

Research Article

MICROBIOLOGICAL QUALITY OF YOGHURT CONSUMED IN KARS

Mehmet ELMALI*

Hilmi YAMAN

Geliş Tarihi : 23.02.2004

Kabul Tarihi : 14.05.2004

Kars'ta Tüketilen Yoğurtların Mikrobiyolojik Kalitesi

Özet: Kars bölgesinde, küçük aile süt işletmelerinde üretilen ve satılan yoğurtların mikrobiyolojik kalitesini belirlemek üzere toplam 100 yoğurt örneği toplandı. 40 örnekte maya ve küf sayısı saptama sınırının altında iken ($< 10^2$), 40 örnekte 10^3 kob/ml, 10 örnekte 10^4 kob/ml ve 10 örnekte 10^5 kob/ml düzeyinde saptandı. *Enterobacteriaceae* 60 örnekte $< 10^2$, 20 örnekte 10^2 kob/ml ve 20 örnekte 10^3 kob/ml düzeyinde saptandı. Mikrokok-stafilokok ve enterokoklar ise 88 örnekte ve 92 örnekte 10^2 kob/ml, 12 ve 8 örnekte ise sırasıyla 10^3 kob/ml düzeyinde saptandı. Analiz edilen yoğurt örneklerinde maya ve küf, *Enterobacteriaceae*, mikrokok ve stafilocok, enterokok ve koliformlar için ortalama değerler (\log_{10} kob/ml) sırasıyla 4.77, 2.75, 1.34, 1.44 ve 0.52 kob/ml olarak saptandı. 100 yoğurt örneğinin 2'sinden koagulaz pozitif stafilocok izole edildi ve bunlar *Staphylococcus aureus* (*S. aureus*) olarak tanımlandı. İzole edilen bütün koliformlar *Escherichia coli* (*E. coli*) olarak tanımlandı. Türk Standartları Enstitüsü yoğurt standardına göre (TS, 1330) analiz edilen yoğurtların % 10.0'u koliform ve % 60.0'ı maya-küf bakımından düşük hijyenik kaliteye sahiptir. Bu çalışmadaki sonuçlar Kars bölgesinde küçük aile işletmelerince üretilen ve satılan yoğurtların üretim yapıldığı yerlerin gerekli hijyene sahip olmadığı veya yetersiz olduğunu işaret etmektedir.

Anahtar Kelimeler: Yoğurt, Mikrobiyolojik Kalite, Hijyen

Summary: A total of 100 yoghurt samples were collected in order to investigate the microbiological quality of yoghurts produced by small family run dairies in Kars region, Turkey. Numbers of yeasts and mould in 40 samples were below the detection level of $< 10^2$ cfu/ml, 10^3 cfu/ml in 40 samples and 10^4 - 10^5 cfu/ml in 10 samples. *Enterobacteriaceae* spp. were detected at a level of $< 10^2$ cfu/ml in 60 samples, 10^2 cfu/ml in 20 samples and 10^3 cfu/ml in 20 samples. *Micrococci-Staphylococci* and *Enterococci* were found to be below the level of $< 10^2$ in 88 and 92 samples, 10^2 cfu/ml in 12 and 8 samples, respectively. Bacterial counts (\log_{10} cfu/ml) were 4.77, 2.75, 1.34, 1.44 and 0.52 for yeast and mould, *Enterobacteriaceae*, *Micrococci* and *Staphylococci*, *Enterococci* and coliforms spp., respectively. Out of 100 yoghurt samples, coagulase positive *Staphylococci* were isolated from two samples and these were identified as *Staphylococcus aureus* (*S. aureus*). All of the coliforms isolated were identified as *Escherichia coli* (*E. coli*). According to Turkish Standards Institute (TS, 1330), 10.0 % and 60.0 % of yoghurts did not fit into criteria of Turkish

* Kafkas Üniversitesi Vet. Fak. Besin Hijyeni ve Teknolojisi Anabilim Dalı, KARS.

