القبول الحسي من قبل المقيمين. لتحسين العائد وجودة القوام دون التأثير سلبًا على الخصائص الحسية، لابد من التوازن بين المكونين للحصول على منتج ذو جودة عالية مرضية للمستهلك.

الكلمات المفتاحية: جبنة بيضاء طرية؛ جبنة بيضاء طرية مدعم جيوياً؛ جبنة بيضاء عالية الإنتاجية.

#### 1. Introduction

The development of traditional food products is an important issue in the food industry (Guerrero et al., 2009). One of the most popular fermented dairy products is cheese. In the Mediterranean basin countries, there are numerous traditional brine cheeses made from cow, buffalo, goat or sheep milk. These cheeses are highly valued by consumers in these countries (Abd-El Salam & Benkerroum, 2006). White cheeses are one of the oldest types of cheese that originated from the Middle East and the Mediterranean around 8000 years ago (Huppertz et al., 2006). Among these cheeses is the fresh Libyan white cheese, a traditional cheese found in specific cities in Libya. The cheese is made without the application of brine from cow milk coagulated with rennet enzyme without starter bacteria. This cheese is produced and consumed locally in Libya (El-Gerbi, 1998). The cheese is variously referred to as "Gjibna Bayda" which means "white cheese" in Arabic or "Gjibna Maasouraa" which means "squeezed cheese". Generally, soft white cheese varieties in North Africa have a short shelf-life from 3 to 15 days (Samet-Bali et al., 2009), showing that they are prone to the growth of undesirable microorganisms (Benkerroum, 2013). Several studies have looked into producing probiotic cheeses out of white cheese and survival of probiotic bacteria in cheese (Buriti et al., 2005; Cárdenas et al., 2014; Dabevska-Kostoska et al., 2015; Dantas et al., 2016 and Haddad et al., 2015). In addition, starter cultures are not used for the white cheese made in Libya. This cheese is produced without the intentional inclusion of starter culture in the artisanal cheese production; instead, the indigenous milk flora contributes to ripening. In the processing of white cheese, the use of mixed-strain starter colonies comprising of unknown genera of lactic acid bacteria has been used (Effat et al., 2020; Dabevska-Kostoska et al., 2015 and Yerlikaya & Ozer, 2014). One of the most critical criteria used in assessing customer preference and approval is cheese flavour. This study was based on the hypothesis that Lactic acid bacteria are characterised in their flavour-forming capacity and recognized for their potential to improve some types of cheese

flavour when used as adjunct cultures. One of the dominant lactic acid bacteria (LAB) group used in the cheese starter cultures is the *Lactobacillus* species (Ehsani et al., 2018; Dabevska-Kostoska, 2015). In addition, the mixture of whole cow's milk with varying amounts of whole milk powder can enhance the nutritional value of the resulting cheese while also improving its sensory qualities (Gomah et al., 2019). The development of new or modified fermented foods is greatly anticipated due to the increasing demand for tasty and healthy foods. To the best of our knowledge, no published studies have been reported on the effects of different manufacturing methods on the fresh Libyan white cheese. Based on this, the aim of this study was to evaluate the effect of adding lactic acid bacteria and different percentages of Powdered milk to cow's whole milk in order to reduce whey production during the rennin-induced coagulation process. Additionally, a sensory evaluation of the resulting cheese was conducted and compared with cheese produced by traditional methods using natural.

#### 2. Materials and Methods

#### 2. 1. Starter culture

The commercial freeze-dried starter culture YO-MIX 726 LYO® used in this study was supplied by Danisco (Germany) and contained *Streptococcus thermophilus*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *Bifidobacterium lactis*, and *Lactobacillus acidophilus*.

## 2. 2. Cheese making

Three batches of traditional white cheese with slight modifications were produced following the method of El-Gerbi (1998), along with three batches for each of four additional cheese types, making a total of 15 batches. For each cheese type, 2 liters (2 L) of fresh raw cow's milk were used, amounting to a total of 30 liters. The milk was sourced from a farm in Janzur City and transported to the Department of Food Technology laboratory at the Ghiran Agriculture Institute in jars maintained at 4 °C, where it was stored under refrigeration until analysis and cheese production. Liquid rennet (coagulating power 1:10,000) obtained from Chr. Hansen (Gayrettepe, Kuala Lumpur, Malaysia) was used for milk coagulation, and calcium chloride (ChemAR®) was applied as a coagulant aid. The starter culture was inoculated at a rate of 2% (v/v). Cheese samples were prepared by adding varying ratios of powdered milk to fresh standard cow's milk to increase total solids (TS). The production schemes for the three groups of white cheeses are provided in (Table ) as follows: 0%, 10%, 20%, and 30% powdered milk per liter of cow's milk. The five experimental groups included: Control: cheese made by the traditional method without starter culture or powdered milk; T1: cheese with starter culture and 0% powdered milk; T2: cheese with starter culture and 10% powdered milk; T3: cheese with starter culture and 20% powdered milk; and T4: cheese with starter culture and 30% powdered milk. The

