

## Diseño, valoración nutricional y sensorial de un queso semiduro usando tres tipos de leche

### Design, nutritional, and sensory evaluation of a semi-hard cheese using three types of milk.

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#### Abstract:

Cheese is one of the most widely consumed dairy products worldwide, and its physicochemical, textural, and sensory properties are strongly influenced by the type of milk used and the ripening process. The aim of this study was to design a semi-hard Mexican Manchego-style cheese using a mixture of cow, goat, and sheep milk and to evaluate its physicochemical composition, proteolysis, texture profile, and sensory acceptance during 180 days of ripening. Cheese was manufactured from a milk mixture composed of 60% cow milk, 25% sheep milk, and 15% goat milk using a standardized cheesemaking process and ripened at 10 °C and 85% relative humidity. During the ripening period, physicochemical parameters, including moisture, protein, fat, ash, water activity, and titratable acidity, were analyzed. Proteolysis was evaluated through the determination of pH 4.6-soluble nitrogen, 12% trichloroacetic acid-soluble nitrogen, and free amino acids. Texture profile analysis (TPA) was performed to determine mechanical properties. Consumer acceptance was evaluated using a hedonic sensory test. Results showed a progressive decrease in moisture and water activity during ripening, accompanied by an increase in ash content and proteolysis-related compounds. Soluble nitrogen fractions and free amino acids increased markedly, indicating intense proteolytic activity throughout maturation. Texture analysis revealed a decrease in hardness during intermediate ripening stages, followed by a slight increase at the end of maturation, while cohesiveness showed a gradual increase. Sensory evaluation indicated that the cheese maintained favorable consumer acceptance throughout the ripening period. These findings demonstrate that mixed milk from cows, goats, and sheep can be successfully used to produce a semi-hard cheese with desirable technological, nutritional, and sensory properties, contributing to the diversification and sustainability of dairy production systems.

#### Keywords:

Manchego style, mixed milk; semi-hard; ripening; texture, sensory acceptance.

#### Resumen:

El queso es uno de los productos lácteos más importantes a nivel mundial, y sus características fisicoquímicas, texturales y sensoriales dependen del tipo de leche utilizada y de las condiciones de maduración. El objetivo de este estudio fue diseñar un queso semiduro tipo Manchego mexicano a partir de una mezcla de leche de vaca, cabra y oveja, y evaluar su composición fisicoquímica, la proteólisis, el perfil de textura y la aceptación sensorial durante 180 días de maduración. El queso se elaboró a partir de una mezcla compuesta

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por 60 % de leche de vaca, 25 % de leche de oveja y 15 % de leche de cabra, empleando un proceso estandarizado de elaboración y maduración a 10 °C y 85% de humedad relativa. Durante el periodo de maduración se analizaron parámetros fisicoquímicos, incluidos la humedad, la proteína, la grasa, las cenizas, la actividad de agua y la acidez titulable. La proteólisis se evaluó mediante la determinación del nitrógeno soluble a pH 4.6, del nitrógeno soluble en ácido tricloroacético al 12% y de los aminoácidos libres. Asimismo, se determinó el perfil de textura mediante análisis de perfil de textura (TPA) y la aceptación sensorial fue evaluada mediante una prueba hedónica con consumidores. Los resultados mostraron una disminución progresiva del contenido de humedad y de la actividad de agua durante la maduración, acompañada de un incremento en la concentración de cenizas y de compuestos derivados de la proteólisis. El nitrógeno soluble y los aminoácidos libres aumentaron significativamente, lo que indica una intensa actividad proteolítica durante el proceso de maduración. En términos de textura, la dureza disminuyó durante las etapas intermedias de maduración y aumentó ligeramente al final del proceso, mientras que la cohesividad aumentó de forma gradual. El análisis sensorial mostró una aceptación favorable del producto durante todo el periodo de maduración. Estos resultados indican que el uso de leche mixta de vaca, cabra y oveja permite obtener un queso semiduro con propiedades tecnológicas, nutricionales y sensoriales adecuadas, lo que además contribuye a la diversificación y sostenibilidad de los sistemas de producción láctea.

**Palabras Clave:**

Queso estilo Manchego, leche mixta; semiduro; maduración; textura; aceptación sensorial.