Standards Institute due to excessive numbers of coliforms, yeast and mould, respectively. The results of this study indicate that yoghurts produced and sold by family run dairies had poor hygienic quality.

Key words: Yoghurt, Microbiological Quality, Hygiene

Introduction

Yoghurt is a nutritious fermented milk product which has high biological value and it can be digested easily. Yoghurt is frequently consumed in Mediterranean countries including Turkey. However, it is a more commercialised product in Europe, whereas it is still made at home and produced in family run dairies widely in Turkey. In order to produce good quality yoghurt, it is required to monitor the chemical and microbiological composition of milk to be used in the production. Poor chemical and microbiological quality of milk, contamination during production, unsuitable storage, lack of personal hygiene and premises will effect the chemical and microbiological quality of yoghurt and its shelf life. Studies carried out on the microbiological quality of yoghurt in Turkey (3,5,11,13) were mainly based on products sold at markets in big developed cities. Therefore, this study aimed to investigate the microbiological quality of yoghurts produced by family run dairies in a rural area in Kars district of Turkey.

Materials and Methods

Samples

A total of hundred yoghurt samples (5 kg each) were collected in quantities of 25 kg for each different four small dairies across Kars region. Samples were transported to laboratory immediately in cold chain (+4°C) and subsequently analysed within 2 hr.

Microbiological analysis:

Ten (10) ml of yoghurt samples were homogenised in a stomacher (Bagmixer) for 2 min in 90 ml 0.1 % (wt/vol) peptone water (Oxoid CM 9). Serial dilutions up to 10^{-5} were carried out using the same solution. The pour plate technique was used to prepare plates for enumeration of coliform spp. microorganisms and drop technique was used for enumeration of other microorganisms. *Staphylococci* and *Micrococci* were isolated on Baird Parker Agar (Oxoid CM 275) aerobically at 37°C for 24-48 hr. Typical black colonies with zones around and atypical black colonies were considered as *Staphylococci*. Small, brown-black colonies without zones around were considered as micrococci. Colonies that were isolated as *Staphylococci* were inoculated into Brain Heart Infusion Broth (Merck 1.10493), and were incubated at 37°C for 24-48 hr. Subsequently, coagulase test (Merck 1.3306) was performed to determine coagulase positive *Staphylococci* and these isolates were then inoculated onto DNase Agar (Merck 1.10449) for identification of *S. aureus* which was confirmed by cell morphology, Gram stain, catalase activity, sensitivity to lysostaphin. *Enterobacteriaceae* counts were performed on Violet Red Bile Glucose Agar (Oxoid CM 485) which was incubated

aerobically at 37°C for 24-48 hr. Pink-red colonies with precipitation were taken into consideration. *Enterococci* were counted on Slanetz Bartley Medium (Oxoid CM 377) incubated aerobically at 37°C for 24-48 hr, red colonies were taken into consideration. Pink-red colonies with precipitation isolated on Violet Red Bile Lactose Agar (Oxoid CM 107) were grown on Endo Agar (Oxoid CM 479) and were incubated at 37°C for 24-48 hr. After the incubation colonies that had shiny metallic colour were chosen and IMVIC test was performed for the identification of *E. coli*. Rose Bengal Chloramphenicol Agar was used to isolate yeast-mould, which was incubated aerobically at 25°C for 4-5 days. All media were purchased from Oxoid Ltd. (Hampshire, UK). Colonies between 30-300 (Pour plate technique) and 5-50 (Drop plate technique) were counted and mean counts were calculated.

Results

The results obtained from microbiological examination of 100 yoghurts produced and sold by four family run dairies are summarised in Table 1. Grouping of the 100 yoghurts according to the degree of contamination can be seen in Table 2.

Table 1. The logarithmic numbers (\log_{10}) of detected microorganisms from yoghurt samples (cfu/ml).
Tablo 1. Yoğurt örneklerinden (kob/ml) saptanan mikroorganizmaların logaritmik sayısı (\log_{10}).

