# Some properties of Tomas cheese and identification of lactic acid bacteria isolated from Tomas cheese

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**Abstract:** In this research, some properties of 16 Tomas cheese samples, collected from different areas in the Elazig province in Turkey, were investigated. The dry matter, fat, salt, ash, protein, titratable acidity, ripening index and pH changed between 39.70- 46.01%; 16.20-21.56%; 3.31- 4.95%; 4.41-6.90%;11.80-23.83%; 0.9-1.8%; 13.18-42.04%; 4.5-6.03, respectively. The total aerobic mesophilic bacteria, yeast-mould and lactic acid bacteria(on MRS and M17 agar) count changed between 6.67-9.01 log cfu/g; 5.30-8.39 log cfu/g; 6.25-8.91 log cfu/g; 6.56- 9.01 log cfu/g, respectively. Coliforms were determined as <1 in all the samples. In the classification was 6 isolates(30%) Lactobacillus paracasei subsp. paracasei, 5 isolates(25%) Lactobacillus brevis, 4 isolates(20%) Lactobacillus plantarum, 2 isolates(10%) Lactobacillus collinoides, 2 isolates(10%) Lactobacillus curvatus subsp. curvatus and one of the isolates(5%) Lactobacillus salivarius. According to the results of the identification of coccal isolates, 16 of the isolates(66.6%) were identified as Enterococcus faecium and 8 of isolates(33.3%) were identified as Lactococcus lactis subsp. lactis. **Keywords:** Tomas cheese, local cheese, lactic acid bacteria

### I. Introduct 🗆 on

Cheese has a history of approximately 8000-9000 years. It arose between the Euphrates and Tigris rivers [1]. That area is part of Iran, Iraq and Turkey[2]. Today, cheese type in the world are known to be more than 4000[3]. Aroma and texture of these cheeses are different from each other. Some of these cheeses are consumed as fresh and some cheeses is consumed after a ripening period under certain conditions [4]. In Turkey, 193 varieties of cheese are produced [5]. Tomas cheese is traditionally produced in Eastern Anatolia region in Turkey. Tomas is a type of Tulum cheese (brynza). First, yogurt produced from sheep and goat's milk is subjected to churning. Later, buttermilk obtained from yoghurt is heated in order to obtain skim-milk cheese. The skim-milk cheese is placed in cloth bags and filtered. Later, it is salted and enriched by adding ingredients such as butter, cream and yoghurt. The mixture is poured and pressed into a leather tulum. Then the mix is matured for 1-3 month. After ripening, it is kneaded and again poured and pressed into a leather tulum. After these operations, the cheese is ready for consumption. Tomas cheese does not have any production standards. Because of small family plant production, its chemical and microbiological quality completely related to the skills of the people engaged in the production. In particular, mold spores infecting the clot in the production environment lead to the formation of different colors, flavors and aromas in the cheese during ripening [6]. Tomas cheese, like all other traditional cheeses, is sometimes manufactured in scant hygienic conditions using different methods. The natural microflora affects the organoleptic properties and the safety of the product. The studies conducted on Tomas cheese are insufficient. This study was performed to determine some properties of the Tomas cheese samples. Tomas cheese isolates can be potentially used during the commercial production of Tomas cheese or other fermented dairy products. This contribute to a continuity of cultural heritage and economic development.

## II. Material and Method

The Tomas cheese samples were gathered from different households in the Elazig province of Turkey.

The samples were analyzed in duplicate for determination of acidity, pH, dry matter, fat, ash, salt, total nitrogen and water soluble nitrogen. The dry matter content was detected by drying of samples at  $100\pm2^{\circ}C$  for 4 h [7]. The fat content was determined through butyrometric method[8]. To determine the ash content, portions (2g) of the cheese samples were turned into ash in a porcelain crucible at 550°C[8]. The pH value was measured using a digital pH meter(Hanna Instruments pH 211 microprocessor pH meter) [8]. The titratable acidity(lactic acid, %) and the salt was detected by the titration method [9]. The total nitrogen was analyzed through the Kjeldahl method practiced by Gripon et al. [10]. In the protein content calculation was used Kjeldahl Nx6.38 as formul. The water soluble nitrogen (WSN) was detected through the Kjeldahl method [11]. The nitrogen content in the extracted cheese was regarded as ripening index and stated as a percentage of the total nitrogen (WSN/TN, %) [12].