## 1. Introduction

Cheese is one of the oldest and most widely consumed dairy products worldwide, valued for its high nutritional density, sensory diversity, and technological versatility [1]. Its quality and final characteristics are strongly influenced by the type of milk used, processing conditions, and ripening parameters. Although cow's milk dominates global dairy production, milk from small ruminants such as goats and sheep has gained increasing scientific and industrial interest due to its distinctive nutritional composition and technological behavior in cheese manufacture [2]. Goat's and sheep's milks differ markedly from cow's milk in protein fractions, fat composition, mineral content, and casein micelle structure [3–5]. These compositional differences directly affect coagulation kinetics, curd firmness, whey expulsion, texture development, and sensory perception of cheeses [6]. Sheep's milk is characterized by higher total solids, protein, and fat contents, which contribute to increased cheese yield and firmer curd formation [7,8]. In contrast, goat's milk has lower  $\alpha$ 1-casein content, resulting in a softer texture and improved digestibility, although it may have specific flavor attributes that influence consumer acceptance [2,9,10].

In Mexico, semi-hard cheeses such as Mexican Manchego-type cheese represent an important segment of both artisanal and industrial dairy production [11–13]. Traditionally, this type of cheese is produced from cow's milk; however, previous

studies have demonstrated that goat's and sheep's milks can be successfully used to produce semi-hard cheeses with differentiated physicochemical, textural, and sensory properties [2,8,10,13,14]. Comparative evaluations have shown that cheeses made from different milk types exhibit significant variations in moisture content, protein and fat composition, rheological behavior, melting capacity, and sensory attributes, which are closely associated with milk composition and ripening time [2]. From a sustainability perspective, the use of milk from small ruminants is a relevant strategy to diversify and enhance the resilience of dairy production systems [15]. Goat and sheep farming is commonly associated with low-input production systems and efficient use of natural pastures, particularly in marginal, mountainous, or semi-arid regions where cattle production is limited [16]. The transformation of these milks into value-added products, such as semi-hard cheeses, improves resource use efficiency, reduces post-harvest losses, and strengthens the economic viability of small-scale producers. Additionally, the higher concentrations of protein, fat, and total solids in small ruminant milk contribute to increased cheese yield, optimizing resource use per unit of product and supporting more sustainable dairy processing chains [17].

Despite growing interest in mixed-milk cheeses, most studies have focused on cheeses made from a single species or binary mixtures, and little information is available on the physicochemical, proteolytic, textural, and sensory evolution of cheeses produced from ternary mixtures of cow,

goat, and sheep milk under controlled ripening conditions. There is a lack of integrated studies that simultaneously evaluate compositional changes, proteolysis dynamics, texture development, and consumer acceptance over extended ripening periods in this type of cheese.

Therefore, the aim of this study was to design a semi-hard Mexican Manchego-style cheese using a mixture of cow's, goat's, and sheep's milk, and to evaluate its physicochemical composition, proteolysis, texture profile, and sensory acceptance during 180 days of ripening under controlled processing conditions. This study seeks to advance understanding of mixed-milk cheese systems and provide scientific evidence for their technological feasibility and potential role in sustainable dairy production.

## 2. Materials and Methods

### 2.1. Experimental Design

The study was conducted under a completely randomized design to evaluate the physicochemical, proteolytic, textural, and sensory characteristics of Mexican Manchego-style cheese made from a mixture of cow's, goat's, and sheep's milk.

Three independent cheesemaking batches (biological replicates) were produced, and analyses were performed in triplicate (analytical replicates) at each sampling time (1, 30, 60, 90, and 120). Ripening time was considered a fixed factor.

### 2.2. Experimental Site

The experiment was conducted in the laboratories of the Food Science and Technology Research Center and the Institute of Agricultural Sciences of the Autonomous University of the State of Hidalgo (UAEH), Mexico.

### 2.3. Raw Materials

Milk from three mammalian species was used for cheesemaking. Cow's milk was obtained from healthy animals maintained at the University Ranch of the Institute of Agricultural Sciences (UAEH). Goat milk was provided by local producers in Ixmiquilpan,

Hidalgo, from Creole goats raised under traditional production systems in the Mezquital Valley. Sheep milk from East Friesian sheep was obtained from a farm located in Atotonilco el Grande, Hidalgo.

