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Pepsin extracted from the proventriculi of geese raised under different fattening techniques and its effect on microbiological quality and cheese production

Pepsina extraída de proventrículos de gansos manejados sob diferentes técnicas de engorda e seu efeito na qualidade microbiológica e fabricação de queijos

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ABSTRACT

Goose pepsin is critically important in cheese coagulation, both in terms of variety and economics. This study was conducted to investigate the potential use of the pepsin enzyme obtained from the proventriculi of geese reared under intensive and extensive fattening systems as a milk coagulant, and to evaluate its effect on the quality of white brined cheese. A total of 10 groups of cheeses (2 feeding systems × 5 levels of enzyme concentration) were produced using pepsin derived from intensively and extensively reared geese, together with chymosin (calf rennet) in the following ratios: 100% chymosin, 50% chymosin+50% pepsin, 75% chymosin+25% pepsin, 25% chymosin+75% pepsin, and 100% goose pepsin. Samples were stored at 4±1°C and chemically and microbiologically analyzed on days 0, 7, 15, 30, 45, 60, 90, and 120 of storage. During storage, a significant decrease was observed in other bacterial counts, while an increase was noted in the counts of psychrotrophic, lipolytic, and proteolytic microorganisms (P<0.05). No *E. coli* growth was detected (<1.0 log CFU/g). Differences in microbiological counts between enzyme groups and storage days were statistically significant (P<0.05). Notably, the groups containing 100% goose pepsin exhibited superior microbiological quality parameters (P<0.05). In terms of chemical parameters, apparent pH decreased during storage, while percentage of lactic acid (% l.a.) and salt content increased (P<0.05). However, no significant differences in apparent pH, acidity, or salt were observed among the experimental groups, regardless of feeding system or enzyme combination (P>0.05).

Index terms: Intensive-extensive handling; quality parameters; white brined cheese.

RESUMO

A pepsina de ganso é de importância crucial na coagulação do queijo, tanto em termos de variedade quanto de economia. Este estudo foi conduzido para investigar o potencial de uso da enzima pepsina, obtida do proventrículo de gansos criados em sistemas de engorda intensivos e extensivos, como coagulante do leite, e para avaliar seu efeito na qualidade do queijo branco em salmoura. Um total de 10 grupos de queijos [2 sistemas de alimentação × 5 níveis de concentração da enzima] foram produzidos utilizando pepsina derivada de gansos criados intensiva e extensivamente, juntamente com quimosina (coalho de vitelo). As amostras foram armazenadas a 4±1°C e analisadas química e microbiologicamente nos dias 0, 7, 15, 30, 45, 60, 90 e 120 de armazenamento. Durante o armazenamento, observou-se uma diminuição significativa na contagem de outras bactérias, enquanto houve um aumento na contagem de microrganismos psicrotróficos, lipolíticos e proteolíticos (P<0.05). Não foi detectado crescimento de *E. coli* (<1.0 log CFU/g). As diferenças nas contagens microbiológicas entre os grupos enzimáticos e os dias de armazenamento foram estatisticamente significativas (P<0.05). Notavelmente, os grupos contendo 100% de pepsina de ganso apresentaram parâmetros de qualidade microbiológica superiores (P<0.05). Em termos de parâmetros químicos, o pH aparente diminuiu durante o armazenamento, enquanto a porcentagem de ácido lático (%l.a.) e o teor de sal aumentaram (P<0.05). No entanto, não foram observadas diferenças significativas no pH aparente, na acidez ou no teor de sal entre os grupos experimentais, independentemente do sistema de alimentação ou da combinação de enzimas (P>0.05).

Palavras-chave: Manejo intensivo-extensivo; parâmetros de qualidade; queijo branco em salmoura.

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Introduction

The history of brined cheese dates back approximately 8.000 years in the Middle East and Mediterranean regions (Zareie et al., 2025). White cheese, widely produced and consumed in Balkan and Middle Eastern countries such as Turkey, Egypt, and Greece, is among the most diverse types of cheese worldwide. It is a salty cheese made from cow, sheep, or goat milk or their mixtures and matured in brine. Turkish-type white cheese is typically matured for 1-3 months at temperatures between 4 and 8 °C. It is characterized by its brined texture, white to white-yellow color, hard to semi-hard texture, slightly acidic and salty taste, and lack of rind formation on the surface. This cheese has a soft texture when fresh but is classified as semi-hard or semi-soft after

brine ripening (Hayaloglu, Güven, & Fox, 2002; Dinkci, 2020; Homayouni et al., 2020; Özcan, Artık, & Aloğlu, 2020; Soltani et al., 2022; Uğur & Öner, 2023; Erkaya Kotan & Hayaloglu, 2024).

Cheese production involves several stages, including coagulation, acidification, curd cutting, salting, molding, and ripening (Bennett & Johnston, 2004). Proteolytic enzymes (enzymatic coagulation) and organic acids (acid coagulation) are used in the coagulation step (Kaya & Patır, 2019). These enzymes are also referred to as coagulants, rennets, or proteases (Vapur, 2021). Beyond curd formation, enzymes play a critical role in cheese maturation, influencing yield, texture, and flavor (Lee & Warthesen, 2010). Animal-derived enzymes include chymosin, pepsin, trypsin, and chymotrypsin (Dervişoğlu, Aydemir, & Yazıcı, 2007). The preparation of animal rennets involves collecting and processing of the abomasum (including preparation, extraction, and activation), determining the chymosin-to-pepsin ratio, purification, standardization, and microbiological and chemical quality control (Oštarić et al., 2022). Trypsin and chymotrypsin are associated with high proteolytic activity and the production of bitter peptides in cheese (Dervişoğlu, Aydemir, & Yazıcı, 2007). Microbial rennet is the most widely used alternative to calf chymosin worldwide due to its cost-effectiveness. However, it may cause undesirable characteristics such as poor melting and bitterness (Fox et al., 2004; Horne & Lucey, 2017; Vapur, 2021). Chymosin, the primary coagulant used in cheesemaking, is an aspartic protease enzyme obtained from the abomasum of young calves (Oštarić et al., 2022; Rocchetti et al., 2024). It specifically cleaves the κ-casein protein in milk, thereby accelerating clot formation (Costabel et al., 2015). Chymosin, along with pepsin and lipase, can also be obtained from other animals such as lambs, kids, buffalo, and camels (Liu et al., 2021; Oštarić et al., 2022).

The chymosin/pepsin ratio in the abomasum varies depending on the animal's age and feeding regimen (Uniacke-Lowe & Fox, 2017). Factors such as age, sex, and diet influence the ratio and quantity of these enzymes in rennet (Rolet-Repecaud et al., 2017). In standard calf rennet, the chymosin-to-pepsin ratio is typically 80:20 (Yegin & Dekker, 2013).

The rising demand for cheese production has increased the need for coagulants. However, several factors such as the long time required to raise calves, low litter size, higher costs compared to poultry, increasing calf prices, religious restrictions (e.g., in India), dietary preferences (e.g., veganism favoring plant-based coagulants), and consumer desire for diverse organoleptic properties have limited the use of chymosin and led researchers to explore alternative coagulant sources (Ahmed, 2021; Anusha, Singh, & Bindhu, 2014; Coşkun & Akgündüz, 2020; Vapur, 2021; Wehaidy et al., 2023). Nevertheless, most plant-derived enzymes used in cheesemaking are not preferred due to their low yield and tendency to produce bitterness and flavor defects (Afsharnezhad, Shahangian, & Sariri, 2019).

Pepsin is an aspartic protease secreted by the gastric mucosa's parietal cells and is the primary enzyme in gastric juice, playing a key role in food digestion. These endopeptidases, adapted to the stomach's acidic environment (pH 1.5-2.0), are activated by hydrochloric acid secretion (Kageyama, 2002; Li et al., 2024; Kabir et al., 2025). In cheese, pepsin's strong proteolytic activity facilitates the breakdown of residual proteins during ripening, thus influencing flavor and texture (Mir Khan & Selamoglu, 2020).

Pepsin and chymosin are enzymes that initiate milk coagulation and micelle formation by hydrolyzing the Phe_{105}-Met_{106} peptide bond in κ-casein; however, their proteolytic properties differ (Yang et al., 2022). Chymosin promotes clot formation through targeted bond cleavage and has a higher milk coagulation/proteolytic activity ratio. In contrast, pepsin exhibits a broader proteolytic activity, particularly hydrolyzing peptide bonds containing aromatic residues such as phenylalanine, tyrosine, and tryptophan, as well as some other amino acid residues (Li et al., 2025).

Because of the anatomical, physiological, and biochemical differences in stomach structure between animals raised under intensive and extensive feeding systems, as well as variations in diet composition and feed particle size, pepsin activity may be affected by differences in gastric function. As a result, enzymes produced under different feeding systems may contribute to cheeses with varying aromatic characteristics (Tüzün & Çiftçi, 2010).

The goose is a species suitable for both intensive and extensive rearing systems, and its proventriculus contains the pepsin enzyme. This study is the first to isolate goose pepsin using gel filtration and to evaluate its potential as a milk coagulant. Additionally, the effects of different feeding systems (intensive vs. extensive) on cheese quality are examined, representing a novel contribution to the literature. Accordingly, the aim of this study aimed to evaluate the feasibility of using pepsin derived from the proventriculi of geese reared under different fattening systems as a coagulant in white brined cheese production and to evaluate its effects on selected microbiological and chemical quality parameters.

Material and Methods

Ethical approval

This study was conducted in Elazig province with the approval of the Firat University Non-Interventional Research Ethics Committee, dated 26 May 2022 (Protocol No. 2022/07-03).

Procurement of stomachs (proventriculus)

In this study, redundancy reared and raised mean the same on a private farm in the Kovancilar district of Elazig province (in Türkiye), and geese reared under extensive (pasture-based) conditions were raised and slaughtered in the Sivrice district of Elazig province (in

Türkiye). In total, 14 forestomachs (proventriculus), 7 from each group, were extracted from 10-week-old male geese with complete flora development for pepsin extraction.

The average daily feed intake for intensive feeding was 500 g/head. The mixed feed consists of corn grits (45%), barley (15%), soybean meal (20%), sunflower meal (7%), wheat bran (5%), vegetable oil (2%), limestone (1.2%), dicalcium phosphate (1.5%), salt (0.3%), and vitamin-mineral premix (3%). The nutrient composition of this ration was calculated to be 19.2% crude protein, approximately 2.850 kcal/kg of metabolic energy, 4.8% crude fiber, 6.2% crude ash, 1.1% calcium, and 0.7% phosphorus.

The average daily feed intake for extensive feeding was 340 g/head. The ration composition includes corn grits (30%), barley (20%), wheat (15%), soybean meal (12%), alfalfa meal (10%), sunflower meal (5%), limestone (1.5%), dicalcium phosphate (1.5%), salt (0.3%), and vitamin-mineral premixes (2%). In addition, geese consume 400-600 g of fresh grass and aquatic plants daily. The nutrient content of this ration was determined as 16.0% crude protein, approximately 2.600 kcal/kg metabolic energy, 7.2% crude fiber, 7.0% crude ash, 0.9% calcium, and 0.6% phosphorus.