Total aerobic mesophilic bacteria counts were examined using plate count agar (PCA). Incubation was performed at 30°C for 48 h. Yeast and mould counts were examined using potato dextrose agar (PDA). Incubation was continued for 5 days at 25°C [13,14]. de Man Rogosa Sharpe (MRS) agar (anaerobically for 72 h at  $30\pm1^{\circ}$ C) [15] and M17 agar (aerobically for 48h at  $30\pm1^{\circ}$ C) were used for lactic acid bacteria counts [16]. Violet red bile (VRB) agar was used to determine coliform group bacteria counts and incubation was continued for 24 h at 37°C [14].

The cell morphology of all lactobacilli isolates was observed using phase contrast microscopy. The isolates were tested for gram reaction. The catalase reaction was determined by adding 3% H<sub>2</sub>O<sub>2</sub> on the isolates. All lactobacilli isolates, for the gas production from the glucose was grown in the MRS broth. The salt tolerance was observed in MRS broth supplemented with the addition of 2% and 4% NaCl. A growth rate at  $15^{\circ}$ C or  $45^{\circ}$ C was tested in the MRS broth. For growth rate, incubation was continued for three days. The hydrolysis of the arginine was examined in the MRS broth. MRS broth was containing 0.2% (v/v) sodium citrate replacing the ammonium citrate and 0.3% of arginine, and not containing glucose and meat extract. Nessler's reagent was used to determine of ammonia [17]. Fermentation or assimilation of various carbon sources was tested. For the lactobacilli was used API 50 CHL (BioMérieux SA. Marcy l'Etoile. France). API 20 STREP was used for lactococci.

The differences between the 16 Tomas cheese samples were analyzed using SPSS 15 as statistical software (SPSS Corporation, Chicago, IL, USA). Multiple comparisons were made between means by ANOVA techniques. Duncan's test were used to compare means [18].

#### III. Results and Discussions

The physicochemical properties of the Tomas cheese samples are indicated in Table 1. According to the results of the statistical evaluation, remarkable differences between the chemical properties of cheese samples were observed. The minimum, maximum and mean ripening index values were 13.18%, 42.04 % and 23.37%, respectively. The microbiological properties of Tomas cheese samples are indicated in Table 2. The coliform group bacteria counts were determined as <1 log cfu/gin all the samples. Forty-four lactic acid bacteria from 16 Tomas cheese samples were isolated and identified. Nearly 45.5% of lactic acid bacteria were *Lactobacillus* (growing on MRS agar). Other lactic acid bacteria (54.5%) identified were *Enterococcus* and *Lactococcus* (growing on M17 agar). In the classification,6 isolates (13.6%) were identified as*Lactobacillus plantarum*; 2 isolates(4.5%) were identified as*Lactobacillus collinoides*; 2 isolates(4.5%) *Lactobacillus plantarum*; 2 isolates(4.5%) were identified as*Lactobacillus collinoides*; 2 isolates(4.5%) *Lactobacillus salivarius*. In the classification of coccal isolates, 16 of the isolates(36.4%) were identified as *Enterococcus faecium* and 8 of isolates(18.2%) identified were *Lactococcus lactis* subsp. *Lactos*. Biochemical properties of lactic acid bacteria (Lactobacillus) are indicated in Table 3.

