



RESEARCH ARTICLE

Microbiological quality of red meat pieces

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Öz

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Abstract

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Amaç: Bu araştırma Türkiye turizmi açısından önemli bir yeri olan Antalya'da faaliyet gösteren et parçalama üniteleri ve marketlerden temin edilen kıyma ve kırmızı et preparatlarında bazı mikrobiyolojik nitelikleri: Toplam mezofilik aerobik bakteri (TMAB), koliform bakteri, *E. coli* ve koagülaz (+) *Staphylococcus* sayısını belirlemek amacıyla yapılmıştır.

Gereç ve Yöntem: Araştırmada her bir et preparatından (Antrekot, kontrnuar, yumurta, kontrfile, tranç, sokum, gerdan, nuar, bonfile) 24 adet olmak üzere toplam 216 adet numune kullanıldı. Ayrıca her bir marketten (Satın alma esnasında parça etten kıyma haline getirilmek suretiyle) 60 adet kıyma numunesi temin edildi. Bütün numuneler TMAB, koliform bakteri, *E. coli* ve koagülaz (+) *Staphylococcus* varlığı yönünden incelenerek istatistiksel olarak değerlendirildi.

Bulgular: Et parçalama ünitesinden temin edilen kırmızı et preparatları kendi içlerinde karşılaştırıldığında, et preparatlarının sahip oldukları TMAB ve koliform bakteri sayıları arasında önemli farklılıklar gözlemlendi ($P<0.01$). Et parçalama ünitesinden elde edilen et preparatlarının *E. coli* ve koagülaz (+) *Staphylococcus* açısından önemli farklılıklara sahip olduğu görüldü ($P<0.01$). Marketlerden elde edilen et preparatları, TMAB ve koliform bakteri, *E. coli* ve koagülaz (+) *Staphylococcus* açısından önemli farklılıklar gösterdiği saptandı ($P<0.01$). Marketlerden temin edilen numunelerden yumurta, kontrfile, nuar ve bonfilede *E. coli*, yumurta, kontrfile, tranç ve bonfilelerde ise koagülaz (+) *Staphylococcus* üremesi tespit edilemedi.

Öneri: Et ve et ürünlerinin mikrobiyolojik kalitesinin iyileştirilmesi, üretimin bütün basamaklarında kontaminasyon kaynaklarının önlenmesi ve genel hijyen kurallarına uyulması ile sağlanabilir.

Anahtar kelimeler: Et, koliform bakterileri, *E. coli*, *S. aureus*

Aim: This research was conducted to determine the number of total mesophilic aerobic bacteria (TMAB), coliform bacteria, *E. coli* and coagulase (+) *Staphylococcus* in minced and red meat sold in meat shredding units and markets in Antalya city which has an important place in terms of tourism in Turkey.

Materials and Methods: Twenty-four samples were taken from each of the meat pieces (Ribeye, silverside, knuckle, striploin, topside, rump, neck, eyeround, tenderloin) giving a total of 216 samples used in this research. In addition 60 minced beef samples (The meat pieces were minced at the time of purchasing) were taken from different markets. All samples are examined and evaluated statistically from the point of the TMAB, coliform bacteria, *E. coli* and coagulase (+) *Staphylococcus*.

Results: Significant differences were observed in the TMAB and coliform bacteria number of meat preparations which were taken from the meat processing unit ($P<0.01$). Also significant differences were determined between the *E. coli* and coagulase (+) *Staphylococcus* counts of meat preparations ($p<0.01$). Statistical differences were determined in TMAB, coliform bacteria, *E. coli* and coagulase (+) *Staphylococcus* numbers of samples which were taken from markets ($P<0.01$). *E. coli*, growth was not determined in the knuckle, eyeround, silverside and tenderloin taken from the markets and coagulase (+) *Staphylococcus* growth was not determined in samples of knuckle, eyeround, silverside and tenderloin.

Conclusion: Improving the microbiological quality of meat and meat products provided with prevention of contamination sources in all steps of production and strict adherence to general hygiene rules.