Processes for extracting, diluting, pH correction, and purification of pepsin from goose proventriculi

Stomach samples were cleaned of contents and surface fats, rinsed with distilled water, placed in plastic bags, and stored at -20 °C. Extraction was performed according to El-Beltagy et al. (2004). The Berridge substrate was prepared following the methods described by Berridge (1952), and enzyme purification was conducted as described by Temiz et al. (2007).

Determination of the coagulating activity (yeast strength)

One unit of milk-clotting activity is defined as the volume of milk (at 35 °C) coagulated by one unit of enzyme within 40 minutes (Anusha, Singh, & Bindhu, 2014). A tenfold (1:10) dilution of the enzyme was used, and this dilution factor was considered in the following equation according to Equation 1, presented below:

$$U = (2400 / T) \times (S / E) \quad (1)$$

U, yeast strength; T, time to clot formation [s]; S, milk quantity [mL]; E, volume of enzyme [mL]

Cheese samples and experimental design

Raw cow's milk used for white brined cheese production was sourced from the Fırat University Agricultural and Animal Research and Application Center (TAHAM). Cheesemaking was performed following the procedure of Hayaloğlu, Güven and Fox (2002) (Figure 1).

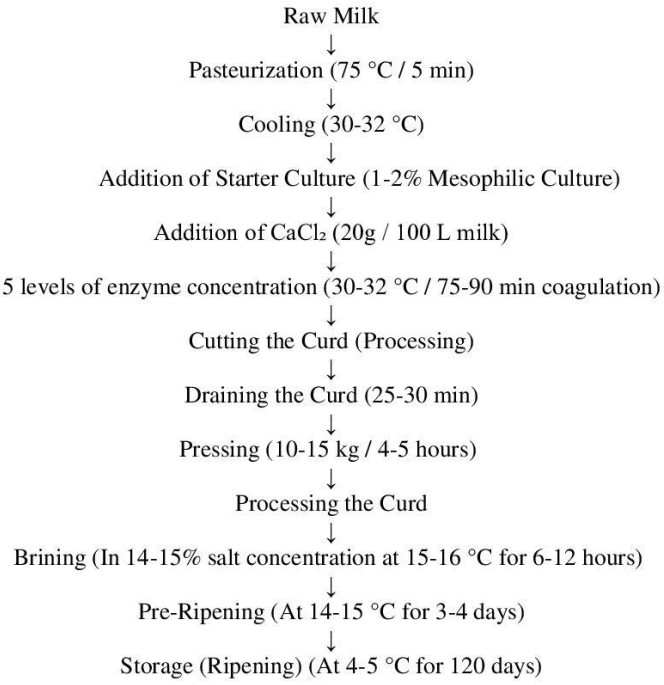

Figure 1: Stages of brined white cheese production.

After the milk was pasteurized, inoculated with *Lactococcus lactis* subsp. *lactis* + *Lactococcus lactis* subsp. *cremoris* (CM 102-10U, Maysa MyStarter Culture, Istanbul, Turkey), and processed into white brined cheese. Coagulation was performed using calf chymosin (RENMAX 600 L, Mayasan Biotech, Istanbul, Turkey). The experimental cheeses were divided into two main groups based on the enzyme source: proventricular extract from intensively fed geese and from extensively fed geese. Each main group was further subdivided into mixtures, in five (5) different percentage proportions, of chymosin and pepsin, considering the sample with 100% chymosin as the control treatment (reference treatment in the studies): 100% chymosin, 50% chymosin + 50% goose pepsin, 75% chymosin + 25% goose pepsin, 25% chymosin + 75% goose pepsin, and 100% goose pepsin. Analyses were conducted using a double parallel design with three replicates (Table 1). Each group utilized 8.4 liters of milk. Cheese blocks (approximately 150 g each, two per package) were placed in 500 mL leak-proof, transparent PET containers (120 mm×155 mm×90 mm; Eva 500 PY-B, Turkey). A 14%-15% brine solution was prepared by dissolving iodine-free rock salt (Hazar, Turkey) in tap water, followed by boiling and cooling. Approximately 250 mL of 14% salt solution was added to each container, at 4±1 °C throughout the experiment and subjected to chemically and microbiologically analyses on storage days 0, 7, 15, 30, 60, 90, and 120.

Microbiological analysis

Cheese samples were aseptically collected; 25 g of each sample was placed into sterile stomacher bags with 225 mL of 0.1% sterile

peptone water (NCM0096A, Neogen, UK) and homogenized using a stomacher (Bag Mixer® 400, Interscience, France), producing a 10^{-1} dilution. After microbiological cultivation, only plates containing 30-300 colonies were counted (USDA/FSIS, 2011). Results were expressed as $\log_{10}$ CFU/g or log CFU/g.

Table 1: Experimental design (pasteurized cow milk: 8.4 liters + rennet + goose pepsin)

Feeding	Experimental groups	Treatments (%)	
		Calf chymosin	Goose pepsin
Intensive	G1	100	0
	G2	50	50
	G3	75	25
	G4	25	75
	G5	0	100
Extensive	G6	100	0
	G7	50	50
	G8	75	25
	G9	25	75
	G10	0	100

Plate Count Agar (PCA) (NCM0010A, Neogen, UK) was used to determine total aerobic mesophilic bacteria (TAMB), with incubation at 35±1 °C for 48 hours (United States Department of Agriculture - USDA, 2015). The same medium was used for psychrotrophic bacteria counts, incubated at 5-7°C for 10 days (USDA, 2011). Man, Rogosa, and Sharpe (MRS) Agar (NCM0190A, Neogen, UK) was used for *Lactobacillus-Leuconostoc-Pediococcus* (*LLP*) counts, incubated at 30±1 °C for 72 hours (ISO 15214, 1998). M17 Agar (Condalab 1318.00, Madrid, Spain) was used to count *lactic streptococci* (*Lactococcus-Streptococcus-Enterococcus*), incubated at 30±1 °C for 48-72 hours (Halkman, 2005). Tributyrin Agar (TBA) (Liofilchem 610215, Abruzzi, Italy) and Calcium Caseinate Agar (Condalab 1069.00, Madrid, Spain) were used to enumerate lipolytic and proteolytic microorganisms, respectively, both incubated at 30±1 °C for 48 hours (Halkman, 2005). Violet Red Bile Agar (VRB) (NCM0025A, Neogen, UK) was used for coliform counts, and Violet Red Bile Glucose Agar (VRBG) (NCM0041A, Neogen, UK) for *Enterobacteriaceae* counts, with incubation at 37±1 °C for 24 hours (ISO 4832, 2006; ISO 21528-2:2017). After incubation, red colonies were counted and subjected to biochemical verification. Five colonies per plate were randomly selected and tested using an oxidase test (Liofilchem 88029N, Abruzzi, Italy) (ISO 21528-2:2017). For *Escherichia coli (E. coli*) enumeration, Tryptone Bile X (TBX) Glucuronide Agar (NCM1001A, Neogen, UK) was used, with incubation at 30 °C for 4 hours followed by 44 °C for 18 hours (ISO 16649-2, 2001). Dichloran Rose Bengal Chloramphenicol (DRBC) Agar (NCM0082A, Neogen, UK) was used for yeast counts, incubated at 25±1 °C for 5 days (ISO 21527, 2008). For *Staphylococcus-Micrococcus* enumeration, Baird Parker Agar (Biokar 055HA, France) supplemented with egg-yolk tellurite (Himedia FD046, India) was used, with incubation at 36±1 °C for 48 hours (ISO 6888-1, 2021).

Chemical analysis

The apparent pH was measured using a pH meter (HI 11310, Hanna Instruments, USA). Total titratable acidity (percentage of lactic acid) was determined by the alkaline titration method (Association of Official Analytical Chemists - AOAC, 2023). Salt content was analyzed using the Mohr method ISO 5943:2006 (IDF 88:2006).

Statistical analysis

Pepsin was separately extracted from the proventriculi of geese reared under intensive and extensive conditions. Cheese samples were produced using pepsin alone or in combination with chymosin at various ratios, and their effects over different storage periods were evaluated. For statistical analysis, three factors were considered: rearing method (intensive vs. extensive), enzyme group (100% chymosin; 50% goose pepsin + 50% chymosin; 25% goose pepsin + 75% chymosin; 75% goose pepsin + 25% chymosin; 100% goose pepsin), and storage day (0, 7, 15, 30, 60, 90, and 120). The effects and interaction effects of these variables on cheese quality parameters were evaluated using multifactorial analysis of variance (ANOVA) based on the General Linear Model (GLM). Although storage time is inherently a quantitative variable, it was treated as a fixed categorical factor in the GLM, as measurements were obtained at predefined discrete storage intervals and comparisons among specific storage days were of primary interest. Statistical analyses were performed using SPSS version 22 (IBM SPSS, IBM Corporation, USA). Results were expressed as mean ± standard deviation, with significance set at P≤0.05 (Collins, Dziak, & Li, 2009).

Mathematical model of the study (Equation 2):

$$Y_{ijkl} = \mu + R_i + G_j + D_k + \left(G_j \times D_k\right) + \\ + \left(R_i \times D_k\right) + \left(R_i \times G_j\right) + \left(R_i \times G_j \times D_k\right) + e_{ijkl} \quad (2)$$

μ=Overall mean
R_i=Effect of rearing method
G_j=Effect of enzyme group
D_k= Effect of storage day
e_{ijkl}= Random error term
$G_j \times D_k$= Interaction between enzyme group and storage day
$R_i \times D_k$= Interaction between rearing method and storage day

$R_i \times G_j$ = Interaction between rearing method and enzyme group
$R_i \times G_j \times D_k$ = Interaction between rearing method, enzyme group, and storage day

Results and Discussion

Microbiological analysis results for white brined cheese

The microbiological analysis results for white brined cheese samples are presented in Table 1. The differences in *LLP* and *E. coli* counts with respect to rearing method (intensive vs. extensive) were not statistically significant (P>0.05), whereas all other microbiological parameters showed significant differences (P<0.05). *E. coli* was observed at <1 log CFU/g in all cheese samples. For all examined bacteria and yeasts/molds, differences between storage days (within-group) and between enzyme groups were statistically significant, except for *E. coli* (P<0.001). As the proportion of goose pepsin increased, the counts of all examined bacteria and yeasts/molds declined. With increasing storage time, the counts of TAMB, *LLP*, *Lactic Streptococcus*, coliforms, *Enterobacteriaceae*, yeasts-molds, and *Staphylococcus-Micrococcus* decreased, whereas psychrotrophic, lipolytic, and proteolytic counts increased. Regarding interaction effects: the enzyme group × storage day interaction was significant for *LLP* (P<0.05), the rearing method × enzyme group interaction was significant for proteolytic bacteria (P<0.05), both the rearing method × storage day interaction (P < 0.01) and the enzyme group × storage day interaction (P<0.001) were significant for coliforms, and only the enzyme group × storage day interaction was significant for *Enterobacteriaceae* and *Staphylococcus-Micrococcus* (P<0.001).

