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Microbiological Properties of Boneless Sheep Meat in Kahramanmaraş

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Abstract: This study was undertaken to determine the microbiological properties of boneless sheep meat sold by supermarkets, local markets and butchers in Kahramanmaraş, Turkey, and also to examine the microbiological quality of the food surfaces and tools used. Of the meat samples, 93 were collected from supermarkets (group 1), 25 from local markets (group 2) and 21 from butchers (group 3). Food surface and tool samples were collected from 139 cutting tables, 139 chopping boards, 139 workbenches, 255 knives, 139 balances and 278 aprons. The mean values of the aerobic plate count (log CFU/g) of the boneless sheep meat samples from the 3 groups were 5.92, 5.70 and 5.83, respectively. Presumptive *Escherichia coli* (% detected) was 1.07 (1/93), 0.25 (1/25) and 0.21 (1/21) in the 3 groups. The mean values of pseudomonads were (log CFU/g) 4.79, 4.83 and 4.59, mean values of yeasts and moulds were 3.36, 3.59 and 4.38, and mean values of psychrotrophs were 4.61, 4.55 and 4.77, respectively. Coagulase positive *Staphylococcus aureus* was detected (20%) in 5 meat samples from local markets. Coagulase positive *S. aureus* was not detected in meat samples from supermarkets and butchers.

The mean values of aerobic bacteria from the cutting tables, chopping boards, workbenches, knives, balances and aprons were (log CFU/ml) 6.67, 6.47, 7.62, 6.76, 7.0 and 6.88, respectively. The average counts of yeasts and moulds from the food surfaces and tools were: cutting tables 6.27, chopping boards 5.0, workbenches 5.51, knives 5.56, balances 6.32 and aprons 6.36 log CFU/ml, respectively. *E. coli* was not detected from the food surfaces or tools. Coagulase positive *S. aureus* was detected on knives (1.9%) and aprons (3.4%).

Key Words: Microbiological properties, boneless sheep meat, tools.

Kahramanmaraş İli'nde Satılan Kemiksiz Koyun Etinin Mikrobiyolojik Özellikleri

Özet: Bu çalışma Kahramanmaraş İli'ndeki süper market, yerel market ve kasaplardaki kemiksiz koyun etinin mikrobiyolojik durumu ile bu yerlerde kullanılan alet ile etin temas edebileceği yüzeylerin mikrobiyolojik özelliklerini belirlemek amacı ile yapılmıştır. Et örneklerinden 93'ü süper marketten, 25'i yerel marketten, 21 tanesi ise kasaplardan alınmıştır. Ayrıca 139 tahta, 139 tezgah, 139 çalışma masası, 255 bıçak, 139 tartı, 278 adet işçi önlüğünün mikrobiyolojik özelliğine de bakılmıştır. Alınan örneklerdeki ortalama aerobik bakteri sayısı sırası ile 5,82, 5,70, 5,83 log kob/g iken, şüpheli *Escherichia coli* % 1,07 (1/93), 0,25 (1/25) ve 0,21 (1/21) olarak tespit edilmiştir. Ortalama pseudomonads sayısı 4,79, 4,83, 4,59 log kob/g iken ortalama maya ve küf 3,36, 3,59, 4,38 log kob/g, olup ortalama psikotrof bakteri sayısı sırası ile 4,61, 4,55, 4,77 log kob/g olarak gözlenmiştir. Kasaplardan alınan 5 et örneğinde koagülaz pozitif *Staphylococcus aureus* tespit edilmiştir.

Tahta, tezgah, çalışma masası, bıçak, tartı ve önlüklerden elde edilen ortalama aerobik bakteri sayısı sırası ile 6,67, 6,47, 7,62, 6,76, 7,0 ve 6,88 log kob/ml olup, ortalama maya ve küf sayısı sıra ile 6,27, 5,0, 5,51, 5,56, 6,32 ve 6,36 log kob/ml'dir. Çalışma yüzeyleri ve aletlerde *E. coli* tespit edilemezken, koagülaz pozitif *S. aureus* bıçak (% 1,9) ve önlükte (% 3,4) gözlenmiştir.

Anahtar Sözcükler: Mikrobiyolojik özellik, kemiksiz koyun eti, alet.

Introduction

Worldwide interest in meat hygiene has increased in recent years. The focus on the hygienic quality of sheep meats is increasing with respect to regulatory bodies considering meat a major cause of food poisoning. When

animals are presented for slaughter the populations of aerobic microorganisms on their hides and hooves, and within their intestinal tracts is high and variable. No absolute guarantee can ever be given that the alimentary tracts of healthy animals will be free from microorganisms pathogenic to humans (1).

Meat is a nutrient-rich substrate that can support the growth of a wide range of microorganisms (2). Bacterial contamination of fresh meat has important implications for food safety and product shelf-life.

Psychrotrophs, pseudomonads, *Staphylococcus aureus*, and yeasts and moulds have been used in meat and poultry products to assess their microbiological safety, sanitation conditions throughout processing, and keeping quality (3). Pseudomonad counts are a practical indicator of hygiene (4). They also help to predict the shelf-life of food (5). Food poisoning caused by *Staphylococcus* is one of the major causes of foodborne illness throughout the world (6). The term 'indicator microorganisms' is vague and can be associated with a wide variety of applications. Indicator microorganisms are generally used to examine the existence of faecal contamination, the quality of a product, and the efficiency of a food production process (7).