Key words: Meat, coliform bacteria, *E. coli*, *S. aureus*





Introduction

Meat and meat products have an important role in food-borne infections and intoxications. While a part of microorganisms growing in meat and meat products can cause decaying without affecting human health; the other part cause infection and intoxication in humans without any deterioration in the meat and meat products. Especially *Staphylococcus aureus*, *Clostridium perfringens*, *Campylobacter jejuni*, *Escherichia coli* O157:H7 and *Salmonella* species are considered to be potential sources of risk in terms of meat and meat products. Microflora of meat pieces are similar to that found in the carcass. However, size of pieces, hand contact, shredding and conditions of sale significantly affect the final load. Çon and Gökalp (1998) reported that cubed meat stored at 10-15°C for 4-5 days occur bad odor, in 7th day there is a sticky and mucous layer on the surface. It is claimed that in hygienic slaughterhouses, for beef aerobic microorganism counts 10^3 - 10^5 cfu/cm², psychrotroph microorganisms 10^2 cfu/cm², coliform microorganisms 10 - 10^2 cfu/cm², for sheep meat aerobic microorganisms 10^3 - 10^6 cfu/cm², psychrotroph microorganisms 10^2 - 10^3 cfu/cm² are normal. Under hygienic conditions, pathogenic microorganisms in meat are low and microflora consists of saprophyte microorganisms.

This research was conducted to determine the number of TMAB, coliform bacteria, *E. coli* and coagulase (+) *Staphylococcus* in minced and red meat sold at meat shredding units and markets in Antalya city which has an important place in terms of tourism in Turkey.

Materials and Methods

The materials used in the study were obtained from a shredding unit and markets during a period of 12 months in Antalya. In the study, each meat sample was taken 24 pieces. Total 216 samples were used. In this research, separately from shredding units and markets; ribeye, silverside, knuckle, striploin, topside, rump, neck, eye round and tenderloin preparations were collected. Also 60 minced meat samples (the meat pieces were minced at the time of purchasing) provided from the markets were analyzed.

Samples were evaluated for TMAB, coliform bacteria, *E. coli* and coagulase (+) *Staphylococcus*. Samples were brought in to the laboratory under aseptic and cold chain than were analyzed. Required serial dilutions were prepared. For TMAB, Plate Count Agar (PCA-Merck 1.05463) 24 h at 35°C; for total coliform bacteria count, Violet Red Bile Agar (VRB

Table 1. Minimum, maximum, and average values of (\log_{10} cfu/g) TMAC and coliform bacteria of samples.

Microorganism	Meat preparations	N	Min	Max	X±Sx
TMAC	Ribeye	24	2.76	3.80	3.25±0.06 ^{bcd}
	Silverside	24	2.72	3.70	2.79±0.06 ^d
	Knuckle	24	2.73	3.84	3.19±0.05 ^{cd}
	Striploin	24	2.89	3.88	3.34±0.05 ^{bc}
	Topside	24	2.85	3.97	3.39±0.06 ^b
	Rump	24	2.91	4.79	3.58±0.08 ^a
	Neck	24	2.92	4.74	3.65±0.05 ^a
	Eye Round	24	2.88	3.79	3.36±0.05 ^{bc}
	Tenderloin	24	2.89	3.51	3.22±0.03 ^{bcd}
Coliform	Ribeye	24	2.48	3.48	2.79±0.06 ^{bc}
	Silverside	24	2.48	3.48	2.79±0.06 ^{bc}
	Knuckle	24	2.48	3.26	2.72±0.04 ^c
	Striploin	24	2.48	3.20	2.73±0.04 ^c
	Topside	24	2.48	3.51	2.84±0.05 ^{bc}
	Rump	24	2.49	3.53	2.96±0.06 ^b
	Neck	24	2.48	4.43	3.15±0.09 ^a
	Eye Round	24	2.51	4.66	3.22±0.10 ^a
	Tenderloin	24	2.48	3.53	2.80±0.06 ^{bc}

a, b, c, d: Different letters in the same row refers significant differences between the averages (P<0.001).



Table 2. Minimum, maximum and average values of *E. coli* and coagulase (+) *Staphylococcus* (log₁₀ cfu/g).