Figures 2, 3, 4, 5 and 6 illustrate the regression relationships between storage time and selected cheese quality parameters, demonstrating the temporal evolution of these traits during ripening and storage.

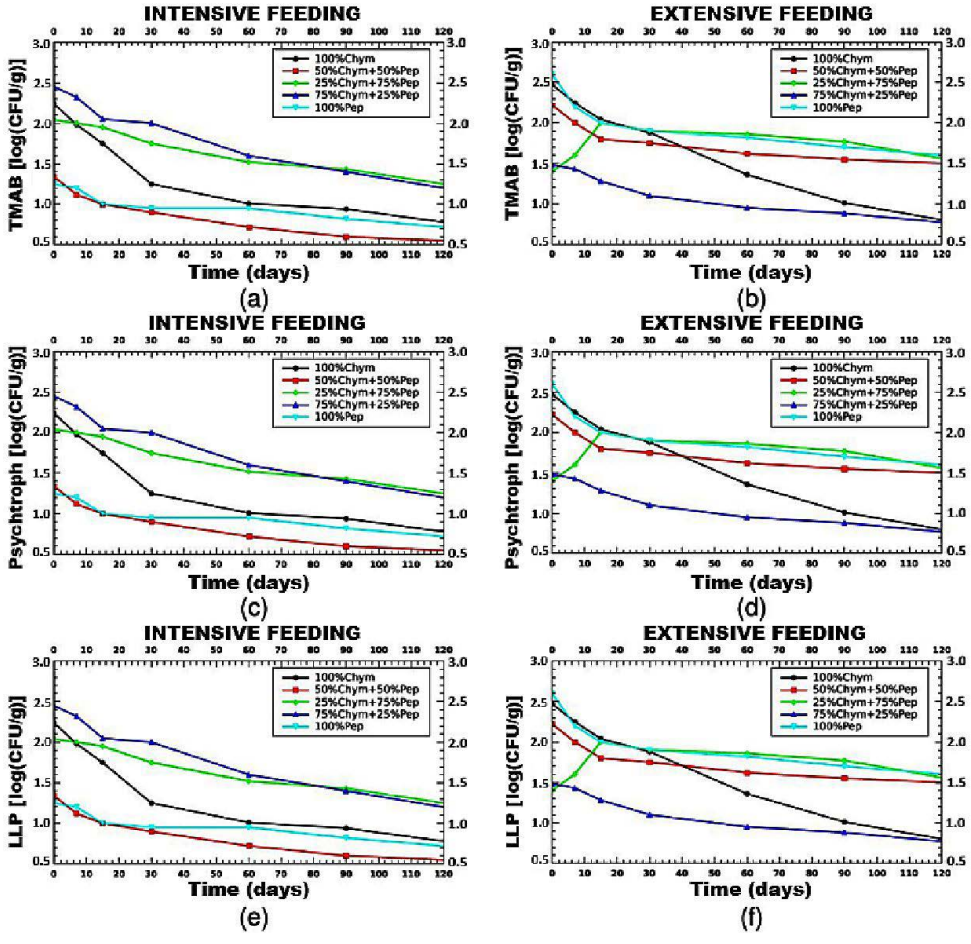

Figure 2: Changes in total aerobic mesophilic bacteria (TAMB) (a, b), psychrotrophic bacteria (c, d), and lactic acid bacteria (*LLP*) (e, f) counts during ripening of white brined cheese produced using different proportions of calf chymosin and goose pepsin under intensive and extensive feeding conditions.

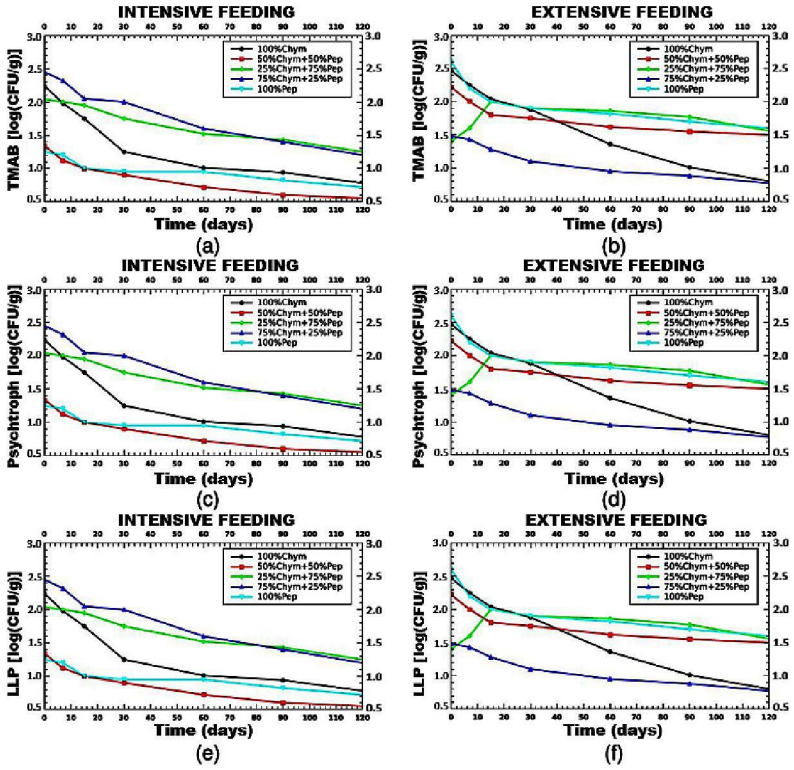

Figure 3: Evolution of *Lactic streptococcus* (*LS*) (g, h), lipolytic (i, j) and proteolytic (k, l) microorganisms in white brined cheese manufactured with different chymosin-goose pepsin combinations under intensive and extensive feeding systems.

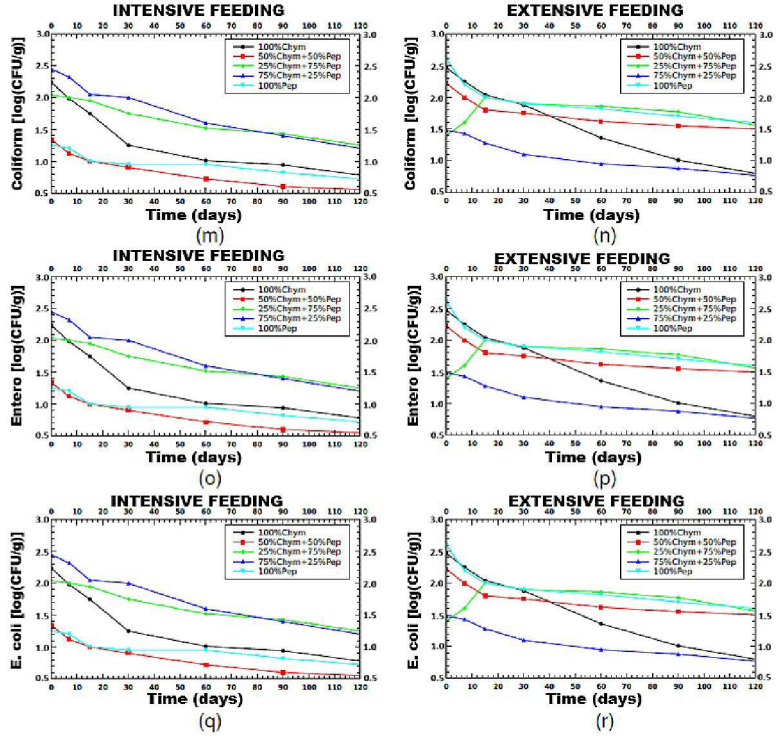

Figure 4: Variation in coliforms (m, n), *Enterobacteriaceae* (o, p), and *Escherichia coli* (q, r) counts during storage of white brined cheese produced with different enzyme ratios under intensive and extensive feeding conditions.

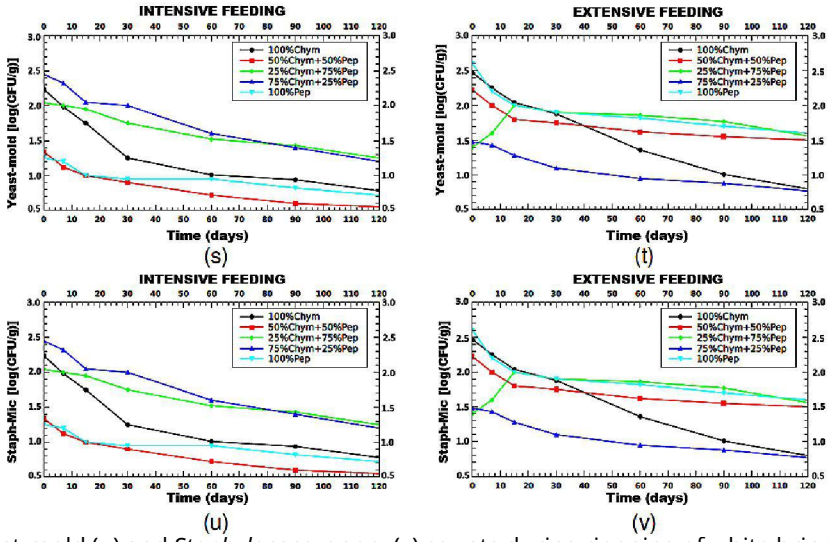

Figure 5: Changes in yeast-mold (u) and *Staphylococcus* spp. (v) counts during ripening of white brined cheese manufactured using calf chymosin and goose pepsin mixtures under different feeding systems.

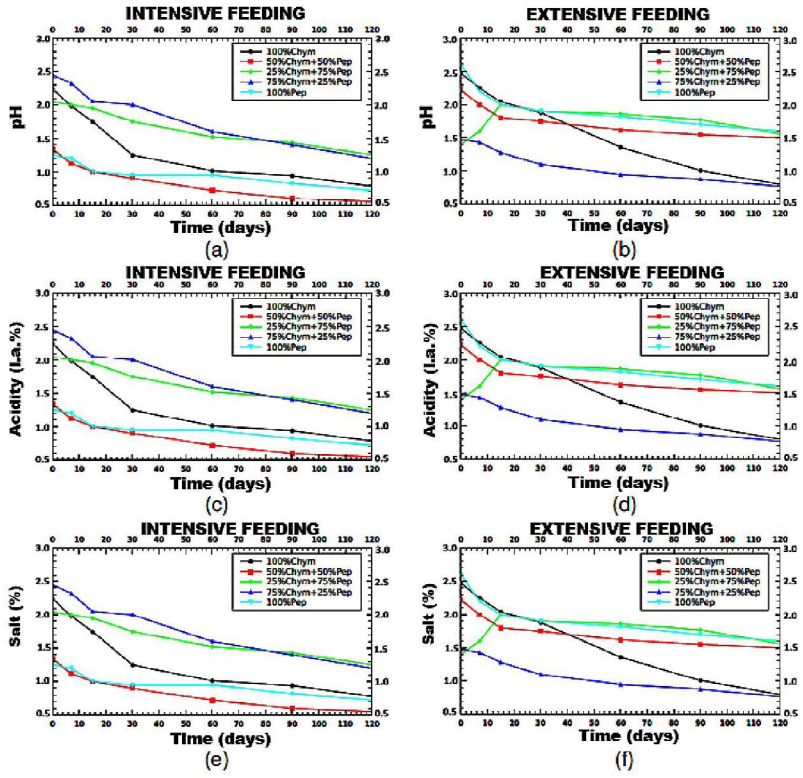

Figure 6: Changes in pH (a, b), acidity (%l.a.) (c, d), and salt (e, f) counts during ripening of white brined cheese produced using different proportions of calf chymosin and goose pepsin under intensive and extensive feeding conditions.