|         |                           |                            | FF                        |                         | sumples and the          |                         |                           |
|---------|---------------------------|----------------------------|---------------------------|-------------------------|--------------------------|-------------------------|---------------------------|
| Samples | Dry matter<br>(%)         | Fat<br>(%)                 | Ash<br>(%)                | Salt<br>(%)             | Protein<br>(%)           | Acidity<br>(%)          | рН                        |
| 1       | $43.95\pm0.03^{\rm c}$    | $16.20 \pm 0.07^{\rm h}$   | $6.55 \pm 0.11^{b}$       | $3.98\pm0.24^{e}$       | $19.80 \pm 0.01^{\circ}$ | 1.3 ±0.05 <sup>ef</sup> | 4.75±0.15 <sup>bcd</sup>  |
| 2       | $39.70\pm0.10^{\rm i}$    | $19.46 \pm 0.35^{\circ}$   | $5.14\pm0.23^{\rm f}$     | $4.27\pm0.28^{\rm d}$   | $13.89 \pm 0.25^{1}$     | $1.1\pm0.05^{\text{g}}$ | $4.63 \pm 0.05^{de}$      |
| 3       | $46.01\pm0.10^{a}$        | $15.65 \pm 0.71^{\circ}$   | $5.20\pm0.29^{\text{ef}}$ | $3.91\pm0.14^{e}$       | $23.83 \pm 0.08^{a}$     | 1.2 ±0.10 <sup>fg</sup> | 4.76±0.01 <sup>bcd</sup>  |
| 4       | $43.39 \pm 0.04^{de}$     | $18.50 \pm 0.35^{\rm f}$   | $6.25\pm0.17^{\rm c}$     | $4.62\pm0.08^{bc}$      | $17.00 \pm 0.02^{f}$     | 1.2±0.10 <sup>fg</sup>  | 4.79±0.01 <sup>bcd</sup>  |
| 5       | $40.24 \pm 0.09^{\circ}$  | $21.25\pm0.06^{\text{b}}$  | $6.55 \pm 0.11^{b}$       | $4.95\pm0.22^{a}$       | $11.80 \pm 0.01^{j}$     | 1.7±0.05 <sup>ab</sup>  | $4.62\pm0.06^{de}$        |
| 6       | $43.35 \pm 0.02^{de}$     | $19.01 \pm 0.41^{d}$       | $6.37 \pm 0.05^{bc}$      | $4.53\pm0.13^{\rm c}$   | 16.98 ±0.03 <sup>f</sup> | 1.6±0.05 <sup>cd</sup>  | 4.78±0.03 <sup>bcd</sup>  |
| 7       | $41.52\pm0.03^{h}$        | $19.54 \pm 0.35^{\circ}$   | $5.79 \pm 0.05^{d}$       | $4.65 \pm 0.47^{bc}$    | $14.56 \pm 0.08^{h}$     | 1.5±0.05 <sup>cd</sup>  | 4.89±0.005 <sup>bc</sup>  |
| 8       | $41.98\pm0.10^{\text{g}}$ | $18.05\pm0.71^{\text{g}}$  | $6.90 \pm 0.11^{a}$       | $4.51\pm0.25^{\rm c}$   | 16.05 ±0.50 <sup>g</sup> | 1.5±0.01 <sup>cd</sup>  | 4.87±0.005 <sup>bc</sup>  |
| 9       | $43.03\pm0.12^{\text{e}}$ | $18.58\pm0.08^{\rm ef}$    | $6.43 \pm 0.11^{bc}$      | $4.94\pm0.12^{a}$       | $17.08 \pm 0.35^{\rm f}$ | 1.4±0.01 <sup>de</sup>  | $4.51\pm0.04^{e}$         |
| 10      | $41.74 \pm 0.15^{gh}$     | $21.05 \pm 0.06^{b}$       | $6.49 \pm 0.05^{bc}$      | $4.95\pm0.37^{a}$       | $12.87 \pm 0.25^{i}$     | 0.9 ±0.10 <sup>h</sup>  | $6.03 \pm 0.02^{a}$       |
| 11      | $44.81 \pm 0.21^{b}$      | $18.45\pm0.25^{\rm f}$     | $5.44\pm0.05^{e}$         | $4.74 \pm 0.11^{b}$     | 20.38 ±0.09 <sup>b</sup> | $1.8\pm0.05^{\rm a}$    | 4.73±0.05 <sup>bcd</sup>  |
| 12      | $42.42 \pm 0.07^{\rm f}$  | 19.68 ±0.05 <sup>c</sup>   | $4.41 \pm 0.02^{g}$       | $3.31 \pm 0.83^{\rm f}$ | 17.36 ±0.06 <sup>e</sup> | 1.5±0.10 <sup>cd</sup>  | $4.94 \pm 0.02^{b}$       |
| 13      | $43.54\pm0.08^{\text{d}}$ | $18.99\pm0.08^{\rm d}$     | $5.79 \pm 0.05^{d}$       | $4.61 \pm 0.17^{bc}$    | 17.68 ±0.25 <sup>d</sup> | 1.6±0.15 <sup>bc</sup>  | 4.79±0.06 <sup>bcd</sup>  |
| 14      | $43.09 \pm 0.02^{e}$      | $18.78\pm0.07^{\text{de}}$ | $6.37 \pm 0.05^{bc}$      | $4.65 \pm 0.49^{bc}$    | $16.98\pm0.09^{\rm f}$   | 1.6±0.10 <sup>bc</sup>  | $4.90 \pm 0.02^{b}$       |
| 15      | $40.19\pm0.10^{\rm i}$    | $19.46\pm0.02^{c}$         | $5.03 \pm 0.11^{\rm f}$   | $3.85\pm0.45^{e}$       | 14.34 ±0.08 <sup>h</sup> | $1.8\pm0.05^{a}$        | 4.79±0.05 <sup>bcd</sup>  |
| 16      | $40.35 \pm 0.04^{\circ}$  | $21.56\pm0.21^{a}$         | $5.79\pm0.05^{\rm d}$     | $4.26\pm0.30^{d}$       | $12.02\pm0.05^j$         | 1.5±0.10 <sup>cd</sup>  | 4.68 ±0.02 <sup>cde</sup> |