milk was pasteurized at 72 °C for 15 seconds and cooled to 37 °C, then divided into equal portions. A solution of 0.02% (w/v) CaCl<sub>2</sub> (Armadale, Australia) was added to each batch, followed by the addition of 0.5 mL of chymosin per liter of milk to induce coagulation for 45 minutes. After coagulation, the curds were cut into 1 cm<sup>2</sup> cubes, and whey was allowed to drain for 15 minutes. The curds were then covered with cloth, left to drain without pressure for approximately 30 minutes, and subsequently pressed for 60 minutes. Once the whey was completely removed, the curds were transferred to high-density polyethylene (HDPE) molds and stored at 5 °C. Chemical analyses and sensory evaluations of the cheeses were performed after 24 hours of storage.

## 2.3. Physico-chemical Analysis

The cheese samples were analysed for their pH values using a digital pH-meter (pH meter, model pH 537 WTW, Germany). Titratable acidity was expressed as a lactic acid (%) with 0.1 N NaOH. Fat was digested in a Gerber butyrometer. Protein content was determined using the Kjeldahl method by measuring the total nitrogen with conversion to protein content using a conversion factor of 6.38. Moisture was removed in an air oven at 105 °C using a drying oven (WTC binder, Germany). All samples were analysed in triplicate as described by the Standard Methods of Hooi et al. (2004). The yield of the cheese was calculated by the methods described by Sulieman *et al.*, (2012).

#### 2.4. Yield calculation:

The actual yield of Domiati cheese was determined by weighing the cheese after its removal from brine and expressing the result as a percentage of the initial milk weight in the corresponding vat (Gomah et al., 2019). The cheese yield was calculated using the following equation:

Cheese Yield (%) = 
$$\frac{\textit{Weight of Cheese}}{\textit{Weight of cheese Milk}} \times 100$$

All experiments were conducted in triplicate to ensure accuracy and reproducibility.

## 2. 5. Sensory evaluation

The fresh white cheese samples sensory evaluation was carried out at the Department of Food Technology, Department of Food Technology laboratory at the Ghiran Agriculture Institute. The experiment was conducted under controlled conditions of light and environment according to the recommendations set out in ISO standard (ISO 8589, 2007) from 10:00 am to 12:00 pm daily. The cheese samples were cut into 5 cm, tempered for 30 min to 20±2 °C before evaluation, and subsequently presented in plastic plates, coded with random 3-digit numbers to the panelists. For the sensory analysis of the fresh white cheese samples, a 5-point Hedonic scale with expressions such as extremely dislike to extremely like

were used to determine the quality of the cheese samples (i.e., appearance, colour, texture, flavour, and overall acceptability) (Sharif et al., 2017). The sensory evaluation was performed by 30 assessors, eight men and twenty-two women ranging between 19 - 60 years selected from staff and students of the Ghiran Agriculture Institute without previous experience in the sensory evaluation of cheese. However, they were familiar with the sensory evaluation techniques.

## 2. 6. Statistical analysis

The data were presented as the mean  $\pm$  standard division of the mean. All statistical tests were carried out in triplicates that followed the analysis of variance (ANOVA) and Tukey's test if the results obtained were significantly different with a 95 % confidence level (p  $\leq$  0.05) using Minitab statistical software program version 18. Meanwhile, the data on sensory evaluation were analysed by randomized block design (RBD).

#### 3. Results and Discussion

#### 3. 1. Physico-chemical profile

The compositional characteristics of the fresh whole milk and powdered milk are given in Table 2. It was observed that the fat content in powdered milk was higher than in fresh milk, measuring 41.21% and 30.86% on a dry weight basis, respectively. As for protein, the values were 28.40% and 25.87% on a dry weight basis, respectively. The lower fat content in fresh milk may be attributed to poor cow nutrition or seasonal variations, as fat levels are generally higher in winter than in summer.

## 3. 2. pH Values During Soft White Cheese Production

Table 3 presents the pH values recorded throughout the different stages of soft white cheese production. The addition of starter culture was found to lower the pH, indicating increased acidity. Moreover, incorporating powdered milk at varying concentrations further reduced the pH of the soft cheese compared to the control sample.