TAMB counts were 8.517 log CFU/g in the intensive group and 8.428 log CFU/g in the extensive group. The rearing method significantly affected TAMB counts ($P<0.01$). Among the enzyme groups, the lowest TAMB count was observed in the 75% pepsin + 25% chymosin group (8.399 log CFU/g), and the highest in the 100% chymosin group (8.678 log CFU/g). Significant differences were observed between groups ($P<0.001$). In terms of storage days, the lowest TAMB count was observed on day 120 (7.570 log CFU/g) and the highest on day 0 (9.621 log CFU/g). TAMB counts decreased progressively over time, and

this difference was statistically significant (P<0.001) (Figure 2, Table 2). Similarly, Ertürkmen, Akbal, & Arısoy (2022) reported TAMB counts ranging from 7.01 to 9.70 log CFU/g on day 0 and from 7.54 to 8.24 log CFU/g on day 90 in white cheeses produced using plant, animal, and microbe-derived coagulants. In another study on the effects of different coagulants in ultrafiltered white cheese, Öner and Arısoy (2019) found TAMB counts ranging from 4.20 to 7.64 log CFU/g on day 1 and from 3.22 to 5.78 log

Table 2: Microbiological analyses of pickled white cheeses.

Raising	TAMB	Psychotroph	LLP	LS	Lipolytic	Proteolytic	Coliform	Entero.	E.coli	Yeast-Mold	*Staph-Mic.*
Intensive	8.517^{a}	4.020^{a}	7.344	8.108^{a}	4.128^{a}	2.722^{a}	1.197^{a}	1.413^{a}	< 1.00	2.251^{a}	1.470^{a}
Extensive	8.428^{b}	3.909^{b}	7.291	7.986^{b}	3.950^{b}	2.566^{b}	1.126^{b}	1.329^{b}	< 1.00	2.159^{b}	1.436^{b}
SEM	0.019	0.026	0.029	0.027	0.037	0.016	0.015	0.026	0.00	0.031	0.012
Groups											
100% Chymosin	8.678^{a}	4.345^{a}	7.648^{a}	8.424^{a}	4.800^{a}	3.052^{a}	1.340^{a}	1.645^{a}	< 1.00	2.505^{a}	1.657^{a}
50% Goose pepsin + 50% Chymosin	8.583^{a}	4.183^{b}	7.457^{b}	8.216^{b}	4.310^{b}	2.761^{b}	1.205^{b}	1.474^{b}	< 1.00	2.334^{ab}	1.495^{b}
25% Goose pepsin + 75% Chymosin	8.456^{b}	4.021^{c}	7.303^{bc}	8.047^{c}	3.991^{c}	2.626^{c}	1.115^{bc}	1.387^{bc}	< 1.00	2.194^{bc}	1.423^{bc}
75% Goose pepsin + 25% Chymosin	8.399^{b}	3.828^{d}	7.203^{c}	7.885^{c}	3.762^{c}	2.456^{d}	1.092^{c}	1.269^{c}	< 1.00	2.058^{cd}	1.371^{cd}
100% Goose pepsin	8.426^{c}	3.445^{e}	6.976^{d}	7.665^{d}	3.333^{d}	2.324^{e}	1.056^{c}	1.080^{d}	< 1.00	1.935^{d}	1.318^{d}
SEM	0.031	0.041	0.046	0.043	0.059	0.025	0.024	0.041	0.00	0.049	0.019
Days											
0	9.621^{a}	2.525^{g}	9.242^{a}	9.394^{a}	2.786^{f}	1.464^{g}	1.770^{a}	2.565^{a}	< 1.00	4.067^{a}	2.396^{a}
7	8.908^{b}	3.214^{f}	8.440^{b}	8.788^{b}	3.237^{e}	2.075^{f}	1.328^{b}	1.777^{b}	< 1.00	3.316^{b}	1.994^{b}
15	8.672^{c}	3.586^{e}	8.122^{c}	8.588^{b}	3.530^{e}	2.421^{e}	1.060^{c}	1.235^{c}	< 1.00	2.642^{c}	1.587^{c}
30	8.489^{d}	3.938^{d}	7.533^{d}	8.062^{c}	3.925^{d}	2.691^{d}	1.001^{c}	1.050^{cd}	< 1.00	1.974^{d}	1.200^{d}
60	8.234^{e}	4.349^{c}	6.589^{e}	7.603^{d}	4.482^{c}	2.986^{c}	< 1.00^{c}	< 1.00^{d}	< 1.00	1.397^{e}	1.012^{e}
90	7.813^{f}	4.867^{b}	5.860^{f}	7.142^{e}	4.979^{b}	3.273^{b}	< 1.00^{c}	< 1.00^{d}	< 1.00	1.049^{f}	< 1.00^{e}
120	7.570^{g}	5.271^{a}	5.438^{g}	6.754^{f}	5.335^{a}	3.598^{a}	< 1.00^{c}	< 1.00^{d}	< 1.00	< 1.00^{f}	< 1.00^{e}
SEM	0.036	0.048	0.055	0.051	0.070	0.029	0.028	0.049	0.00	0.058	0.023
P values											
Raising	<0.01	<0.01	>0.05	<0.01	<0.01	<0.001	<0.01	<0.05	>0.05	<0.05	<0.05
Groups	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	>0.05	<0.001	<0.001
Day	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	>0.05	<0.001	<0.001
Raising x Groups	>0.05	>0.05	>0.05	>0.05	>0.05	<0.05	>0.05	>0.05	>0.05	>0.05	>0.05
Raising x Day	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	<0.01	>0.05	>0.05	>0.05	>0.05
Groups x Day	>0.05	>0.05	<0.05	>0.05	>0.05	>0.05	<0.001	<0.001	>0.05	>0.05	<0.001
Raising x Group x Day	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05
Effect size											
Raising	0.070	0.062	0.012	0.068	0.075	0.267	0.074	0.035	N/A	0.031	0.028
Groups	0.460	0.675	0.462	0.573	0.717	0.790	0.396	0.433	N/A	0.378	0.578
Day	0.940	0.943	0.966	0.937	0.885	0.964	0.825	0.866	N/A	0.948	0.964
Raising x Groups	0.807	0.004	0.020	0.003	0.023	0.077	0.036	0.011	N/A	0.005	0.006
Raising x Day	0.784	0.009	0.015	0.016	0.005	0.025	0.142	0.047	N/A	0.020	0.028
Groups x Day	0.157	0.125	0.216	0.204	0.170	0.191	0.531	0.533	N/A	0.212	0.479
Raising x Group x Day	0.054	0.035	0.067	0.049	0.045	0.122	0.072	0.042	N/A	0.018	0.024

^{a,b,c,d,e,f}: The differences within each factor in the same column are expressed with different letters; SEM: Standard error of the mean; N/A: Not applicable; TAMB: Total aerobic mesophilic bacteria; LLP: *Lactobacillus-Leuconostoc-Pediococcus*; LS: *Lactic streptococcus*; Entero: *Enterobacteriaceae*; Staph.-Mic.: *Staphylococcus-Micrococcus*.

CFU/g on day 120 at 4 °C, and from 4.25 to 7.68 log CFU/g on day 1 and from 3.53 to 5.95 log CFU/g on day 120 at 8 °C. In the present study using goose pepsin, the observed decline in TAMB during storage is consistent with these previous findings (Öner & Arısoy, 2019; Ertürkmen, Akbal, & Arısoy, 2022). This reduction was likely due to increased acidity and salt concentration in the cheese matrix (Kara & Akkaya, 2013). High mesophilic counts indicate conditions favorable to the growth of human and animal pathogens, raising concerns about hygiene and safety (Öner & Arısoy, 2019). Differences in TAMB levels among studies may be attributed to variations in the microbial load of raw milk, pasteurization procedures, post-pasteurization contamination, and the thermotolerance of indigenous microbial strains (Ertürkmen, Akbal, & Arısoy, 2022). Furthermore, increasing acidity and salt concentration in cheese blocks may also suppress TAMB (Yılmaztekin, Özer, & Atasoy, 2004).

Psychrotrophic bacteria, which produce lipases and proteases that degrade milk components, can survive pasteurization and, when present in high numbers, cause bitterness, rancidity, texture defects, and discoloration (Çakmakçı, Bakırcı, & Akyüz, 1996). However, these effects are generally not observed unless counts exceed 10^6 CFU/g (Öner & Arısoy, 2019). In the current study, the counts of psychrotrophic bacteria increased in all groups during storage. Psychrotrophic bacterial counts were 4.020 log CFU/g in the intensive group and 3.909 log CFU/g in the extensive group. The rearing method had a significant effect on these counts ($P<0.01$). Among the enzyme groups, the lowest psychrotrophic bacterial count was observed in the 100% pepsin group (3.445 $\log_{10}$CFU/g), and the highest in the 100% chymosin group (4.345 log CFU/g) ($P<0.001$). The lowest count was on day 0 (2.525 log CFU/g) and the highest on day 120 (5.271 log CFU/g), with counts increasing progressively over time ($P<0.001$) (Figure 2, Table 2). In this study, the increase in psychrotrophic bacteria during storage is primarily attributed to the low maturation temperature (4±1 °C) and long storage duration, rather than enzyme treatments. Since increased acidity and salt concentration, and a decrease in pH, reduced the number of mesophilic and starter culture-associated microorganisms, psychrotrophic bacteria well adapted to cold storage conditions gained a competitive advantage and progressively increased throughout maturation. Öner & Arısoy (2019) reported counts ranging from <1.0 to 7.75 log CFU/g on day 1 and from <1.0 to 5.78 log CFU/g on day 120 at 4 °C, and from <1.0 to 7.75 log CFU/g on day 1 and from <1.0 to 6.01 log CFU/g on day 120 at 8 °C.