Table 1. Some physicochemical properties of Tomas cheese samples and their statistical evaluations

a, b, c, d, e, f, g, h, i, i, j; There is no difference between group means with the same letter.

The reason for remarkable differences between the properties of cheese samples may be the absence of any production standard and production recipe for Tomas cheese. Besides, the heat treatment applied to the milk and milk type may cause the main difference between cheese samples. The manufacturing steps is very effective

on the quality and properties of the final cheese. The texture and flavor of cheese develops with the progress of ripening, which characterise the different cheese varieties. The biochemical changes that take place during ripening are very complex. Starter cultures and enzymes of starter cultures, non starter lactic acid bacteria and their enzymes also participate in the ripening process [19].

| Samples | Total aerobic mesophilic<br>bacteria<br>(log cfu/g) | Yeast and<br>moulds<br>(log cfu/g) | Coliform<br>bacteria<br>(log cfu/g) | Lactic acid bacteria<br>(on MRS agar)<br>(log cfu/g) | Lactic acid bacteria<br>(on M17 agar)<br>(log cfu/g) |
|---------|---|------------------------------------|-------------------------------------|--|--|
| 1       | $7.65 \pm 0.04$                                     | $7.72\pm0.06$                      | <1                                  | $7.85 \pm 0.15$                                      | $7.87 \pm 0.23$                                      |
| 2       | $8.53 \pm 0.25$                                     | $8.39\pm0.04$                      | <1                                  | $8.50\pm0.12$  | $8.59 \pm 0.01$                                      |
| 3       | $7.93 \pm 0.47$                                     | $8.16\pm0.09$                      | <1                                  | $7.99\pm0.08$  | $8.09\pm0.08$  |
| 4       | $7.67 \pm 0.06$                                     | $7.53\pm0.06$                      | <1                                  | $7.48\pm0.02$  | $7.64\pm0.18$  |
| 5       | $6.73 \pm 0.03$                                     | $6.25\pm0.24$                      | <1                                  | $6.25 \pm 0.24$                                      | $7.25 \pm 0.24$                                      |
| 6       | $7.28 \pm 0.44$                                     | $7.69\pm0.08$                      | <1                                  | $7.56\pm0.07$  | $7.63\pm0.05$  |
| 7       | $9.01 \pm 0.01$                                     | $8.60\pm0.01$                      | <1                                  | $8.91 \pm 0.02$                                      | $9.01 \pm 0.004$                                     |
| 8       | $7.15 \pm 0.15$                                     | $6.71\pm0.15$                      | <1                                  | $6.95\pm0.24$  | $7.15 \pm 0.15$                                      |
| 9       | $6.77\pm0.07$                                       | $6.58\pm0.05$                      | <1                                  | $6.99\pm0.08$  | $6.86\pm0.24$  |
| 10      | $8.10\pm0.06$                                       | $8.14\pm0.05$                      | <1                                  | $7.99 \pm 0.04$                                      | $8.37\pm0.11$  |
| 11      | $6.73\pm0.19$                                       | $5.30\pm0.30$                      | <1                                  | $6.71 \pm 0.03$                                      | $7.22\pm0.02$  |
| 12      | $6.67 \pm 0.16$                                     | $6.87\pm0.02$                      | <1                                  | $6.43\pm0.14$  | $6.56 \pm 0.17$                                      |
| 13      | $7.47 \pm 0.12$                                     | $7.07\pm0.13$                      | <1                                  | $7.25 \pm 0.24$                                      | $7.13 \pm 0.01$                                      |
| 14      | $8.12 \pm 0.11$                                     | $7.90\pm0.07$                      | <1                                  | $8.31\pm0.02$  | $8.43\pm0.03$  |
| 15      | $8.17\pm0.05$                                       | $6.45\pm0.26$                      | <1                                  | $6.73\pm0.03$  | $8.81 \pm 0.20$                                      |
| 16      | $7.70\pm0.19$                                       | $7.13\pm0.20$                      | <1                                  | $7.59\pm0.01$  | $7.32\pm0.16$  |