Microorganism	Meat Preparation	N	Min	Max	X±Sx
<i>E. coli</i>	Ribeye	24	<1	<1	<1
	Silverside	24	<1	<1	<1
	Knuckle	24	<1	<1	<1
	Striploin	24	1.76	2.76	2.47±0.06 ^b
	Topside	24	1.81	2.83	2.47±0.06 ^a
	Rump	24	1.85	2.83	2.67±0.08 ^a
	Neck	24	<1	<1	<1
	Eye Round	24	<1	<1	<1
	Tenderloin	24	<1	<1	<1
Coagulase (+) <i>Staphylococcus</i>	Ribeye	24	2.48	3.08	2.52±0.09 ^b
	Silverside	24	2.48	3.08	2.52±0.09 ^b
	Knuckle	24	<1	<1	<1
	Striploin	24	<1	<1	<1
	Topside	24	<1	<1	<1
	Rump	24	2.50	3.58	2.83±0.05 ^a
	Neck	24	2.51	2.80	2.73±0.04 ^a
	Eye Round	24	2.48	2.86	2.78±0.05 ^a
	Tenderloin	24	<1	<1	<1

a, b: Different letters in the same row refers significant differences between the averages (P<0.001). <1: Growth was not observed.

Merck 1.01406) double layered, 18-24 hat 35°C; for *E. coli*, Violet Red Bile + MUG (Fluorocult VRB Merck 1.04030) medium was used and incubated 18 hours at 37°C. After incubation the colonies were calculated and evaluated. For coagulase (+) *Staphylococcus* determination, Baird Parker Agar (BPA Merck 1.05406) with egg yolk tellurite emulsion (EYT Merck 1.03785) was poured and incubated at 35°C for 24-48 h. After 48 h, 1-1.5 mm narrow diameter, black, shiny, convex colonies and also 3 mm wide-scale boundary zone surrounded by a clear ring-shaped colonies were seen. Coagulase test (Oxoid DR0595 Staphylas Test) was administered to determined appropriate colonies.

The obtained data was evaluated by analysis of variance in the Statistical Analysis System, a computer program. Differences between groups were revealed by Duncan multiple comparison test. P<0.05 level was accepted statistically significant.

Results

The results of microbiological analysis of samples are shown with minimum, maximum, mean and standard error values in Tables 1, 2, 3 and 4. Evaluations of data for the examined

microorganisms of samples are shown in Table 5 and comparisons of the seasons are shown in Table 6.

Discussion

Microbiological quality characteristics of meat samples obtained from the meat shredding unit and the supermarkets were tried to be determined. All samples were examined in terms of TMAB, coliform bacteria, *E. coli* and coagulase (+) *Staphylococcus* and were statistically evaluated.

The number of TMAB of the meat preparations obtained from the meat shredding units were determined between 2.79-3.65 log₁₀ cfu/g; the same number in the preparations obtained from the supermarkets was determined between 3.49-4.18 log₁₀ cfu/g (Tables 1 and 3). These observed values are lower than the values determined for raw red meat in Turkish Food Codex (Anonymous 2011). In a research done by Çalıcıoğlu et al (2005) on beef carcass surface contamination in Elaziğ; the count of TMAB collected from 48 samples were found as 3.70-4.90 log₁₀ cfu/cm², and they were ≥4.0 log₁₀ cfu/cm² in 45.4% of all samples. In the microbiological studies conducted on beef carcasses, Ingram and Roberts (1976) and Cook et al (1997) determined the number of





TMAB as \log_{10} 4.48 cfu/cm² and \log_{10} 2.68-7 cfu/cm² respectively. The researchers in a study in which beef carcasses in different categories were studied (in terms of carcass microflora) determined the number of TMAB as \log_{10} 3.04-3.40 cfu/cm² (McEvoy et al 2000). Considering the number of TMAB, the collected values are within the scope of the first class meat classification done by some researchers (Little and De Louvois 1998). It is especially pleasing to determine this situation in Antalya which has an important role in Turkish tourism.

When red meat preparates obtained from the meat shredding unit and supermarkets were compared, it was observed that there were meaningful differences among the numbers of TMAB ($P < 0.01$) (Tables 1 and 3). It was determined that silverside had the lowest values in preparations obtained from the meat shredding unit; tenderloin had the lowest value in preparations obtained from the supermarkets.