Lactic acid bacteria, which influence the characteristic flavor, aroma, and shelf life of foods, play a central role in cheese ripening. They also contribute to biological preservation by secreting antimicrobial substances such as acetic acid, bacteriocins, and lactic acid (Demir, İlhak, & Öksüztepe, 2023). The rearing method did not significantly affect *LLP* counts ($P>0.05$). In the present study, *Lactobacillus-Leuconostoc-Pediococcus* (*LLP*) counts declined continuously during storage in all experimental cheese samples ($P<0.001$). *LLP* counts were 7.344 log CFU/g in the intensive group and 7.291 log CFU/g in the extensive group. Among enzyme groups, the lowest *LLP* count was observed in the 100% pepsin group (6.976 log CFU/g), and the highest in the 100% chymosin group (7.648 log CFU/g) ($P<0.001$). The lowest count was observed on day 120 (5.438 log CFU/g), and the highest on day 0 (9.242 log CFU/g) (Figure 2, Table 2). Ertürkmen, Akbal and Arısoy (2022) reported *LLP* counts of 7.60-8.28 log CFU/g on day 0 and 6.25-7.61 log CFU/g on day 90. The decline in *LLP* toward the end of storage observed in this study aligns with the findings of Ertürkmen, Akbal and Arısoy (2022). Karaca and Güven (2004), in their study on the effects of microbiologically derived proteolytic and lipolytic enzymes on white cheese characteristics and ripening rates, reported *LLP* counts between 7.30 and 8.15 log CFU/g (1.4×10^6 - 2×10^7 CFU/g). For probiotic microorganisms, salt concentration and dissolved oxygen in cheese are considered the most critical limiting factors for growth (Fox et al., 2017; Yılmaztekin, Özer, & Atasoy, 2004).

Lactic Streptococcus, used as starter cultures in the dairy industry, impart flavor and aroma to the product and play an important role in cheese ripening (Ateş & Patır, 2000). *Lactic Streptococcus* counts were 8.108 log CFU/g in the intensive group and 7.986 log CFU/g in the extensive group. Counts decreased significantly in all groups over storage ($P<0.001$). The rearing method had a significant effect on these counts ($P<0.01$). Overall differences were significant, although counts on days 7 and 15 were similar. All enzyme groups also differed significantly in terms of bacterial counts. Among the enzyme groups, the lowest count was observed in the 100% pepsin group (7.665 log CFU/g) and the highest in the 100% chymosin group (8.424 log CFU/g) ($P<0.001$). The lowest count was on day 120 (6.754 log CFU/g), and the highest on day 0 (9.394 log CFU/g) (Figure 3, Table 2). Ertürkmen, Akbal and Arısoy (2022) reported *Lactic Streptococcus* counts of 8.30-9.41 log CFU/g on day 0 and 7.52-8.66 log CFU/g on day 90. The decline in *Lactic Streptococcus* counts during storage is attributed to factors such as decreasing pH, increased salt concentration, and the depletion of fermentable carbohydrates (Chaves & Gigante, 2016).

Lipolytic microorganisms degrade milk fat in dairy products, resulting in undesirable flavors, aromas, tastes, and odors. These bacteria are less prevalent in products made from pasteurized milk with added starter cultures. The extent of lipolysis is also influenced by the lipase concentration present in the rennet (Demir, İlhak, & Öksüztepe, 2023). Lipolytic microorganism counts were 4.128 log CFU/g in the intensive group and 3.950 log CFU/g in the extensive group. The rearing method significantly affected these counts ($P<0.01$). Lipolytic counts also differed significantly among the enzyme groups. The lowest count was in the 100% pepsin group (3.333 log CFU/g), and the highest in the 100% chymosin group (4.800 log CFU/g) ($P<0.001$). Counts increased

continuously during storage, from 2.786 log CFU/g on day 0 to 5.335 log CFU/g on day 120 (P<0.001) (Figure 3, Table 2).

Proteolytic microorganisms hydrolyze proteins, negatively affecting taste and aroma. They tend to proliferate during storage, compromising the structural integrity of the product and reducing shelf life. In the present study, proteolytic counts increased continuously in all cheese groups throughout the storage period, with significant differences among enzyme treatments. Proteolytic activity in cheese may originate from coagulant enzymes, milk-derived enzymes, starter cultures, and non-starter bacteria (Demir, İlhak, & Öksüztepe, 2023). Proteolytic microorganism counts were 2.722 log CFU/g in the intensive group and 2.566 log CFU/g in the extensive group. The rearing method had a significant effect (P<0.001). The lowest count was in the 100% pepsin group (2.324 log CFU/g), and the highest in the 100% chymosin group (3.052 log CFU/g) (P<0.001). Counts increased from 1.464 log CFU/g on day 0 to 3.598 log CFU/g on day 120 (P<0.001) (Figure 3, Table 2).

While declining pH and increasing salt concentration typically create unfavorable conditions for most microorganisms, certain species such as lipolytic and proteolytic bacteria are more resistant to these stressors and may continue to proliferate (Al-Nabulsi et al., 2020).

Coliform bacteria are recognized as hygiene indicators. Their presence in food products signals insufficient sanitation, inadequate thermal processing, or post-processing contamination (Demir, İlhak, & Öksüztepe, 2023). Coliform counts were 1.197 log CFU/g in the intensive group and 1.126 log CFU/g in the extensive group. The rearing method significantly affected coliform counts (P<0.01). In the present study, significant differences in coliform counts were observed on days 0, 7, and 15, while counts were similar on subsequent days. The lowest count was in the 100% pepsin group (1.056 log CFU/g), and the highest in the 100% chymosin group (1.340 log CFU/g) (P<0.001). Counts declined steadily, from 1.770 log CFU/g on day 0 to 1.001 log CFU/g on day 30. No coliform growth (< 1.0 log CFU/g) was detected from day 60 onward (P<0.001) (Figure 4, Table 2). Ertürkmen, Akbal and Arısoy (2022) reported coliform counts of 3.76-6.73 log CFU/g on day 0 and <1.0-6.83 log CFU/g on day 90. The decrease in coliform counts observed toward the end of the storage period in this study is consistent with their findings. It is believed that lactic acid bacteria and molds exert an inhibitory effect on coliform development during ripening (Ertürkmen, Akbal, & Arısoy, 2022).

Enterobacteriaceae counts decreased significantly across all groups during storage (P<0.001). Counts were 1.413 log CFU/g in the intensive group and 1.329 log CFU/g in the extensive group. The rearing method had a significant effect (P<0.05). The lowest count was in the 100% pepsin group (1.080 log CFU/g), and the highest in the 100% chymosin group (1.645 log CFU/g). Significant differences were observed between groups (P<0.001). The lowest count was on day 30 (1.050 log CFU/g), and the highest on day 0 (2.565 log CFU/g) (Figure 4, Table 2).

No *E. coli* growth was detected in any cheese sample from day 0 onward; *E. coli* remained below detectable levels (<1.0 log CFU/g) (P>0.05) (Figure 4, Table 2).

Yeasts and molds can grow over a broad pH range (2-9), at suitable storage temperatures (10-35°C), and at water activity levels ≥0.85. Their presence indicates inadequate hygiene during production or storage. With both lipolytic and proteolytic activity, yeasts and molds can cause gas formation, unwanted pores, and off-flavors and odors in foods (Frazier & Westhoff, 1978; Gönül & Bostan, 2018). In the present study, yeast-mold counts declined continuously from the beginning of storage across all enzyme groups. Yeast-mold counts were 2.251 $\log_{10}$CFU/g in the intensive group and 2.159 log CFU/g in the extensive group (P<0.05). The lowest count was in the 100% pepsin group (1.935 log CFU/g), and the highest in the 100% chymosin group (2.505 log CFU/g) (P<0.001). Counts decreased from 4.067 log CFU/g on day 0 to 1.049 log CFU/g on day 90, with no detectable growth from day 120 onward (<1.0 log CFU/g). The decrease in yeast-mold counts was statistically significant (P<0.001) (Figure 5, Table 2). Ertürkmen, Akbal and Arısoy (2022) reported yeast-mold counts of 2.69-6.13 log CFU/g on day 0 and 5.65-6.47 log CFU/g on day 90. Öner and Arısoy (2019) reported yeast-mold counts of <1.0-7.35 log CFU/g on day 1 and <1.0-5.74 log CFU/g on day 120 at 4°C, and <1.0-7.37 log CFU/g on day 1 and <1.0-5.53 log CFU/g on day 120 at 8°C.

Staphylococcus-Micrococcus counts were 1.470 log CFU/g in the intensive group and 1.436 log CFU/g in the extensive group (P<0.05). The lowest count was observed in the 100% pepsin group (1.318 log CFU/g), and the highest in the 100% chymosin group (1.657 log CFU/g) (P<0.001). Counts decreased from 2.396 log CFU/g on day 0 to 1.012 log CFU/g on day 60, with no detectable growth from day 90 onward (<1.0 log CFU/g). The decrease was statistically significant (P<0.001) (Figure 5, Table 2).The low pH, high salt concentration, and activity of lactic acid bacteria and the associated acidity are likely responsible for inhibiting these bacteria (Al-Nabulsi et al., 2020).

Factor interactions were not statistically significant for TAMB, psychrotrophic bacteria, *Lactic Streptococcus*, lipolytic microorganisms, *E. coli*, and yeast-mold counts (P>0.05) (Table 1).

The observed microbial changes are primarily driven by low storage temperature, while the differences among treatments reflect the influence of enzyme type and proportion. Variations in microbiological findings across studies on white brined cheese may result from the use of raw vs. pasteurized milk; differences in starter culture types and coagulant enzymes; endogenous milk enzymes; variations in production protocols; storage and ripening conditions; initial microbial load; brine salt concentration; added chemical components; moisture content; and cheese pH.

In the present study, microbial growth was lower in the pepsin groups compared to the chymosin groups, suggesting that goose pepsin may exhibit antimicrobial properties. Due to its lower proteolytic activity, goose pepsin causes less protein hydrolysis, resulting in slower acidification and thereby limiting microbial proliferation (Egito et al., 2001; Sousa, Ardö, & McSweeney, 2001).

Differences in the gastric microbial flora and pepsin activity of extensively reared animals may influence the microbiological properties of the resulting pepsin. Additionally, geese raised under extensive conditions consume natural diets with more diverse microbial flora, which may affect the stability of the enzymes obtained from them. The reduced microbial growth observed in pepsins derived from extensively reared geese compared to those from intensively reared birds can be attributed to the influence of rearing conditions on gastric flora and corresponding differences in pepsin activity (Yadav & Jha, 2019; Stocco et al., 2025).