#### 3. 4. Soft White Cheese Yield:

Cheese yield, a key economic indicator in cheesemaking, reflects the efficiency of cheese production. The results shown in Table 4 and illustrated in Figure 1 indicate that adding powdered milk to fresh cow's milk significantly enhanced the yield of soft white cheese compared to the control samples made with fresh milk alone. The highest yield (74.43%) was obtained from cheese produced with 300 g of powdered milk per liter of fresh cow's milk (30%), compared to only 12.23% for the control sample and lower yields in the other treatments. Whey content followed an opposite trend: the highest whey volume was recorded in the control treatment (0.88 kg) and in cheese made with fresh cow's milk and starter culture (0.83 kg), decreasing progressively to 0.33 kg in treatment T4 (30% powdered

milk). These findings are consistent with the results reported by Hattem and Hassabo (2015) and Gomah et al. (2019). Furthermore, the addition of starter culture alone (T1) improved the yield of soft white cheese by approximately 5% compared to the control made solely from fresh milk.

## 3. 5. Sensory evaluation

The differences in the sensory characteristics among the five types of white cheese on consumers acceptance were recorded in supplementary Table 1. According to the results, there was no significant difference ( $P \ge 0.05$ ) in the appearance, colour, taste and texture compared to the soft cheese (control) with the types of cheeses that adding 10% and 20% powdered milk, as shown in Figure (2). In this case, the addition 10% and 20% powdered milk in fresh white cheese did not affect the appearance, colour and texture compared well with the traditional white cheese. On the other hand, a significant difference ( $P \le 0.05$ ) in the addition 30% powdered milk received the lowest consumer rating. Adding starter culture to the milk without adding powdered milk also received a lower rating due to the acidity and sweetness that formed in the cheese. This indicates that Libyan consumers prefer sensory properties similar to traditional soft white cheese. This evaluation contradicts what Gomah et al. (2019) reported, who stated that increasing the addition of powdered milk improved the cheese's taste, texture, and overall acceptability. Their results indicated that adding 40% powdered milk to the cheese received the highest overall score compared to the other treatments (Figure 3). Haddad et al., (2015) found that the probiotic soft Jordanian white cheese produced with the addition of L. acidophilus and B. lactis to the curd were the least preferred. In this study, the addition starter culture had effect on the overall acceptance. In addition to that, fresh white cheese with starter culture had significant differences (P  $\geq$  0.05) on the overall acceptance rating. Heterofermentative lactobacilli have varietal effects on cheese characteristics including negative, no effect, and positive (Irlinger, et al., 2017). Dantas et al., (2016) reported that the addition of L. casei Zhang for Minas Frescal cheese negatively affected the sensory acceptance, and they suggested that optimization of L. casei Zhang dosage should be performed during the manufacturing. According to the studies on the effects of LAB on white cheese flavour and texture, some studies stated that LAB as a non-starter culture, adjunct or probiotic positively affected cheese flavour, such as L. brevis in Turkish white cheese (Kayagil & Canan, 2009) and soft Moroccan white cheese (Ouadghiri, et al., 2005); L. acidophilus La-5 and L. casei 01 in Coalho cheese (Oliveira, et al., 2012). L. acidophilus La-5 and S. thermophilus improved the sensory stability in Minas fresh cheese (de Paula, et al., 2020; Souza & Saad, 2009) while some L. paracasei strains contributed to the development and production of compounds related to flavour in short-aged cheddar cheese (Stefanovic, et al., 2018). On the other hand, some research stated that adding probiotics did not yield significant difference on cheese flavour and

texture, such as *Lb. paracasei* subsp. *paracasei* LBC 82 in fresh Minas cheese (Buriti et al., 2005); and *L. casei* ATCC 39392 and *L. plantarum* ATCC 8014 in UF-white cheese (Zomorodi et al., 2011).

#### 4. Conclusions

This work demonstrated the adding lactic acid bacteria lowered the pH, improved acidity, and enhanced the overall taste and flavor of the cheese. It also led to a relative improvement in sensory acceptance, but with a slight decrease in texture. Adding powdered milk (at 10%, 20%, and 30%) significantly increased cheese yield, reaching 74.43% at 30%, with some decline in sensory evaluation as the percentage of powdered milk increased, especially at 30%. The best sensory combination was achieved when using 10% powdered milk with the bacterial starter culture, with a balanced taste, texture, and appearance.

**Table 1:** Different treatments for soft white cheese are under study.

Treatment	Fresh milk	Powdered Milk	Starter culture
• Control	✓	*	*
• T1	✓	*	✓
• T2	✓	10%	✓
• T3	✓	20%	✓
• T4	✓	30%	✓

**Table 2:** Physiochemical properties of different types of soft white cheese.

Type of Milk	Total solid (%)	Moisture (%)	Non- Fat total solid (%)	Protein (%)	Fat (%)	Titratable acidity*	pН
Fresh milk	10.6	89.4	8.10	3.01	2.50	0.16	6.62
powdered Milk	96.63	3.37	68.43	25.0	28.20	0.14	6.70