The differences determined in red meat preparations are likely to arise from positions of meat preparations on carcass, the manipulations carried out, ambient air, transportation, cooling and the processes that workers applied on meat preparations. Thus, some researchers (Çon and Gökalp 1998)

reported that the flora determined in the pieces of meat is similar to the flora in the carcass, but size of the piece, hand contact, shredding and conditions of marketing considerably affect the final microbial load.

The number of coliform bacteria in red meat samples obtained from the meat shredding unit was between 2.72-3.22 \log_{10} cfu/g, the same number in meat preparations obtained from the supermarkets was between 2.80-3.71 \log_{10} cfu/g (Table 1 and 3). Gill et al (2000) determined 3.33 \log_{10} cfu/cm²; Emswiler et al (1976) determined 1.70 \log_{10} cfu/g of coliform bacteria in carcass. Little and De Louvois (1998) determined the coliform bacteria in ready-to-eat meat between 10²-10⁴ cfu/g.

Eisel et al (1997) found a high number of coliform bacteria in packed beef. The reasons including the widespread presence of coliform bacteria in nature, their ability to grow outside of human and animal body and having some fecal strains can be accepted to be the indicators of sanitation. In other words, it is crucial because it reflects inadequate hygienic conditions during transportation, storing and processing. When it is evaluated in this respect, limited number or absence of coliform bacteria in food products is considered positive in

Table 3. Minimum, maximum and average values of TMAC and coliform bacteria (\log_{10} cfu/g) of samples.

Microorganism	Meat preparation	N	Min	Max	X±Sx
TMAC	Ribeye	24	3.08	5.42	4.10±0.10 ^a
	Silverside	24	3.23	5.98	4.08±0.14 ^a
	Knuckle	24	3.15	4.98	3.74±0.08 ^{bc}
	Striploin	24	3.26	5.91	3.96±0.12 ^{ab}
	Topside	24	2.98	5.85	4.15±0.12 ^a
	Rump	24	3.20	5.58	4.18±0.10 ^a
	Neck	24	3.00	5.76	4.15±0.12 ^a
	Eye Round	24	3.26	4.88	3.86±0.09 ^{ab}
	Tenderloin	24	2.75	4.30	3.49±0.07 ^c
Coliform	Ribeye	24	2.80	4.51	3.53±0.08 ^{ab}
	Silverside	24	2.59	5.26	3.47±0.13 ^{ab}
	Knuckle	24	2.64	4.56	3.31±0.08 ^{bc}
	Striploin	24	2.54	4.51	3.37±0.10 ^{abc}
	Topside	24	2.58	5.54	3.68±0.13 ^a
	Rump	24	2.58	5.42	3.71±0.13 ^a
	Neck	24	2.52	4.66	3.62±0.12 ^{ab}
	Eye Round	24	2.49	4.62	3.39±0.12 ^{abc}
	Tenderloin	24	2.48	3.53	2.80±0.06 ^{bc}

a, b, c: Different letters in the same row refers significant differences between the averages ($P < 0.001$) <1: Growth was not observed.



Table 4. Minimum, maximum and average values of *E. coli* and coagulase (+) *Staphylococcus* (\log_{10} cfu/g).

Microorganism	Meat preparation	N	Min	Max	X±Sx
<i>E. coli</i>	Ribeye	24	1.78	3.83	2.40±0.21 ^a
	Silverside	24	1.79	3.92	2.60±0.14 ^a
	Knuckle	24	<1	<1	<1
	Striploin	24	<1	<1	<1
	Topside	24	1.90	3.75	1.76±0.27 ^b
	Rump	24	1.78	3.56	1.77±0.26 ^b
	Neck	24	1.20	3.02	1.86±0.27 ^b
	Eye Round	24	<1	<1	<1
	Tenderloin	24	<1	<1	<1
Coagulase (+) <i>Staphylococcus</i>	Ribeye	24	2.52	3.90	2.87±0.23 ^{ab}
	Silverside	24	2.48	3.92	2.60±0.24 ^{ab}
	Knuckle	24	<1	<1	<1
	Striploin	24	<1	<1	<1
	Topside	24	2.49	3.91	2.64±0.25 ^{ab}
	Rump	24	2.56	4.11	2.85±0.25 ^{ab}
	Neck	24	2.52	3.94	3.17±0.14 ^a
	Eye Round	24	2.49	3.96	2.62±0.25 ^{ab}
	Tenderloin	24	<1	<1	<1

a, b, c: Different letters in the same row refers significant differences between the averages ($P < 0.001$) <1: Growth was not observed.