All animals were clinically healthy and in similar stages of lactation. Milk was collected under hygienic conditions, transported under refrigeration (4 °C), and analyzed prior to processing to determine its physicochemical composition.

### 2.4. Cheese-making Procedure

Mexican Manchego-style cheeses were manufactured following the method described by Lobato-Calleros et al. [18], with minor modifications. For each batch, 100 kg of milk was processed. A ternary mixture was prepared as follows: 60% cow's milk, 25% sheep's milk, and 15% goat's milk. This formulation was selected to balance the technological properties of bovine milk with the higher protein and fat contents of sheep milk and the functional characteristics of goat milk.

The milk mixture was filtered, pasteurized at 63 °C for 30 min, and cooled to 35 °C. A commercial starter culture composed of *Lactococcus lactis* and *Lactococcus lactis* subsp. *cremoris* (French Bioprox M195, Mexico) was added at a rate of 20 g/100 kg of milk and allowed to ripen for 30 min.

A calcium chloride solution (6% w/v) was added at a rate of 333 mL/100 kg of milk and equilibrated for 5 min. Rennet (Cuamix, 280 IMCU/mL; Cuamex, Jalisco, Mexico), diluted in potable water (1:10), was added at 10 mL/100 kg of milk. After coagulation ( $\approx$ 30 min), the curd was cut into 1 cm cubes and allowed to heal for 5 min.

The curd was stirred for 10 min and gradually heated to 42 °C at a rate of 1 °C every 3 min. This temperature was maintained for 30 min during whey drainage. The curd was then dry-salted with 440 g of sodium chloride per batch and equilibrated for 20 min. Curd was transferred into stainless steel molds (1 kg capacity), pressed for 24 h at 20 °C, vacuum-packaged, and ripened at 10 °C and 85% relative humidity for 120 days.

### 2.5. Chemical Composition and Proteolysis

Cheese samples were analyzed for moisture (AOAC 948.12), fat (Gerber method; AOAC 933.05), protein

(Kjeldahl method; AOAC 991.20), and ash (AOAC 935.42) [19].

Water activity (aw) was measured at 21 °C using a hygrometer (Aqualab CX-2 Dew Point; Decagon Devices Inc., Pullman, WA, USA). Titratable acidity was determined by titration with 0.1 N NaOH and expressed as lactic acid percentage. pH was measured at 25 °C using a homogenized slurry (10 g cheese + 10 mL deionized water).

Proteolysis was evaluated by measuring pH 4.6-soluble nitrogen, 12% trichloroacetic acid (TCA)-soluble nitrogen, and total free amino acids, as described by Folkertsma and Fox [20].

## 2.6. Texture Analysis

Texture profile analysis (TPA) was performed using a CT3 texture analyzer (Brookfield Engineering Laboratories, Middleboro, MA, USA) equipped with a cylindrical probe (TA4/1000).

Cheese samples were cut into cubes (1 cm<sup>3</sup>) and equilibrated at 20 °C for 30 min. Samples were subjected to two compression cycles with a 1 s interval at a crosshead speed of 1 mm/s and a deformation of 50%. The following parameters were calculated: hardness, cohesiveness, springiness, resilience, adhesiveness, and chewiness [21].

## 2.7. Sensory Evaluation

Sensory evaluation was conducted at 1, 90, and 180 days of ripening using a consumer acceptance test.

A total of 75 untrained consumers (38 females and 37 males; age range 20–30 years) participated in the study. Cheese samples (2 × 2 × 2 cm) were tempered at 12 °C, coded with random three-digit numbers, and presented in randomized order.

Panelists evaluated appearance, aroma, flavor, texture, and overall acceptability using a nine-point hedonic scale (1 = extremely dislike; 9 = extremely like). All participants provided informed consent prior to evaluation.

## 2.8 Statistical analysis

Data were analyzed using analysis of variance (ANOVA) under a completely randomized design, considering ripening time as a fixed factor. When significant differences were detected ( $p < 0.05$ ),

Tukey's multiple comparison test was applied. Assumptions of normality and homogeneity of variance were verified prior to analysis. Statistical analyses were performed using SAS software (version 9.4; SAS Institute Inc., Cary, NC, USA).