Table 2. Some microbiological properties of Tomas cheese samples

In this study, differences between the acidity of cheese samples were observed. According to estimates, the reason for difference between acidity values difference in ripening periods of cheeses and materials. Because, the acidity is associated with the ripening periods of cheeses. This is also true for pH values. In the study conducted by AteşandPatır[20] the pH of the Tulum cheese samples was decreased continuously during the ripening period. The pH values was determined as 5.84 and 5.46, respectively at the first day and at the ninety day as higher from pH values determined in this study. The acidity was 0.67 at the beginning of ripening. The acidity was 0.92 at the end of the ninety day as lower from pH values determined in this study. The reason for this difference may be the difference in the production methods. The duration of the ripening period of cheeses samples used in this study could have lasted longer. In this research, the reason for differences between the fat amounts of cheese samples may be the difference in the amount of cream added. Furthermore, ripening period may have influenced the amount of fat. The oxidation or lipolysis can degrade fat of cheese. The fat in cheese is important for the sensation and cheese flavor [21]. The amount of salt varied depending on the choice of manufacturers. Additionally, the difference in the ripening period may be reason for the difference in the amount of salt. Many researchers indicated that steady increase in the amount of salt in the progress of ripening [22,23]. Ateşand Patır[20] determined that the amount of salt continuously increased during the ripening period. The amount of salt of Tulum cheese in their study was (as mean values) 4.17% and 4.81%, respectively at the first day and the ninety day. The amount of salt of Tulum cheeses in this study is similar to their findings.

A total of 20 Tomas cheese samples were analyzed by Kurtetal.[24], and the acidity (SH), moisture, dry matter, protein, fat, non fat dry matter, fat in dry matter, salt and ash contents (as mean values) were 102.5, 52.5%, 47.49%, 22.26%, 18.13%, 29.37%, 38.18%, 3.05%, 3.42%, respectively. Thedry matter and protein contents are higher than the values obtained in this work. The salt and ash content is lower. The fat content is similar in both studies. The reason for difference between researches may be difference in ripening periods of cheeses and materials. Gündüz[25]produced Tomas cheese in laboratory conditions using *Penicillium roqueforti Thom.* Samples were used as experimental materials after the production, in the first, second, third, and fourth months. The total aerobic mesophilic bacteria,lactic acid bacteria, proteolitic bacteria, *Candida lipolytica*, *P. roqueforti, Lactobacillus plantarum, Lactobacillus bulgaricus, Streptococcus lactis, Streptococcus cremoris* and *S. thermophiles* count in the fourth month (as mean value) were determined as 7.36x10<sup>7</sup>, 5.82x10<sup>7</sup>, 1.54x10<sup>7</sup>, 1.26x10<sup>7</sup>, 0.84x10<sup>7</sup>, 0.54x10<sup>7</sup>cfu/g, respectively. The fat in dry matter, degree of ripening, acidity (lactic acid) and pH (as mean values) were determined as 49.91%, 47.45%, 2.098, and 6.08, respectively. The results of this study were lower than the results determined by Gündüz.