Table 5. Microflora of minced meat samples (\log_{10} cfu/g).

Microorganism	N	Min	Max	X±Sx
TMAC	60	3.23	7.59	5.22±0.13
Coliform	60	2.58	6.88	4.51±0.11
<i>E. coli</i>	60	2.30	4.86	2.61±0.17
Coa (+) <i>Staphylococcus</i>	60	2.80	4.79	3.42±0.13

terms of hygiene of the enterprise and reliability of foodstuff.

The numbers of *E. coli* in meat preparations obtained from the meat shredding unit and the supermarkets showed significant differences ($P < 0.01$, Tables 2 and 4). The number of *E. coli* in meat samples obtained from the meat shredding unit was determined between 2.47-2.67 \log_{10} cfu/g (Table 2); and the number of *E. coli* in meat samples obtained from the supermarkets was between 1.76-2.60 \log_{10} cfu/g (Table 4). It was observed that this microorganism was under detectable level in ribeye, silverside, knuckle, neck, eye round, tenderloin obtained from the meat shredding unit, and it was under detectable level in the samples which were obtained from the supermarket including knuckle, striploin, eye round and tenderloin (Tables 2 and 4).

Little and De Louvois (1998) determined 10^2 - 10^5 cfu/g of *E. coli* in carcasses. The researchers stated that *E. coli* must not exceed 10^4 cfu/g. It is indicated in Turkish Food Codex (Anonymous 2011) that in raw meat, *E. coli* O157 must not be present in 25 g. Emswiller et al (1976) determined the *E. coli* in beef carcass as 1.24 \log_{10} cfu/g. Gill and McGinnis (1999) established the presence of *E. coli* in carcass at the rate of 3.46 cfu/cm². Gill and Jones (1999) reported that they did not find *E. coli* in carcass but filet had *E. coli* at the rate of 2.14 cfu/100 cm². Bell (1997) determined that the rate of *E. coli* in carcasses obtained from cows whose skins contain fecal contamination goes over the rate of 2 \log_{10} , and the same rate in carcasses obtained from cows having clean skin is <2 \log_{10} . As it has fecal origin, the presence of *E. coli* in food indicates that there is direct or indirect fecal contamination in food.

Significant differences were observed in the number of coagulase positive *Staphylococcus* in the meat preparations obtained from the meat shredding unit and the supermarkets ($P < 0.01$) (Tables 2 and 4). The number of coagulase positive *Staphylococcus* in the meat samples obtained from the meat shredding unit was found between 2.52-2.83 (Table 2), the same number was found between 2.60-3.17 \log_{10} cfu/g





Table 6. Changes in microflora of samples depending on the season.

Microorganism	Season	N	Min	Max	X±Sx
TMAC	Spring	15	3.93	6.41	5.24±0.19 ^b
	Summer	15	4.54	7.59	6.31±0.23 ^a
	Fall	15	4.08	6.66	4.91±0.20 ^b
	Winter	15	3.23	4.79	4.31±0.21 ^c
Coliform	Spring	15	2.58	5.45	4.21±0.17 ^b
	Summer	15	4.38	6.88	5.44±0.19 ^a
	Fall	15	3.75	5.91	4.42±0.15 ^b
	Winter	15	2.86	4.79	3.96±0.11 ^b
<i>E. coli</i>	Spring	15	1.70	3.45	2.66±0.28 ^{ab}
	Summer	15	2.11	4.86	3.54±0.29 ^a
	Fall	15	1.62	3.88	2.35±0.32 ^b
	Winter	15	1.54	3.20	1.90±0.36 ^b
Coagulase (+) <i>Staphylococcus</i>	Spring	15	3.15	4.26	3.73±0.09 ^{ab}
	Summer	15	3.26	4.79	4.09±0.11 ^a
	Fall	15	2.62	3.89	3.37±0.09 ^b
	Winter	15	2.36	3.20	2.48±0.40 ^c