## 3. Results and discussion

### 3.1 Milk composition

The compositional analysis of the milks used for cheesemaking revealed significant differences among species, particularly in fat, protein, and total solids content (Table 1). Sheep milk exhibited the highest fat (4.93%) and protein (4.86%) contents, as well as the greatest total solids (14.09%), confirming its well-documented suitability for cheesemaking due to its contribution to higher yield and firmer curd formation [22,23].

Table 1. Composition of milk samples and the mixture used in the cheesemaking of Manchego-style cheese

Milk type	Fat (%)	Protein (%)	Moisture (%)	Ash (%)	Total solids (%)	Carbohydrates (%)
Cow	3.40 ± 0.08	2.65 ± 0.05	88.86 ± 0.20	0.67 ± 0.02	11.13 ± 0.18	4.42 ± 0.07
Goat	4.00 ± 0.09	3.39 ± 0.06	87.66 ± 0.22	0.79 ± 0.02	12.37 ± 0.20	4.16 ± 0.06
Sheep	4.93 ± 0.10	4.86 ± 0.07	85.90 ± 0.25	0.85 ± 0.03	14.09 ± 0.23	3.46 ± 0.06
Mixed	4.83 ± 0.09	2.65 ± 0.05	88.34 ± 0.21	0.64 ± 0.02	11.65 ± 0.19	3.54 ± 0.07

Values are expressed as mean ± standard deviation of three experimental replicates

Goat milk showed intermediate values of fat and protein, higher than those of cow milk but lower than those of sheep milk. These results are consistent with previous comparative studies [24], and reflect structural characteristics such as smaller fat globules and differences in casein fractions, which can influence curd formation and texture development. Cow milk had the lowest protein and total solids contents, which may negatively affect cheese yield compared with small ruminant milk [25]. The mixed milk composition reflected the proportional contributions of each species, suggesting that the final cheese characteristics result from a combined technological effect. Previous studies have reported that mixed-milk systems can enhance functional and sensory

properties by integrating compositional advantages of different species [8,26,27].

Overall, these compositional differences are expected to play a key role in determining the cheese's physicochemical evolution, texture development, and sensory properties during ripening.

### 3.2 Cheese composition

The physicochemical composition of the Mexican Manchego-style cheese produced from mixed milk showed significant changes throughout ripening (Table 2), reflecting typical biochemical and physical transformations associated with cheese maturation. Moisture content decreased from 44.39% on day 1 to 41.22% on day 180 ( $p < 0.001$ ), primarily due to progressive water loss and structural rearrangement of the casein matrix. This behavior is characteristic of semi-hard cheeses and contributes to increased firmness and solids concentration [23,25,28].

Table 2. Physicochemical composition of the Mexican Manchego-style cheese made from a mixture of cow, goat, and sheep milk changes during ripening

Ripening time (days)	pH	Lactic acid (%)	SN/TN (%)	TCA SN/TN (%)	Free amino acids (mg Leu/100 g protein)
1	6.02 ± 0.03	0.85 ± 0.02	8.0 ± 0.5	2.0 ± 0.2	48 ± 10
30	5.45 ± 0.04	0.90 ± 0.03	16.0 ± 0.7	4.0 ± 0.3	83 ± 15
60	5.25 ± 0.03	0.95 ± 0.03	18.0 ± 0.8	5.0 ± 0.4	574 ± 60
90	5.08 ± 0.05	1.00 ± 0.04	21.0 ± 0.9	6.0 ± 0.5	825 ± 80
120	5.20 ± 0.04	1.15 ± 0.05	22.0 ± 1.0	7.0 ± 0.6	998 ± 90
150	5.20 ± 0.03	1.30 ± 0.05	28.0 ± 1.2	9.0 ± 0.7	1187 ± 100
180	5.05 ± 0.04	1.40 ± 0.06	29.0 ± 1.3	9.5 ± 0.8	1296 ± 110

Abbreviations are: MNFS, moisture in the non-fat substance; FDM, fat content on a dry basis weight; S/M, salt in the moisture phase of the cheese. Data are means of three replicate trials; means within a row with different superscripts show significant ( $p < 0.05$ ) differences between ripening times

Ash content increased from 3.6% to 5.35%, indicating mineral concentration as moisture decreased. Simultaneously, water activity declined from 0.93 to 0.90, suggesting reduced water availability and improved microbiological stability, a common feature in ripened cheeses [25,29,30].