Karin Kaymagi cheese is similar to Tulum cheese and usually manufactured from sheep's milk. It is pressed into the rumen and matured. Its fat content is very high. Turgutetal.[26] were identified Lactobacillus strains from Karin Kaymagi cheese. *L. plantarum* type 1 and type 2, *L. brevis* type 1 and type 3, *L. acidophilus* type 3, *L. delbrueckii* subsp. *delbrueckii* and *L. fermentum* were identified as 47.56%, 21.9%, 9.75%, 1.97% and

2.43%, respectively. 24.4% of bacterial flora consisted of *L.mesenteroides* subsp. *mesenteroides/dextranicum* type 2 isolates. *L. plantarum* and *L. brevis* are in the dominant cultures in both studies. The mean dry matter, fat, and protein contents of the samples were found as 74.66%, 43.36% and 25.51%, respectively. The values obtained in our study are lower than these values. Because Karin Kaymagi cheese made from sheep's milk. In their study, the total aerobic mesophilic bacteria, lactic acid bacteria and yeast and mold counts were found as 6.477-8.17 log cfu/g,6.649- 8.041 log cfu/g and 3.556-5.308 log cfu/g, respectively. *S. aureus* and coliform group bacteria counts were determined under the acceptable limit (<10 log cfu/g). Except for the yeast mold count and *S.aureus*, these counts are similar to the numbers obtained in our study. Çakmakçıetal. [27] analyzed 13 samples of Karin Kaymagi cheese. The acidity (lactic acid), dry matter, fat, non fat dry matter, fat in dry matter, protein, water soluble protein, ash, salt, salt in dry matter and degree of ripening(as mean values) were determined as 1.01%, 69.10 %, 39%, 30.10%, 57.18%, 19.01%, 3.77%, 6.19%, 4.36%, 6.15% and 19.27%, respectively. The results of this study were different from the results determined by Çakmakçıetal. The reason for this could be the difference in process or materials.

| tes         | Growth  | Growt | Gas          | Growth | Growth | $NH_3$        | Catalas | Gram's   | API 50 CHL           |
|-------------|---------|-------|--------------|--------|--------|---------------|---------|----------|----------------------|
| Isolates    | at 15°C | h at  | production   | in 2%  | in 4%  | production    | e test  | staining |                      |
| Is          |         | 45°C  | from glucose | NaCl   | NaCl   | from arginine |         |          |                      |
| TB09        | -       | -     | +            | +      | +      | -             | -       | +        | Lb. brevis           |
| TB13        | +       | -     | -            | +      | +      | +             | -       | +        | Lb. plantarum        |
| TB15        | -       | -     | -            | +      | +      | +             | -       | +        | Lb. brevis           |
| <b>TB16</b> | +       | -     | +            | +      | -      | -             | -       | +        | Lb. collinoides      |
| <b>TB18</b> | +       | -     | -            | +      | +      | +             | -       | +        | Lb. paracasei subsp. |
|             |         |       |              |        |        |               |         |          | paracasei            |
| TB19        | -       | -     | -            | +      | +      | +             | -       | +        | Lb.curvatussubsp.    |
|             |         |       |              |        |        |               |         |          | curvatus             |
| TB20        | +       | -     | -            | +      | +      | -             | -       | +        | Lb. paracasei subsp. |
|             |         |       |              |        |        |               |         |          | paracasei            |
| TB24        | -       | -     | +            | +      | -      | +             | -       | +        | Lb. salivarius       |
| TB27        | +       | -     | -            | +      | +      | -             | -       | +        | Lb. paracasei subsp. |
|             |         |       |              |        |        |               |         |          | paracasei            |
| TB29        | +       | -     | -            | +      | +      | +             | -       | +        | Lb. plantarum        |
| TB34        | -       | -     | +            | +      | +      | +             | -       | +        | Lb. brevis           |
| TB39        | -       | -     | -            | +      | +      | -             | -       | +        | Lb. curvatus subsp.  |
|             |         |       |              |        |        |               |         |          | curvatus             |
| TB41        | -       | -     | -            | +      | +      | -             | -       | +        | Lb. brevis           |
| TB42        | +       | -     | -            | +      | +      | -             | -       | +        | Lb. plantarum        |
| TB43        | +       | -     | -            | +      | +      | +             | -       | +        | Lb. paracasei subsp. |
|             |         |       |              |        |        |               |         |          | paracasei            |
| TB48        | +       | -     | +            | +      | -      | -             | -       | +        | Lb. collinoides      |
| TB53        | +       | -     | +            | +      | +      | +             | -       | +        | Lb. brevis           |
| TB57        | -       | -     | -            | +      | +      | +             | -       | +        | Lb. paracasei subsp. |
|             |         |       |              |        |        |               |         |          | paracasei            |
| TB60        | +       | -     | -            | +      | +      | -             | -       | +        | Lb. paracasei subsp. |
|             |         |       |              |        |        |               |         |          | paracasei            |
| <b>TB66</b> | -       | -     | -            | +      | +      | +             | -       | +        | Lb. plantarum        |