Chemical analysis results for white brined cheese samples

The pH value significantly influences the chemical characteristics of cheese (Oluk, 2023). During storage, free fatty acids and amino acids released through lipolysis and proteolysis cause a decline in pH (Dermiki et al., 2008). In addition, non-starter lactic acid bacteria originating from residual *LAB* surviving heat treatment or introduced via equipment, utensils, or ambient air may contribute to changes in pH (Ertürkmen, Akbal, & Arısoy, 2022). Moynihan et al. (2014) reported that cheeses produced using different coagulant enzymes often exhibit similar pH values. According to the Turkish Standards Institute (TSI 591, 2023), white brined cheeses must have a pH of at least 4.5. The pH values of the experimental cheese samples decreased continuously during storage across all groups. The pH was 6.075 in the intensive group and 6.104 in the extensive group. Among the enzyme groups, the lowest pH was observed in the 100% chymosin group (6.015), and the highest in the 100% goose pepsin group (6.182). The difference between enzyme groups was statistically significant ($P<0.001$). The lowest pH value was observed on day 120 (5.586), and the highest on day 0 (6.481) (Figure 6, Table 3). Differences in pH over the storage period were statistically significant ($P<0.001$), while the rearing method did not significantly affect pH ($P>0.05$). Türkmen and Güler (2022) investigated the effects of chymosin type and brine concentration on the chemical composition, texture, microstructure, and color of white cheese, reporting pH values of 4.92-5.07. Ahmed (2021), in a study on white cheeses made with free (F) and immobilized (Z) calf (1), bovine (2), and chicken (3) pepsins over a 90-day ripening period, reported the following pH ranges: F1: 5.21-6.45, Z1: 5.38-6.55, F2: 5.24-6.36, Z2: 5.29-6.44, F3: 5.32-6.41, and Z3: 5.35-6.53. Öner and Arısoy (2019) reported pH values of 4.42-4.72 on day 1 and 4.21-4.56 on day 120 at 4 °C, and 4.48-4.70 on day 1 and 4.18-4.54 on day 120 at 8 °C. In another study examining camel and calf chymosin blends on proteolysis, residual coagulant activity, microstructure, and sensory properties of white cheese, Gümuş and Hayaloğlu (2019) reported pH values of 5.45-5.60 on day 1 and 5.35-5.45 on day 90. Çepoğlu & Güler-Akın (2013) reported pH values of A: 5.55-5.62, B: 5.59-5.65, and C: 5.61-5.88 after 60 days of ripening with different coagulants. Karaca and Güven (2010) examined the effects of lipolytic and proteolytic enzymes on white cheese and reported pH values of 4.64-4.72 on day 1 and 4.40-4.46 on day 90. Vapur (2021) investigated full-fat white cheeses made with various starter ratios and coagulants, reporting pH values of 4.72-5.10 on day 1 and 4.73-5.00 on day 90. Yetişemeyen et al. (1998) reported pH values of 4.72-5.45 on day 1 and 4.53-5.25 on day 60 for white brined cheese made using animal and microbial enzymes and the ultrafiltration method. The decrease in pH observed in this study toward the end of storage is consistent with findings by Yetişemeyen et al. (1998), Karaca and Güven (2010), Vapur (2021), Gümuş and Hayaloğlu (2019), Öner and Arısoy (2019), and Ahmed (2021). This decline is likely due to the activity of lactic acid bacteria from milk and starter cultures, as well as increased proteolysis (Türkmen & Güler, 2022). Additionally, amino acid synthesis and the formation of free fatty acids from psychrotrophic bacteria-induced proteolysis and lipolysis may also contribute to this decline, aligning with the microbiological findings (Fathollahi et al., 2010).

Brined cheeses rely on the inhibitory effect of lactic acid and the stability of the curd to suppress undesirable microorganisms, making acid development a critical quality criterion (Hayaloğlu et al., 2005). Titratable acidity arises from lactic acid formed via lactose fermentation by LAB, as well as from liberated fatty acids and amino acids. Other microorganisms also contribute to acidity by producing acetic, formic, butyric, and other organic acids. Acidity plays key roles in flavor, aroma, texture development, ripening, and microbial inhibition (Hayaloğlu, Güven, & Fox, 2002; Öner & Sarıdağ, 2019). It has also been suggested that casein and paracasein breakdown increases acidity (Ertürkmen, Akbal, & Arısoy, 2022). Çepoğlu and Güler-Akın (2013) reported that different coagulant enzymes significantly affect titratable acidity. According to Turkish Standards Institute 591 (TSI, 2023), acidity in brined cheese must not exceed 3° SH. Titratable acidity (lactic acid%) increased significantly throughout storage in all groups ($P<0.001$). Acidity was 0.390% in the intensive group and 0.367% in the extensive group ($P<0.001$). The lowest acidity was in the 100% goose pepsin group (0.297%), and the highest in the 100% chymosin group (0.452%) ($P<0.001$). Acidity increased from 0.179% on day 0 to 0.722% on day 120, with significant differences across storage days ($P<0.001$) (Figure 6, Table 3). Ahmed (2021) reported acidity values of F1: 0.18%-1.22%, Z1: 0.16%-1.16%, F2: 0.19%-1.29%, Z2: 0.18%-1.18%, F3: 0.20%-1.18%, and Z3: 0.19%-1.21% over a 90 day ripening period.

Table 3: Some chemical analyses of pickled white cheeses.

Raising	pH	Acidity (percentage of lactic acid)	Salt (%)
Intensive	6.075	0.390[a]	6.094[a]
Extensive	6.104	0.367[b]	5.933[b]
SEM	0.015	0.004	0.015
Groups			
100% Chymosin	6.015[c]	0.452[a]	6.088[a]
50% Goose pepsin + 50% Chymosin	6.046[bc]	0.411[b]	6.014[a]
25% Goose pepsin + 75% Chymosin	6.077[bc]	0.386[c]	6.059[a]
75% Goose pepsin + 25% Chymosin	6.127[ab]	0.349[d]	6.007[a]
100% Goose pepsin	6.182[a]	0.297[e]	5.899[b]
SEM	0.024	0.006	0.024
Days			
0	6.481[a]	0.179[g]	0.788[g]
7	6.315[b]	0.216[f]	6.351[f]
15	6.213[bc]	0.280[e]	6.541[e]
30	6.137[cd]	0.347[d]	6.766[d]
60	6.072[d]	0.409[c]	7.032[c]
90	5.822[e]	0.500[b]	7.231[b]
120	5.586[f]	0.722[a]	7.384[a]
SEM	0.029	0.007	0.029
P values			
Raising	>0.05	<0.001	<0.001
Groups	<0.001	<0.001	<0.001
Day	<0.001	<0.001	<0.001
Raising x Groups	>0.05	<0.001	<0.001
Raising x Day	>0.05	>0.05	<0.01
Groups x Day	>0.05	<0.001	>0.05
Raising x Group x Day	>0.05	>0.05	<0.05
Effect size			
Raising	0.013	0.112	0.283
Groups	0.177	0.726	0.201
Day	0.827	0.966	0.996
Raising x Groups	0.003	0.143	0.220
Raising x Day	0.010	0.052	0.155
Groups x Day	0.039	0.450	0.213
Raising x Group x Day	0.019	0.141	0.241

SEM: Standard error of the mean.

Öner and Arısoy (2019) reported acidity values of 76.70-99.4 SH (0.69%-0.89%) on day 1 and 82.50-124.25 SH (0.74%-1.12%) on day 120 at 4 °C, and 76.00-110.5 SH (0.68%-0.99%) on day 1 and 84.4-130.1 SH (0.76%-1.17%) on day 120 at 8 °C. Çepoğlu and Güler-Akın (2013) reported acidity values of A: 0.565%-0.620%, B: 0.515%-0.540%, and C: 0.525%-0.560%. Karaca and Güven (2010) reported acidity values of 2.02%-2.06% on day 1 and 2.41%-2.52% on day 90. Vapur (2021) reported values

of 0.52%-0.85% (57.73-94.98 SH) on day 1 and 0.67%-0.82% (74.85-90.63 SH) on day 90. Yetişemeyen et al. (1998) recorded 0.29%-0.60% (32.49-66.79 SH) on day 1 and 0.34%-0.64% (37.66-71.29 SH) on day 60 of storage. The increase in acidity toward the end of the storage period in the present study aligns with the findings of Yetişemeyen et al. (1998), Karaca and Güven (2010), Öner and Arısoy (2019), and Ahmed (2021). Rising acidity levels, alongside declining pH, are consistent markers of cheese maturation. These changes are largely attributed to degradation products formed during proteolysis and lipolysis, as well as changes in dry matter content (Yılmaztekin et al., 2004). Additionally, acidity variation may result from microbial activity, brine temperature, and the extent of whey expulsion.

As salt content in cheese samples increases, a concomitant rise in dry matter occurs due to reverse salt and water diffusion, where water migrates out of the cheese matrix (Soltani, et al., 2022). Dry matter variation may be influenced by the characteristics of the raw milk, cheesemaking techniques, cheese variety, and degree of maturation. Higher fat content within the cheese matrix also binds more water, thereby increasing dry matter levels (Tarakçı & Akyüz, 2009). Salt, as an inorganic component of the dry matter, becomes more concentrated as cheeses lose moisture during ripening. It not only prevents the development of off-flavors but also enhances water-soluble nitrogen content. Additionally, salt plays a critical role in reducing water activity and regulating key biochemical processes such as paracasein hydrolysis, glycolysis, lipolysis, and proteolysis, as well as influencing enzymatic activity, microbial growth, composition, flavor, aroma, rheological properties, and texture (Guinee, 2004; Türkmen & Güler, 2022). According to the Turkish Food Codex (2015), salt content in the dry matter of brined cheeses must not exceed 7.5%, while the Turkish Standards Institute (TSI, 2023) sets the upper limit at 10%. Salt content in dry matter (%) increased continuously across during storage in all groups. It was 6.094% in the intensive group and 5.933% in the extensive group. The lowest salt content was in the 100% goose pepsin group (5.899%), and the highest in the 100% chymosin group (6.088%). Salt content rose from 0.788% on day 0 to 7.384% on day 120. All factors exhibited statistically significant differences ($P<0.001$) (Figure 6, Table 3). Öner and Arısoy (2019) reported salt contents of 3.48%-5.63% on day 1 and 4.18%-6.20% on day 120 at 4°C, and 4.11%-5.96% on day 1 and 3.70%-6.69% on day 120 at 8°C. Gümuş and Hayaloğlu (2019) found salt contents of 4.09%-5.79% on day 1 and 5.00%-6.23% on day 90. Çepoğlu and Güler-Akın (2013) recorded salt contents of A: 3.13%-4.14%, B: 3.46%-4.26%, and C: 3.33%-4.32%. Karaca and Güven (2010) reported 4.10%-5.78% on day 1 and 3.56%-4.01% on day 90. Vapur (2021) noted a sharp increase, from 3.69%-5.21% on day 1 to 9.31%-13.34% on day 90. Karaca and Güven (2004) found salt levels of 7.58%-9.05% on day 1 and 13.24%-14.37% on day 90. Yetişmeyen et al. (1998) reported 9.90%-16.84% on day 1 and 14.67%-18.14% on day 60. In the present study, the increase in salt content (% in dry matter) toward the end of the storage period aligns with the findings of Yetişmeyen et al. (1998), Karaca and Güven (2004), Vapur (2021), Gümüş and Hayaloğlu (2019), and Öner and Arısoy (2019). Salt uptake by cheese is influenced by brine concentration, temperature, and immersion time (Al-Nabulsi et al., 2020). During cheese production, osmotic pressure differences drive Na^+ and Cl^- ions from the brine into the cheese matrix, progressing from the surface to the center over time, thereby increasing both salt and dry matter content (Fox et al., 2017; Simal et al., 2001).

Factor interactions were not statistically significant for pH ($P>0.05$). In contrast, the rearing method × enzyme group and enzyme group × storage day interactions were significant for salt content, and all factor interactions were significant for titratable acidity ($P<0.05$) (Table 3).