The pH remained relatively stable (5.25–5.50), attributable to the buffering capacity of peptides and

amino acids released during proteolysis. Similar patterns have been reported in semi-hard and mixed-milk cheeses, where initial acidification is followed by stabilization due to biochemical transformations during ripening [23,36]. Proteolysis progressed significantly, as evidenced by the increase in pH 4.6-soluble nitrogen from 8% to 29% of total nitrogen. This fraction reflects the breakdown of caseins into soluble peptides, mainly due to residual rennet activity and microbial enzymes. Comparable increases have been reported in ripened cheeses, confirming proteolysis as a key biochemical process during maturation. Free amino acids increased markedly from 48 to 1296 mg Leu/100 g protein, indicating advanced protein degradation. This accumulation is directly related to flavor development, as amino acids serve as precursors of volatile compounds.

These results demonstrate that the evolution of physicochemical properties in mixed-milk cheese is governed by the combined effects of moisture loss, enzymatic activity, and mineral concentration, which together define the structural and biochemical characteristics of the final product.

### 3.3 Texture profile

Texture profile analysis revealed progressive structural modifications during cheese ripening. Hardness decreased from the initial stage to the intermediate ripening stage and increased slightly at the final stage. This behavior reflects the dynamic balance between proteolysis-induced softening and moisture loss-induced firming. See Table 3.

Proteolysis plays a key role in the weakening of the casein matrix through the enzymatic hydrolysis of  $\alpha$ - and  $\beta$ -caseins by residual coagulant enzymes, indigenous milk proteases, and enzymes produced by lactic acid bacteria. This degradation reduces structural integrity and results in softer textures during the early ripening stages [23,37]. Similar reductions in hardness during the initial stages of maturation have been reported in semi-hard cheeses, including Manchego-type and mixed-milk cheeses [23].

The slight increase in hardness observed at advanced ripening stages can be attributed to moisture loss and compaction of the protein matrix. As water content decreases, the casein network becomes more compact, increasing mechanical resistance. This phenomenon has been widely described in aged cheeses and is associated with dehydration processes that partially counteract proteolysis [25,28].

Cohesiveness showed a slight increase throughout ripening, suggesting progressive structural reorganization within the protein matrix. The peptides generated during proteolysis may interact with casein structures, contributing to internal cohesion and matrix stabilization [37]. Similar behavior has been observed in traditional ripened cheeses, where biochemical changes influence mechanical properties. Chewiness followed a trend similar to hardness, as expected, since this parameter depends on hardness, cohesiveness, and elasticity. The reduction observed during intermediate ripening stages reflects the weakening of the casein network, whereas the subsequent increase is associated with moisture loss and structural consolidation of the matrix. Although some textural parameters did not show statistically significant differences ( $p > 0.05$ ), the observed trends are consistent with previously reported mechanisms describing the evolution of cheese texture during ripening. These results confirm that both proteolysis and water redistribution are key factors influencing the mechanical properties and final texture of mixed-milk cheeses.

### 3.4 Sensory acceptance during ripening

Sensory evaluation showed that consumer acceptance of the Mexican Manchego-style cheese made from a mixture of cow, goat, and sheep milk remained stable throughout the ripening period. The mean hedonic scores for overall acceptability were consistently around 6 on the nine-point scale, indicating moderate to high consumer acceptance across all evaluated stages (1, 90, and 120 days of ripening).