Sample	Parameter	Coliform spp	Yeast and mould	<i>Enterobacteriaceae</i>	<i>Micrococci-Staphylococci</i> spp.	<i>Enterococci</i> spp.
	Mean	0.52	4.77	2.75	1.34	1.44
Yoghurt		(3.3×10^0)	(5.9×10^4)	(5.7×10^2)	(2.2×10^1)	(2.8×10^1)
n:100	Sx	0.019	0.035	0.027	0.024	0.024
	Min	0.00	<2.30	<2.30	<2.30	<2.30
	Max	1.47	5.94	3.64	2.60	2.60

Yeast and mould were detected at the level of $<10^2$ cfu/ml in 40 samples, 10^3 cfu/ml in 40 samples, 10^4 and 10^5 cfu/ml in 10 samples. *Enterobacteriaceae* spp. were found in 60 samples a level of less than $<10^2$ cfu/ml, for the remaining of samples 10^2 and 10^3 cfu/ml were counted for each 20 samples. *Micrococci* and *Staphylococci* were $<10^2$ cfu/ml in 88 samples whereas 12 samples had 10^2 cfu/ml. *Enterococci* were found at the levels of $<10^2$ in 92 samples and it was 10^2 cfu/ml in 8 samples. The average counts of yeast and fungi were 5.9×10^4 cfu/ml or $4.77 \log_{10}$ cfu/ml. *Micrococci* and *Staphylococci* were found to be 2.6×10^1 cfu/ml or $1.41 \log_{10}$ cfu/ml. *Enterococci* gave an average number of 2.4×10^1 cfu/ml or $1.38 \log_{10}$ cfu/ml and it was 3.3×10^0 cfu/ml or $0.52 \log_{10}$ cfu/ml for coliforms. Coagulase (+) *Staphylococci* were isolated from two yoghurt samples and were identified as *S. aureus*. Out of 34 coliforms, 10 isolates were identified as *E. coli*. On the basis of Turkish Standards Institute (TSE) (2), microbiological standard of yoghurt (TSE, 1330), 10.0 % and 60.0 % of analysed

yoghurt samples did not fit into Turkish Standards Institute based on coliform (*E. coli*) and yeast-mould contamination, respectively. The pH of yoghurt samples ranged between 4.2-5.0.

Table 2. The level and distribution of isolated microorganisms from yoghurt samples (cfu/ml)

Tablo 2. Yoğurt örneklerinden izole edilen mikroorganizmaların dağılımı ve düzeyi (kob/ml).

Sample	Level of microorganism spp cfu/ml	Coliform spp.	Yeast and mould	<i>Enterobacteriaceae</i>	<i>Micrococci-Staphylococci</i> spp.	<i>Enterococci</i> spp.
	<10	66				
	10 ¹	24				
Yoghurt	<10 ²	total 90	40	60	88	92
n:100	10 ²	10	-	20	12	8
	10 ³	-	40	20	-	-
	10 ⁴	-	10	-	-	-
	10 ⁵	-	10	-	-	-

Discussion

In this study, the average number of coliform bacteria were found to be 3.3×10^0 cfu/ml or $0.52 \log_{10}$ cfu/ml with the percentage of 10.0%. Many studies have been done regarding microbiological quality of yoghurt (3,6,7,11,12,14). Cakiroglu (3) reported isolation of 8.0 % coliform group bacteria that showed an average count of 2.2×10^2 cfu/ml. 6.66 % of the isolates was identified as *E. coli*. Khalaf and Shareef (7) isolated *E. coli* at the percentage of 6.0 % from 100 samples. Salih et al. (12) examined 42 yoghurt samples and coliforms were isolated from 7.1 % of the samples which had the average number of 7.2×10^2 cfu/ml. Oz (11) analysed 50 yoghurt samples, of which contained an average number of 1.2×10^2 cfu/ml coliforms. Sireli and Ozdemir (14) examined 50 fruit yoghurt samples and found similiar results. Ibrahim et al. (6) reported the presence of coliforms in the range of $<1.0 \times 10^1 - 4.7 \times 10^5$ cfu/ml whereas Yazici (16) reported $<1.0 \times 10^1 - 1.2 \times 10^4$ cfu/ml coliforms in their study in which 60 yoghurt samples were examined. Aboul-Khier et al. (1) isolated coliforms from 47.6 % of 21 samples while Mann (8) examined 870 yoghurt samples and isolated *E. coli* from 99.0 % of samples. Dayisoğlu (4), isolated coliform bacteria from 35 % samples in the range of $4.0 \times 10^2 - 1.0 \times 10^3$ cfu/ml whereas Tayar et al. (15) reported the isolation of coliforms from 25 5 of the samples in the range of $2.9 \times 10^2 - 3.0 \times 10^4$ cfu/ml in Bursa. The results of these studies (3,6,8,9,12) have shown that their total isolation rates of coliforms are lower than what was found in this study. On the contrary our results are lower than the results reported by Dayisoğlu (4) and Tayar et al. (15) This may be explained by low numbers of yoghurt samples included by these researchers. However, the mean levels of contamination (3,7,11,12,14) and detection range (1,6,8) are higher. These can be