Compositional differences in cheese may also be attributed to factors such as lactation stage, genetics, moisture content, feeding regimen, and geographic location (Al Haj & Al Kanhal, 2010).

The storage-dependent decrease in pH, coupled with increases in acidity, particularly lactic acid and salt concentration, inhibits the growth of bacteria, yeasts, and molds, ensures microbial stability, and serves as an indicator of cheese ripening (McSweeney, 2004).

The lower acidity observed with increasing proportions of goose pepsin in the coagulant mixture may be due to goose pepsin's comparatively lower proteolytic activity. Conversely, the higher salt content recorded in the 100% chymosin groups may be attributed to chymosin's stronger proteolytic effect, which promotes greater whey expulsion and consequently accelerates salt ingress into the cheese matrix (Hayaloglu et al., 2004).

Conclusions

This study demonstrated that pepsins extracted from geese reared under different systems, intensive versus extensive, differ in their microbiological and chemical characteristics. The findings suggest that pasture-based feeding enhances the development of gut microbiota in geese, thereby influencing the enzymatic properties of extracted pepsin. The observed microbiological differences between chymosin and goose pepsin further indicate that goose pepsin may exhibit intrinsic antimicrobial properties or different enzymatic activity levels compared to traditional chymosin.

Author Contribution

Conceptual idea: Beyazgül, P.; Methodology design: Beyazgül, P.; Data collection: Beyazgül, P., Tonbak, F.; Özmen, G. K.; Baykalir, Y.; Şimşek, U. G.; Arslan, A.; Deniz, A. R.; Yilmaz, O.; Data analysis and interpretation: Beyazgül, P.; Baykalir, Y.; Şimşek, U. G.; Writing and editing: Beyazgül, P.; Baykalir, Y.; Şimşek, U. G.

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Data availability statement

Data available upon request to authors.

References

Afsharnezhad, M., Shahangian, S. S., & Sariri, R. (2019). A novel milk-clotting cysteine protease from *Ficus johannis*: purification and characterization. *International Journal of Biological Macromolecules*, *121*:173-182.

Ahmed, M. A. (2021). Quality and properties evaluation of white soft cheese made by free or immobilized coagulant enzymes durıng storage. *The Egyptian Journal of Applied Sciences*, *36*(5):29-44.

Al haj, O. A., & Al Kanhal, H. A. (2010). Compositional technological and nutritional aspects of dromedary camel milk. *International Dairy Journal*, *20*(12):811-821.

Al-Nabulsi, A. A. et al. (2020). Factors affecting the viability of *Staphylococcus aureus* and production of enterotoxin during processing and storage of white-brined cheese. *Journal of Dairy Science*, *103*(8):6869-6881.

Anusha, R., Singh, M. K., & Bindhu O. (2014). Characterization of potential milk coagulants from *Calotropis gigantea* plant parts and their hydrolytic pattern of bovine casein. *European Food Research and Technology*, *238*:997-1006.

Association of Official Analytical Chemists - AOAC. (2023). *Official methods of analysis*. (22th Edition), USA, Washington DC: Association of Official Analytical Chemists.

Ateş, G., & Patır, B. (2000). Starter kültürlü tulum peynirinin olgunlaşması sırasında duyusal, kimyasal ve mikrobiyolojik niteliklerinde meydana gelen değişimler üzerine araştırmalar. *Fırat Üniversitesi Sağlık Bilimleri Dergisi*, *15*(1):45-56. Available in: https://search.trdizin.gov.tr/en/yayin/detay/27214/starter-kulturlu-tulum-peynirinin-olgunlasmasi-sirasinda-duyusal-kimyasal-ve-mikrobiyolojik-niteliklerinde-meydana-gelen-degisimler-uzerine-arastirmalar.

Bennett, R. J., & Johnston, K. A. (2004). General aspects of cheese technology. In P. F. Fox. et al. (Eds.)(3rd Edition*) Cheese: Chemistry, physics and microbiology*. London, UK: Elsevier Academic Press, v.2, (pp.281-304).

Berridge, N. J. (1952). Some observations on the determination of the activity of rennet. *Analyst*, *77*:57-60.

Chaves, K. S., & Gigante, M. L. (2016). Prato cheese as suitable carrier for *Lactobacillus acidophilus* La5 and *Bifidobacterium* Bb12. *International Dairy Journal*, *52*:10-18.

Collins, L. M., Dziak, J. J., & Li, R. (2009). Design of experiments with multiple independent variables: A resource management perspective on complete and reduced factorial designs. *Psychological Methods*, *14*:202-224.

Costabel, L. M. et al. (2015). Influence of chymosin type and curd scalding temperature on proteolysis of hard cooked cheeses. *Journal of Dairy Research*, *82*(3):375-384.

Coşkun, H., & Akgündüz, S. N. (2020). Peynir mayası üretiminde helal ve haram yaklaşımlar. *Journal of Halal and Ethical Research*, *2*(2):34-53.

Çakmakçı, S., Bakırcı, İ., & Akyüz, N. (1996). Characteristics of raw milk to be processed into drinking milk and the tests to be applied. *Drinking Milk (İçme Sütü)*, 49-64.

Çepoğlu, F., & Güler-Akın, M. B. (2013). Effects of coagulating enzyme types (commercial calf rennet, *Aspergillus niger var*. *awamori* as recombinant chymosin and *Rhizomucor miehei* as microbial rennet) on the chemical and sensory characteristics of white pickled cheese. *African Journal of Biotechnology*, *12*(37):5588-5594.

Demir, P., İlhak, O. İ., & Öksüztepe, G. (2023). The impact on shelf life of Tulum cheese of modified atmosphere packaging. *Emirates Journal of Food and Agriculture*, *35*(12):1-12.

Dermiki, M. et al. (2008). Shelf-life extension and quality attributes of the whey cheese "Myzithra Kalathaki" using modified atmosphere packaging. *Journal of Food Science and Technology*, *41*(2):284-294.

Dervişoğlu, M., Aydemir, O., & Yazıcı, F. (2007). Peynir yapımında kullanılan pıhtılaştırıcı enzimler ve kazein fraksiyonları üzerine etkileri. *Food*, *32*(5):241-249.

Dinkci, N. (2020).Traditional cheeses of Marmara region, Türkiye. In C. A. Oluk., & O. B. Karaca. (Eds.) *Current developments in Food in nutrition research Traditional cheeses from selected regions in Asia, Europe, and South America*. Bentham Books, v.1, (pp.142-167).

Egito, A. S. et al. (2001). Milk-clotting properties of enzyme extracts from sunflower and albizia seeds and specific hydrolysis of bovine κ-casein. *International Dairy Journal*, *17*(7):816-825.

El-Beltagy, A. E. et al. (2004). Purification and characterization of an acidic protease from the viscera of bolti fish (*Tilapia nilotica*). *Food Chemistry*, *86*(1):33-39.

Erkaya Kotan, T., & Hayaloglu, A. A. (2024). Volatilome profile (HS-SPME/GC-MS) and proteolysis in beyaz peynir (white-brined cheese) made using different probiotic adjunct cultures and ripening under brine or vacuum package systems, and chemometric analysis. *International Dairy Journal*, *152*:105894.

Ertürkmen, P., Akbal, S., & Arısoy, Z. (2022). Bitkisel, hayvansal ve mikrobiyal enzim kullanılarak elde edilen beyaz peynirlerin fizikokimyasal ve mikrobiyal içeriklerinin belirlenmesi. *Turkish Journal of Agriculture-Food Science and Technology*, *10*(11):2252-2262.

Fathollahi, I. et al. (2010). Influence of proteolysis and soluble calcium levels on textural changes in the interior and exterior of Iranian UF white cheese during ripening. *World Academy of Science, Engineering and Technology*, *4*:705-710.

Fox, P. F. et al. (2004). *Cheese: Chemistry, physics and microbiology general aspects.* (3th ed), v.1, 47p .

Fox, P. F. et al. (2017). *Fundamentals of Cheese Science*. Springer, Boston, MA.

Frazier, W. C., & Westhoff, D. C. (1978). Foods in relation to disease (5th ed). In W. C. Frazier., & D. C. Westhoff. *Food Microbiology.* New York: McGrow-Hill Book Company, (pp.399-439).

Gönül, Ö., & Bostan, K. (2018). Kremaların mikrobiyolojik kalitesi üzerine nisin ve lisozimin etkisi). *Aydın Gastronomy*, *2*(1):1-13.

Guinee, T. P. (2004). Salting and the role of salt in cheese. *International Journal of Dairy Technology*, *57*(2-3):99e109.

Gümus, P., & Hayaloglu, A. A. (2019). Effects of blends of camel and calf chymosin on proteolysis, residual coagulant activity, microstructure, and sensory characteristics of beyaz peynir. *Journal of Dairy Science*, *102*(7):5945-5956.

Halkman, A. K. (2005). *Gıda mikrobiyolojisi uygulamaları*. Başak Matbaacılık, Ankara, Türkiye.

Hayaloglu, A. A. et al. (2004). Proteolysis in Turkish White-brined cheeses made with different coagulants. *International Dairy Journal*, *14*(7):599-610.

Hayaloglu, A. A. et al. (2005). Influence of starters on chemical, biochemical, and sensory changes in Turkish white-brined cheese during ripening. *Journal of Dairy Science*, *88*(10):3460-3474.

Hayaloglu, A. A. Güven, M., & Fox, P. F. (2002). Microbiological, biochemical and technological properties of Turkish White cheese Beyaz Peynir. *International Dairy Journal*, *12*(8):635-648.

Homayouni, A. et al. (2020). Cheese as a potential food carrier to deliver probiotic microorganisms into the human gut: A review. *Current Nutrition & Food Science*, *16*(1):15-28.

Horne, D. S., & Lucey, J. A. (2017). Rennet-induced coagulation of milk. *In:* L.H Paul. et al. *Cheese*. (4th ed), Academic Press, v.1, (pp.115-143).

International Organization for Standardization - ISO. (1998). *15214:1998. Microbiology of food and animal feeding stuffs. Horizontal method for the enumeration of mesophilic lactic acid bacteria. Colony-count technique at 30°C*. Available in: <https://www.en-standard.eu/bs-iso-15214-1998-microbiology-of-food-and-animal-feeding-stuffs-horizontal-method-for-the-enumeration-of-mesophilic-lactic-acid-bacteria-colony-count-technique-at-30-c/?gad_source=1&gclid=CjwKCAiA29auBhBxEiwAnKcSqm-XVVDENxDft6h8KpP5mEL66QQR3hDIa6lhwIfr65B6RFjwTqmHbBoCLz8QAvD_BwE>.

International Organization for Standardization - ISO. (2001). *ISO 16649-2:2001(en) Microbiology of food and animal feding stuffs-Horizontal method for the detection and enumeration of β-glucuronidase-positive Escherichia coli. Part 2, Colony-count technique a 44°C using 5-brome-4chloro-3-indoly-beta-Dglucuronide, Geneve, Switzerland*. Available in: <https://www.iso.org/obp/ui/en/#iso:std:iso:16649:-2:ed-1:v1:en>.