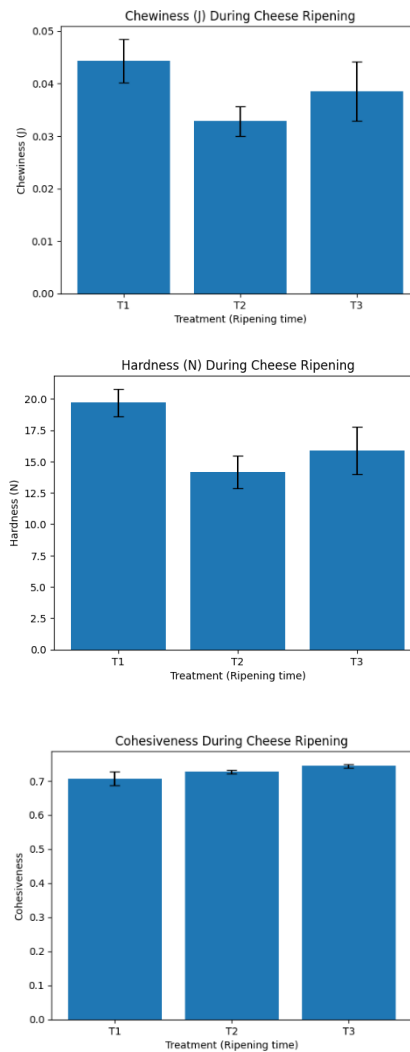


Figure 1. Boxplots of texture profile analysis (TPA) parameters of Mexican Manchego-style cheese made from a mixture of cow, goat, and sheep milk during ripening.

At the early stage of ripening (day 1), sensory scores exhibited low variability, suggesting a more uniform perception among consumers. This behavior is likely associated with the mild flavor profile and softer texture characteristic of fresh or minimally ripened cheeses. As ripening progressed, variability in sensory scores increased, particularly at 120 days. This trend indicates greater heterogeneity in consumer perception, which can be attributed to the development of more complex and intense flavor profiles. The biochemical processes occurring during ripening, particularly proteolysis

Table 3. Texture profile analysis (TPA) parameters of Mexican Manchego-style cheese made from a mixture of cow, goat, and sheep milk during ripening.

Treatment (days)	C1 Hardness (N)	Adhesiveness (J)	Resilience	Fracturability (N)	C2 Hardness (N)	Cohesiveness	Elasticity (mm)	Firmness (N)	Chewiness (J)
T1 (1)	19.69 ± 1.11 <sup>a</sup>	0.00006 ± 0.00006 <sup>a</sup>	0.33 ± 0.01 <sup>a</sup>	19.69 ± 1.11 <sup>a</sup>	17.40 ± 1.04 <sup>a</sup>	0.71 ± 0.02 <sup>a</sup>	3.18 ± 0.06 <sup>a</sup>	13.95 ± 1.04 <sup>a</sup>	0.044 ± 0.004 <sup>a</sup>
T2 (90)	14.17 ± 1.32 <sup>a</sup>	0.00006 ± 0.00005 <sup>a</sup>	0.36 ± 0.02 <sup>a</sup>	14.17 ± 1.32 <sup>a</sup>	12.75 ± 1.11 <sup>a</sup>	0.73 ± 0.01 <sup>a</sup>	3.20 ± 0.03 <sup>a</sup>	10.27 ± 0.88 <sup>a</sup>	0.033 ± 0.003 <sup>a</sup>
T3 (120)	15.86 ± 1.88 <sup>a</sup>	0.00006 ± 0.00003 <sup>a</sup>	0.35 ± 0.01 <sup>a</sup>	15.86 ± 1.88 <sup>a</sup>	14.23 ± 1.51 <sup>a</sup>	0.74 ± 0.01 <sup>a</sup>	3.25 ± 0.09 <sup>a</sup>	11.81 ± 1.44 <sup>a</sup>	0.039 ± 0.006 <sup>a</sup>

Values are expressed as mean ± standard deviation (n = 3)

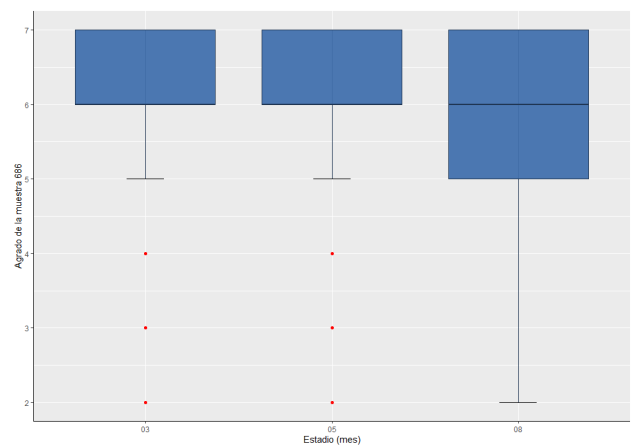
Different superscript letters within the same column indicate significant differences among ripening times (p < 0.05), according to Tukey's test. Hardness: maximum force during first compression; Cohesiveness: ratio of the area under the second compression to the first; Springiness: height recovery after first compression; Resilience: ratio of upstroke to downstroke energy during the first compression; Adhesiveness: negative work required to detach the probe; Chewiness: product of hardness × cohesiveness × springiness.

and lipolysis, lead to the accumulation of peptides, free amino acids, and volatile compounds that contribute to flavor and aroma development [23,37].