| Table 3. Biochemical properties of lactic acid bacter | a (Lactobacillus) (+ = positive result | - = negative result) |
|---|--|----------------------|
|---|--|----------------------|

In study conducted by AteşandPatır[20] on Tulum cheeses, the total aerobic mesophilic bacteria count was described between  $1.1 \times 10^8$ - $7.7 \times 10^8$  cfu/g after the ripening process. The yeast and mould count was between  $3.4 \times 10^4$ - $2.6 \times 10^7$  cfu/g. The coliform group bacteria was detected as <10 cfu/g in one sample on the fifteenth day of ripening, as <10 cfu/gin all samples in the ninetieth day. The total aerobic mesophilic bacteria, yeast-mould, and coliform group bacteria counts obtained in this study is similar to counts determined by Ateş and Patır[20]. Coliform group microorganisms are continually decrease during ripening period. They may be lost completely disappeared at different times according to the type of cheese. pH values continually decrease during ripening [28]. In this study, it may be the reason for determined as <1 log cfu/gof coliform group bacteria count.

Kılıçand Gönç [29] isolated and identified *Lactobacillus*, *Enterococcus* and *Lactococcus* fromIzmir Tulum cheeses. They approved the use of *Lactococcus lactis* subsp. *lactis*, *Lactococcus lactis*subsp. *cremoris*, *L. casei* and *E. faecalis* in the production of Izmir Tulum cheese. Öneretal.[30] determined that lactobacilli constitute the dominant flora of in Tulum cheese. However, *Lactococcus* was determined as 15.8% in cheese flora. In the study conducted by Öksüztepeetal.[31], *L. lactis* subsp. *lactis*, *L. mesenteroides* subsp. *cremoris* and *L. lactis* subsp. *cremoris* played very important roles in the ripening of Savak Tulum cheese as the major species. The study conducted by BüyükyörükandSoyutemiz [32]was done to isolate strains (*L. lactis* subsp. *lactis* and *L. lactis* subsp. *cremoris*)belonging to the *Lactococcus* genus in İzmir Tulum cheese. *L. lactis* subsp. *lactis* were identified as 18 of the isolates(20%). Eleven isolates (12.8%) were *L. lactis* subsp. *cremoris*. In this study, *L. lactis* subsp. *lactis* was identified 18.2% (8 of isolates).

#### **IV. Conclusion**

Consequently, in this study, strains and species of several *Lactobacillus*, *Lactococcus* and *Enterococcus* species have been isolated, identified. These findings are similar to the findings of other studies. Although Tomas cheese is a type of Tulum cheese, is different from Tulum cheese in terms of materials used for production. The characterization in this study will lead the selection of suitable strains to be used in the production of Tomas cheese. These species may also be used in the production of various fermented dairy products. This may be an improvement for the dairy industry. Investigation and identification of traditional foods contribute to a continuity of cultural heritage and economic development.

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