<sup>\* =</sup> Titratable acidity was as percent lactic acid

**Table 3:** Changes in pH values during the stages of different types of white cheese manufacturing.

Type of Treatment	Before adding the starter	After adding the starter	After coagulation	Cheese
Control	6.65	6.52	6.45	6.41
T1	6.55	6.35	5.60	5.30
T2	6.44	6.37	5.99	5.75
T3	6.37	6.35	6.25	6.08
T4	6.31	6.31	6.23	6.10

Control= Traditional White cheese; T1= cheese with starter culture and 0% powdered milk; T2= cheese with starter culture and 10% powdered milk; T3= cheese with starter culture and 20% powdered milk; and T4= cheese with starter culture and 30% powdered milk

**Table 4:** Production rate of soft white cheese using different treatments and ratios of powdered milk.

Type of Treatment	pН	Weight of Cheese Milk (kg)	Weight of Cheese (kg)	Weight of Whey (kg)	Cheese yield (%)
Control	6.55	1.0	0.12	0.88	12.23
T1	6.65	1.0	0.17	0.83	16.97
T2	6.44	1.1	0.30	0.81	26.70
T3	6.37	1.2	0.66	0.54	55.10
T4	6.31	1.3	0.97	0.33	74.43

Control= Traditional White cheese; T1= cheese with starter culture and 0% powdered milk; T2= cheese with starter culture and 10% powdered milk; T3= cheese with starter culture and 20% powdered milk; and T4= cheese with starter culture and 30% powdered milk.

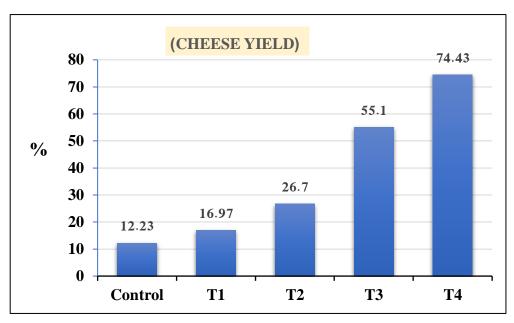
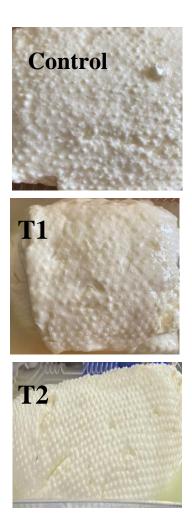


Figure 1: Impact of various treatments on soft cheese yield.







**Figure 3**: Soft white cheese using different treatments and ratios of powdered milk.

## **Supplementary Table 1:**

Sensorial acceptance of white cheese made from cow milk with and without starter culture using different parameters and proportions of powdered milk.

Type of Treatment	Appearance	Colour	Taste	Odor	Texture
Control	$4.53 \pm 0.64^{a}$	± 4.20 1.08a	± 4.13 1.13a	± 4.00 1.13a	± 4.47 0.92a
T1	$2.87 \pm 1.13^{b}$	$\begin{array}{c} 3.27 \pm \\ 1.03^a \end{array}$	$\begin{array}{c} 2.47 \pm \\ 1.25^{\mathrm{b}} \end{array}$	$2.87 \pm 1.25^{a}$	$\begin{array}{c} 2.93 \pm \\ 1.03^{b} \end{array}$
T2	$3.47 \pm 1.19$	$3.67 \pm 1.11^{a}$	$\begin{array}{l} 3.47 \pm \\ 1.19^{ab} \end{array}$	$\begin{array}{c} 3.47 \pm \\ 1.30^a \end{array}$	$\begin{array}{l} 3.60 \pm \\ 1.24^{ab} \end{array}$
Т3	$3.93\pm0.88^{ab}$	$\begin{array}{c} 3.53 \pm \\ 1.06^a \end{array}$	$\begin{array}{l} 3.07 \pm \\ 1.03^{ab} \end{array}$	$\begin{array}{c} 3.00 \pm \\ 1.20^a \end{array}$	$\begin{array}{l} 3.87 \pm \\ 1.06^{ab} \end{array}$
T4	$3.27 \pm 1.34^{b}$	$\begin{array}{c} 3.33 \pm \\ 1.05^a \end{array}$	$2.25 \pm 1.25^{b}$	$\begin{array}{c} 3.80 \pm \\ 0.94^{a} \end{array}$	$\begin{array}{c} 3.20 \pm \\ 1.32^b \end{array}$
P value	0.001 *	0.136	< 0.001 **	0.041	0.004 *

Control= Traditional white cheese; T1= cheese with starter culture and 0% powdered milk; T2= cheese with starter culture and 10% powdered milk; T3= cheese with starter culture and 20% powdered milk; and T4= cheese with starter culture and 30% powdered milk.

Means that do not share in the same column are significantly different (Tukey's Test P<0.05)

P-values  $\leq 0.05$  of each independent variable are statistically significant; StDev = standard deviation of mean

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