The increase in flavor intensity during extended ripening may lead to divergent consumer responses, with some individuals valuing the complexity of aged cheeses, while others perceive these characteristics as too strong. Similar patterns have been reported in cheeses made from goat and sheep milk, where intermediate ripening stages often correspond to optimal sensory acceptance [23,37].

Despite the observed variability at advanced stages, overall acceptability remained within the positive range throughout the ripening period, indicating that incorporating milk from different species did not negatively affect consumer perception. On the contrary, the combination of cow, goat, and sheep milk may contribute to a more balanced sensory profile by integrating complementary flavor and texture attributes.

These results suggest that mixed-milk cheeses can achieve consistent consumer acceptance during ripening, supporting their potential as differentiated dairy products with both technological and sensory advantages



Boxplots represent the distribution of overall acceptability scores obtained from a consumer hedonic test at different ripening times (1, 90, and 120 days). The central line within each box indicates the median, the box limits represent the interquartile range (25th–75th percentiles), and whiskers indicate the minimum and maximum values. Outliers are shown as individual points.

Scores were based on a nine-point hedonic scale (1 = extremely dislike; 9 = extremely like). Different letters indicate significant differences among ripening times (p < 0.05), according to Tukey's multiple comparison test.

Figure 2. Boxplot of overall sensory acceptability scores of Mexican Manchego-style cheeses made from a mixture of cow, goat, and sheep milk during ripening

#### 4. Conclusions

The results of this study demonstrate that producing a semi-hard Mexican Manchego-style cheese from a mixture of cow, goat, and sheep milk is technologically feasible and yields a product with favorable physicochemical, textural, and sensory characteristics. The compositional differences among the milks used in the formulation, particularly the higher fat and protein contents of sheep milk, contributed to improved technological properties of the cheese matrix and supported curd formation and structural development during ripening. These results are consistent with previous studies indicating that milk from small ruminants can enhance cheese yield and modify texture and flavor attributes.

During the 180-day ripening period, significant physicochemical and biochemical changes were observed. Moisture content decreased gradually, while ash content increased and water activity declined, reflecting progressive dehydration and mineral concentration within the cheese matrix. In parallel, the pH remained relatively stable and lactic acid content increased, indicating sustained metabolic activity of lactic acid bacteria throughout ripening.

Proteolysis was one of the most important biochemical processes observed during maturation. The increase in pH-4.6 soluble nitrogen, TCA-soluble nitrogen, and free amino acids confirmed the progressive degradation of caseins and peptides, which plays a key role in the development of flavor and aroma compounds in ripened cheeses. These biochemical transformations are characteristic of semi-hard cheeses and contribute to the product's final sensory profile.

Texture profile analysis revealed moderate structural modifications during ripening. Hardness decreased during the intermediate stage of maturation and increased slightly after prolonged ripening, a pattern associated with the combined effects of proteolysis and moisture loss. Cohesiveness increased slightly over time, suggesting structural reorganization of the casein network as ripening progressed.

Sensory evaluation indicated that the cheese maintained good consumer acceptance throughout the ripening period. Median liking scores remained within the positive range of the hedonic scale, suggesting that the maturation process did not adversely affect the product's sensory quality.

Although variability among individual responses was observed at later ripening stages, the overall acceptance remained favorable.

In conclusion, the use of mixed milk from cow, goat, and sheep represents a promising strategy for developing differentiated semi-hard cheeses with desirable technological and sensory properties. Furthermore, incorporating milk from small ruminants may contribute to the diversification and sustainability of dairy production systems, particularly in regions where goat and sheep farming plays an important economic role.

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#### Conflict of interest

The authors declare that they have no conflicts of interest.

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