related to the quality of raw milk, unsuitable storage of milk, lack of personal (workers) and environmental hygiene.

In this study, the average levels of yeast and mould were obtained as 5.9×10^4 cfu/ml or $4.77 \log_{10}$ cfu/ml. Cakiroglu (3) found that 44.0 % of yoghurt samples examined contained an average count of 6.6×10^2 cfu/ml yeast and mould, while Sezgin (13) reported an average of 2.9×10^3 cfu/ml. Sireli and Ozdemir (14) reported the presence of yeast and mould in 14.0 % of 50 samples examined in their study. These results show that the contamination levels and total isolation rates of yoghurt samples are lower than our results obtained in the present study. However, Oz (11) analyzed 50 yoghurt samples and 84.0 % of those samples contained an average of 1.2×10^6 cfu/ml yeast and mould whereas 1.0×10^5 cfu/ml yeast and mould were counted in 55.0 % of 20 yoghurt samples by Duru and Ozgunes (5). Metin (9) isolated yeast-mould from 85 % samples in the range of 2.2×10^2 - 1.0×10^7 cfu/ml whereas Tayar et al. (15) reported the presence of yeast-mould in the 60 % of samples in the range of 4.2×10^3 - 8.4×10^4 cfu/ml. Minabelema and Stella (10) analyzed 100 yoghurt samples and isolated yeast-mould from 75.0 % in the range of 1.0×10^2 - 1.0×10^6 cfu/ml. These studies show that the level of yeast and mould in the 100 yoghurt samples examined in this study is lower than those examined by Oz (11), Duru and Ozgunes (5), Metin (9), Tayar et al. (15) and Minabelema and Stella (10). This may be due to the use of previous batch of yoghurt as a starter culture instead of ready made yoghurt starter and production or storage of products in an unsuitable environment which promotes growth / contamination of yeast and mould.

The average counts of *Enterobacteriaceae* spp. found in this study were 5.7×10^2 cfu/ml or $2.75 \log_{10}$ cfu/ml. Cakiroglu (3) reported 4.9×10^2 cfu/ml *Enterobacteriaceae* which is similar to the results of this study. In addition, 14.0 % of the samples were reported containing *Enterococci* by Sireli and Ozdemir (14). These results were lower than the results obtained in this study, which indicates better hygienic quality of product and the premises. *Enterococci* were $< 10^2$ cfu/ml in 82 samples and 10^2 cfu/ml in 8 samples with the average counts of 2.4×10^1 cfu/ml or $1.38 \log_{10}$ cfu/ml, whereas Cakiroglu (3) reported 4.9×10^2 cfu/ml *Enterococci* count in their study. Sireli and Ozdemir (14) however, found that out of 50 samples, 8.0 % contained *Enterococci*.