(Table 3) for the samples obtained from the supermarkets. Phillips et al (2001a, b) found coagulase positive *Staphylococcus* in 24.3% of beef carcasses, in 27.5% of beef, in 24.1% mutton carcasses and in 38.6% of mutton. Desmarchler et al (1999) encountered coagulase positive *Staphylococcus* in beef carcasses which were obtained from 3 different slaughterhouses. They were determined at the rate of 62% in the enterprise-A, at the rate of 85% in the enterprise-B and at the rate of 89% in the enterprise-C. They reported that brisket and flank regions were higher in terms of coagulase positive *Staphylococcus* contamination rate than the rump region after internal organs were taken out. They discovered that hands of workers in one of the slaughterhouses had high rates of coagulase positive *Staphylococcus*. In the study conducted on meat cutting boards in meat processing enterprises, Little and DeLouvois (1998) found high levels of *E. coli*, coliform and *S. aureus*. The researchers stated that *S. aureus* must not exceed 10⁴ cfu/g. Vanderline et al (1998) reported that beef carcasses have coagulase positive *Staphylococcus*. Emswiller et al (1976) discovered the average presence of *S. aureus* in raw meat and ready-to-eat raw meat products at the rate of 0.74 log₁₀ cfu/g. The researchers stated that the changes observed in the number of *S. aureus* during storage are not significant. The collected data are appropriate for Turkish Food Codex Anonymous (2011) and it is obser-

ved that they do not exceed 10⁴ kob/g as reported by some researchers (Little and DeLouvois 1998). Also, the undetectable level of coagulase positive *Staphylococcus* in some meat preparations is the indicator of production in hygienic conditions. The numbers of TMAB, coliform bacteria, *E. coli* and coagulase (+) *Staphylococcus* of the minced meat samples obtained from the supermarkets were determined as 5.22, 4.51, 2.61 and 3.42 log₁₀ cfu/g, respectively (Table 5). Sancak et al (1993) determined the total number of TMAB and the number of coagulase (+) *Staphylococcus* of the minced meat exposed for sale in the province of Van as 2.3x10⁵-1.4x10¹⁰ cfu/g and 0-9.2x10⁶ cfu/g, respectively. The researchers also established the number of coliform and *E. coli* as 4.0x10⁶ and 4.1x10⁵ cfu/g, respectively. Gönülalan and Köse (2003) established the TMAB as 7.4x10⁵-5.3x10⁹; the number of coliform as 8.6x10¹-4.5x10⁸; the number of *E. coli* as 1.0x10¹-5.2x10⁵; the number of coagulase (+) *Staphylococcus* as 1.0x10¹-6.7x10⁶ cfu/g in the minced meat exposed for sale in Kayseri. Başkaya et al (2004) determined the TMAB as 3.1x10⁴-6.3x10⁷; the number of coliform as 3.3x10³-6.2x10⁴; the number of *E. coli* as 1.0x10⁴-1.4x10⁴; the number of coagulase (+) *Staphylococcus* as 8.0x10²-8.2x10³.

Microbial criteria related to the minced meat have been established in Turkey (Anonymous 2011). Accordingly, in two of minced meat samples, obtained from meat selling area, the total number of aerobe mesophilic bacteria can be present up to 5x10⁶ cfu/g and the remaining three samples must not exceed 5x10⁵ cfu/g (Tables 5 and 6). It is seen that the values established in this study are lower than the values determined by other researchers (Sancak et al 1993, Gönülalan and Köse 2003, Başkaya et al 2004) and they are in accordance with the values established by Turkish Food Codex. While the number of coagulase (+) *Staphylococcus* determined in the research is lower than the values reported by many researchers, it is close to the limit values established by Turkish Food Codex. When the climatic flora of the minced meat samples were compared, there were meaningful differences among the groups (P<0.01, Table 5). It is seen that the numbers of all examined microorganisms in the minced meat samples in summer are higher in general. In this regard, it is necessary to be more careful during shredding, transporting, packing and other processes in summer and to take better care of good hygiene applications and controlled production.

Conclusions

Consequently, it was observed that the microbial flora of red meat preparations and minced meat provided for consumption in Antalya has good quality. In this respect, it is concluded that the activity of Hazard Analysis and Critical Control Points (HACCP) system, which finds application area all around the world, must be made more active and applicable in food business.



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