International Organization for Standardization - ISO. (2008). *21527-1:2008 (en). Microbiology of food and animal feeding stuffs-Horizontal method for the enumeration of yeasts and moulds-Part 1: Colony count technique in products with water activity greater than 0.95*. Available in: <https://www.iso.org/obp/ui/#iso:std:iso:21527:-1:ed-1:v1:en>.

International Organization for Standardization - ISO. (2017). 21528-2:2017. Microbiology of the food chain. Horizontal method for the detection and enumeration of Enterobacteriaceae Colony-count technique. Available in:<https://www.en-standard.eu/bs-en-iso-21528-2-2017-microbiology-of-the- food-chain-horizontal-method-for-the-detection-and-enumeration-of-enterobacteriaceae-colony-count-technique/?gad_source=1&gclid=CjwKCAiA29auBhBx EiwAnKcSqk7JRhENvMa6s94pjtGm 3dtRgeV00rCB4pPd OWRvetAOr0ELFl5 v4hoCBZcQAvD_BwE>.

International Organization for Standardization - ISO. (2006). *4832:2006. Microbiology of food and animal feding stuffs-Horizontal method for the enumeration on coliforms-Colony-count technique*. Available in: <https://www.iso.org/standard/38282.html>.

International Organization for Standardization - ISO. (2021). 6888-1, 2021. Microbiology of food and animal feeding stuffs-Horizontal method for the enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species)-Part 1: technique using Baird-Parker agar medium. Available in:< https://www.iso.org/standard/76672.html>.

International Organization for Standardization- ISO. (2006). *5943:2006/IDF 88:2006; Cheese and processed cheese product-determination of chloride conten-potentiometric titration method*. ISO: Geneva, Switzerland, 2006. Available in:< chrome-extension://efaidnbmnnnibpcajpcglclefindmkaj/https://cdn.standards.iteh.ai/samples/43922/41446e4ae977424a9f4b6453101325d3/ISO-5943-2006.pdf>.

Kabir, S. et al. (2025). Exploration of the micellization behavior of sodium dodecyl sulfate in aqueous solution of gastric enzyme pepsin: Assessment of the consequences of sodium electrolytes and temperature. *International Journal of Biological Macromolecules*, *291*:138990.

Kageyama, T. (2002). Pepsinogens, progastricsins, and prochymosins: Structure, function, evolution, and development. *Cellular and Molecular Life Sciences*, *59*(2):288-306.

Kara, R., & Akkaya, L. (2013). Investigation of *Brucella abortus* and *Brucella melitensis* at Cheeses in Afyonkarahisar, Turkey. *British Journal of Dairy Sciences*, *3*(1):5-8.

Karaca, O. B., & Güven, M. (2004). Mikrobiyolojik kaynaklı proteolitik ve lipolitik enzim kullanımının beyaz peynirlerin özellikleri ve olgunlaşma hızları üzerine etkileri). *Gıda*, *7*(8):125.

Karaca, O. B., & Güven, M. (2010). Lipolitik ve proteolitik enzim kullaniminin beyaz peynirlerin özellikleri üzerine etkileri. *Cukurova University Journal of Natural and Applied Sciences*, *22*(3):107-116.

Kaya, O., & Patır, B. (2019). Investigation of some quality characteristics of rennets of animal origin sold in Elazığ province. *FUSABIL*, *33*(3):161-168.

Lee, K. D., & Warthesen, J. J. (2010). Mobile phases in reverse-phase HPLC for the determination of bitter peptides in cheese. *Journal of Food Science*, *61*(2):291-294.

Li, Y. et al. (2024). Investigation on the interaction mechanism of trilobatin with pepsin and trypsin by multi-spectroscopic and molecular docking methods. *Journal of Molecular Liquids*, *415*:126424.

Li, M. et al. (2025). Characterization of different types of micellar casein isolates and comparison of their enzymatic coagulation behavior by chymosin and pepsin. *Food Research International*, *221*:117382.

Liu, X. et al. (2021). Advances in research on calf rennet substitutes and their effects on cheese quality. *Food Research International*, *149*:110704.

McSweeney, P. L. H. (2004). Biochemistry of cheese ripening: Introduction and overview. *International Journal of Dairy Technology*, *57*(2-3):127-144.

Mir Khan, U., & Selamoglu, Z. (2020). Use of enzymes in dairy industry: a review of current progress. *Archives of Razi Institute*, *75*(1):131.

Moynihan, A. C. et al. (2014). Effect of camel chymosin on the texture, functionality, and sensory properties of low-moisture, part-skim Mozzarella cheese. *Journal of Dairy Science*, *97*(1):85-96.

Oluk, A. C. (2023). Effect of production variations on the composition, textural and microstructural properties, and volatile compounds of Turkish white cheese during ripening. *LWT*, *173*:114348.

Oštarić, F. et al. (2022). Challenging sustainable and innovative technologies in cheese production: A review. *Processes*, *10*(3):529.

Öner, Z., & Arısoy, Z. (2019). Effect of different coagulating enzymes on the chemical and microbiological properties of white cheese produced by ultrafiltration technique. *Kırklareli University Journal of Engineering and Science*, *5*(1):68-86.

Öner, Z., & Sarıdağ, A. M. (2019). The changes during maturatıon of the white cheese produced from goat milk. *Food*, *44*(3):523-533.

Özalp, E. et al. (1979). Research on the hygiene index of some microorganisms in brine white cheese made with cow's milk (Salamuradaki tuz miktarının inek sütü ile yapılan beyaz peynirler üzerine etkisi). *Ankara Universitesi Veteriner Fakültesi Dergisi*, 26(03.04):277-286. Available in:< https://doi.org/10.1501/Vetfak_0000001021>

Özcan, Y., Artık, N., & Aloğlu, H. Ş. (2020). The effect of different starter culture use on the lactose changes of white cheese during ripening. *Kırklareli University Journal of Engineering and Science*, *6*(1):71-83.

Rocchetti, G., et al. (2024). Rennets differing in chymosin-to-pepsin ratio shape the metabolomic and sensory profile of Grana Padano PDO cheese during ripening. *Food Research International*, *195*:114958.

Rolet-Repecaud, O. et al. (2017). Quantification of pepsin in rennet using a monoclonal antibody-based inhibition ELISA. *Food Science and Technology*, *76*:190-196.

Simal, S. et al. (2001). Water and salt diffusion during cheese ripening: Effect of the external and internal resistances to mass transfer. *Journal of Food Engineering*, *48*(3):269-275.

Soltani, M. et al. (2022). Perspectives and recent innovations on white cheese produced by conventional methods or ultrafiltration technique. *International Dairy Journal*, *125*:105232.

Sousa, M. J., Ardö, Y., & McSweeney, P. L. H. (2001). Advances in the study of proteolysis during cheese ripening. *International Dairy Journal*, *11*(4-7):327-345.

Stocco, G. et al. (2025). Effects of animal rennet, fermentation-produced chymosin, and microbial coagulants on bovine milk coagulation properties. *Journal of Dairy Science*, *108*(5):4614-4625.

Tarakçı, Z., & Akyüz, N. (2009). Effects of packaging materials and filling methods on selected characteristics of Otlu (Herby) cheese. *International Journal of Food Properties*, *12*(3):496-511.

Temiz, H. et al. (2007). The partial purification and properties of pepsin obtained from Turkey proventriculus. *Biotechnology and Bioprocess Engineering*, *12*(4):450-456.

Turkish Food Codex (TFC). (2015). Cheese Communiqué. Ministry of Food, Agriculture and Livestock, Official Gazette of the Republic of Turkey (Number: 29261), Ankara, Türkiye. Available in: <https://www.resmigazete.gov.tr/eskiler/2015/02/20150208-16.htm>.

Turkish Standards Institution - TSI. (2023). (TS 591). *White cheese standard*. Ankara, Türkiye.

Available in: <https://www.akib.org.tr/files/downloads/2022/09/dd94613114b940dd94613114b940dd94613114b940dd946131.pdf >.

Türkmen, D., & Güler, Z. (2022). Influence of chymosin type and brine concentration on chemical composition, texture, microstructural and colour properties of Turkish white cheeses. *International Dairy Journal*, *133*:105408.

Tüzün, C. G., & Çiftçi, İ. (2010). Kanatlılarda sağlıklı bağırsak mikroflorası gelişimi üzerine beslemenin etkileri. *Tavukçuluk Araştırma Dergisi*, *9*(1):48-55.

Uğur, E., & Öner, Z. (2023). Effects of coagulants on peptide profiles and ACE-inhibitory activity in ultrafiltered white (Beyaz) cheese. *Journal of Food Composition and Analysis*, *123*:105622.

Uniacke-Lowe, T., & Fox, P. F. (2017). Chymosin, pepsins and other aspartyl proteinases: Structures, functions, catalytic mechanism and milk-clotting properties. In L. H. Paul. et al. *Cheese*. Academic Press. (pp.69-113).

United States Department of Agriculture - USDA/FSIS. (2011). *Microbiology laboratory guidebook*. Metot 3.01. Quantitative analysis of bacteria in foods as sanitary indicators. Available in <https://www.fsis.usda.gov/news-events/publications/microbiology-laboratory-guidebook>.

Vapur, U. E. (2021). Usage suggestions of different origins of rennet in cheese production, advances in rennet technology and overview of thermolabile microbial rennet. *Akademik Et ve Süt Kurumu Dergisi*, (1):38-51.

Wehaidy, H. R., et al. (2023). The catalytic and kinetic characterization of *Bacillus subtilis* MK775302 milk clotting enzyme: Comparison with calf rennet as a coagulant in white soft cheese manufacture. *Journal of Genetic Engineering and Biotechnology*, *21*(1):61.

Yadav, S., & Jha, R. (2019). Strategies to modulate the intestinal microbiota and their effects on nutrient utilization, performance, and health of poultry. *Journal of Animal Science and Biotechnology*, *10*:2.

Yang, M. et al. (2022). Kinetics of pepsin-induced hydrolysis and the coagulation of milk proteins. *Journal of Dairy Science*, *105*(2):990-1003.

Yegin, S., & Dekker, P. (2013). Progress in the field of aspartic proteinases in cheese manufacturing: structures, functions, catalytic mechanism, inhibition, and engineering. *Dairy Science & Technology*, *93*:565-594.

Yetişmeyen, A. et al. (1998). Ultrafiltrasyon tekniği ile salamura Beyaz peynir üretiminde kalite üzerine değişik maya enzimlerinin etkisi. *Gıda*, *23*(1):3-9.

Yilmaztekin, M., Özer, B. H., & Atasoy, A. F. (2004). Survival of *Lactobacillus acidophilus* LA-5 and *Bifidobacterium bifidum* BB-02 in white-brined cheese. *International Journal of Food Sciences and Nutrition*, *55*(1):53-60.

Zareie, Z. et al. (2025). Enhancing the microbial dynamics, volatile profile, and ripening efficiency of white brined cheese using *Lactiplantibacillus plantarum* L33 as a probiotic co-culture. *Food Research International*, *203*:115912.