Micrococci and *Staphylococci* were found less than 10^2 cfu/ml in 88 and 10^2 cfu/ml in 12 samples. The average counts were 2.6×10^1 cfu/ml or $1.41 \log_{10}$ cfu/ml. Two coagulase positive *Staphylococci* were isolated from yoghurt samples and they were identified as *S. aureus*. Cakiroglu (3) found the average *Micrococci* and *Staphylococci* counts of 3.7×10^2 cfu/ml and isolated 3 coagulase positive *Staphylococci*. Sireli and Ozdemir (14) reported isolation of 10.0 % *Micrococci* and *Staphylococci* out of 50 samples but they were unable to isolate *S. aureus* in their samples.

In conclusion, the results of this study indicate that yoghurts produced and sold by family run dairies in Kars region require better hygienic conditions to fit into the established values based on TSE (TS 1330) in the microbiological criteria for yoghurt.

References

1. **Aboul-Khier, F.A., El-Bassiony, T., El-Rab, G.H.:** *Enterobacteriaceae* in some milk products in Sohag. J. Assut Vet. Med., 1985; 28: 79-83.
2. **Anonymous.:** Yoghurt (TS 1330). Turkish Standard Institute (TSE). Ankara. Turkey. 1989.
3. **Cakiroglu, S.H.:** Ankara garnizonundaki askeri birliklerde tüketilen yoğurtların kimyasal ve mikrobiyolojik kalitelerinin saptanması. Theises. 1997; Institute of Health Science, University of Ankara. Ankara.
4. **Dayisoylu, K.S.:** Van piyasasında üretien ve satışı sunulan yoğurtların fiziksel, kimyasal, mikrobiyolojik ve duyuşal özellikleri üzerine bir araştırma. Theises. 1992; Institute of Health Science, University of Van. Van.
5. **Duru, S., Ozgunes, H.:** Research on quality of hygienic marketing yoghurt and buttermilk samples in Ankara. Gıda Derg., 1981; 6 (4): 20-23.
6. **Ibrahim, M.K.E., El-Batavy, M.A., Girgis E.S.:** Evaluation of yoghurt on the Cairo market. Egyptian J. Dairy Sci., 1989; 17 (1): 125-136.
7. **Khalaf, S.H., Shareef, A.Y.:** The bacteriological quality of kishfa and yoghurt in Mosoul City. Food Microbiol., 1985; 2: 241-242.
8. **Mann, J.E.:** Yoghurt. Int. Dairy Industries. 1981; 46 (3): 28-33.
9. **Metin, F.:** Ankara'da imal edilen yoğurtların kalite sorunları üzerinde araştırma. Ankara Gıda Kontrol, Eğitim ve Araştırma Enstitüsü Müdürlüğü. 1979; 54: 1-37.
10. **Minabelema, D.G., Stella N.I.:** Yeasts as primary contaminants in yogurts produced commercially in Lagos, Nigeria. J. Food Prot., 1987; 50 (3): 193-198.
11. **Oz, K.:** Quality of consumed yoghurt in Konya. Theises. 1990; Institute of Health Science, University of Selcuk. Konya.
12. **Salih, A.W.M., Al-Abdullah, J.M., Hameed, A., Abdulkarim, N.:** Studies on the hygienic quality and nutritive status of market yoghurt in Baghdad. Dairy Science Abstract. 1984; 46 (11): 34-835.
13. **Sezgin, E.:** Ankara'da tüketilen yoğurtların yapımında kullanılan mayaların bazı teknik ve biyolojik nitelikleri üzerinde araştırmalar. J. Faculty of Agriculture of Ankara University. 1979; 722: 418.
14. **Sireli, T.U., Ozdemir, H.:** Microbiological quality of fruit yoghurts marketed in Ankara. J.Vet. Medical of Ankara University. 1998; 45 (2-3): 287-293.
15. **Tayar, M., Anar, T., Sen, C.:** Bursa'da tüketime sunulan yoğurtların kalitesi. Gıda Derg., 1993; 18 (3): 203-205.
16. **Yazici, F.:** Samsun ilinde tüketime sunulan yoğurtların duyuşal, fiziksel, kimyasal ve mikrobiyolojik nitelikleri üzerinde bir araştırma. Theises. 1991; Institute of Applied Science, University of 19 Mayıs. Samsun.