An estimation of the numbers of bacteria in the environment may be necessary to test the standards of hygiene and efficiency of cleaning procedures in supermarkets, local markets, and butchers, to observe routes of contamination or to educate staff (8).

The aim of the present study was to monitor the microbiological quality of boneless sheep meat sold by supermarkets, local markets and butchers and, parallel to this, the microbiological quality of the food surfaces and tools used in the retail process.

Materials and Methods

Sampling of Boneless Sheep Meats

From January, 2001, to June, 2002, 139 samples of fresh boneless sheep meats were collected monthly from supermarkets, local markets and butchers in Kahramanmaraş, Turkey. A total of 139 samples representing the microbiological quality of fresh boneless sheep meat was collected from 93 supermarkets (group 1), 25 local markets (group 2) and 21 butchers (group 3) throughout Kahramanmaraş. All the samples were packed in insulated containers with chiller packs. They were placed in a refrigerator until analysed. The samples were analysed daily.

Microbiological Methods

The samples were analysed for the presence of aerobic microorganisms, psychrotrophs, pseudomonads, presumptive *Escherichia coli*, *S. aureus* (coagulase positive *Staphylococci*), and yeasts and moulds. The microbiological methods applied are shown in Table 1 (9-11). Confirmation tests for the identification of bacteria were as follows: coliforms were tested for gas production on brilliant green bile lactose broth (Merck, Darmstadt, Germany) by incubation at 37 ± 1.0 °C for 24 ± 3 h. The tests for the production of indole from tryptophan were performed by adding to the test medium 0.3 to 0.5 ml of Kovac's indole reagent. Fluorescent pseudomonads on Pseudomonas Agar containing CFC (cetrimide, fucidin and cephaloridine) supplement were enumerated under 254-

Table 1. Microbiological methods.

Microbes	Medium	Incubation conditions			
		Temp (°C)	Time	Atmosphere	Reference
Aerobic bacteria	Plate Count Agar	30.0	48-72 h	Aerobic	(9)
Coliforms	Violet Red Bile Agar	30.0	48 h	Aerobic	(9)
<i>Escherichia coli</i>	Violet Red Bile Agar Endo Agar	37	24-48 h	Aerobic	(9,10)
		37	24 h	Aerobic	
Pseudomonads	Pseudomonas Agar with CFC Supplement	25	48 h	Aerobic	(10)
Yeasts and Moulds	Sabaroud Dextrose Agar	25	5 days	Aerobic	(10)
Psychrotrophs	Plate Count Agar	7	10 days	Aerobic	(9,10)
<i>Staphylococcus aureus</i>	Baird Parker Agar with egg yolk emulsion supplement	37	24-48 h	Aerobic	(11)

nm UV light (12). *S. aureus* was confirmed using cell morphology, the arrangement of the cells, Gram reaction (13), catalase activity (11), the oxydase test and coagulase activity (11).

The results were expressed as log CFU/g.

Sampling of Food Surfaces and Tools

To test the standard of hygiene and efficiency of cleaning procedures in supermarkets, local markets and butchers, the swab technique was used. Sterile cotton swabs were rubbed over an area within a 5 x 5 cm square on the food surfaces. One swab was used for a predetermined area of a cutting table, chopping board, workbench, balance or workers' apron and for both sides of knives. The rubbed swabs were placed into tubes and 10 ml of 0.1% peptone water was added. The tubes were shaken and left to stand for 20-30 min. The enumeration of aerobic plate count (APC), yeasts and moulds, coliforms and presumptive *E. coli* and coagulase positive *S. aureus* is described above (8). The results were expressed as log CFU/ml.

Statistical Analysis

The differences among the groups (group 1, supermarkets; group 2, local markets; group 3, butchers) were compared by one-way ANOVA. In order to compare the correlation between 2 groups the paired samples t-test was applied. All data were analysed using SPSS (SPSS Production Facility Release 7.5, SPSS Inc., 1995).

Results

Microbiological properties of boneless sheep meat samples

For the boneless sheep meat samples from supermarkets, local markets and butchers the mean values of aerobic counts were 5.92, 5.70, and 5.83 log CFU/g, respectively. There was no significant difference among the aerobic counts from samples obtained from the 3 different sources. The results are shown in Table 2.

Presumptive *E. coli* was detected in 1 sample each of the 3 sources. There was no significant difference in the distribution of the numbers of presumptive *E. coli* from the samples obtained from supermarkets, local markets and butchers.

The mean values of pseudomonads for the samples from the 3 groups were 4.79, 4.83, 4.59 log CFU/g, respectively.

The mean values of yeasts and moulds were 3.36, 3.59 and 4.38 log CFU/g from supermarkets, local markets and butchers, respectively.

The average counts of psychrotrophs from supermarkets, local markets and butchers were 4.62, 4.55, and 4.77 log CFU/g, respectively. The psychrotrophs were significantly correlated ($P < 0.001$) with pseudomonads. This suggested that *Pseudomonas* spp. comprise a major proportion of psychrotrophic microorganisms in most cases.

Table 2. The mean values of bacterial counts found in 139 boneless sheep meat samples.

	Samples from supermarkets Group 1 (n: 93)	Samples from local markets Group 2 (n: 25)	Samples from butchers Group 3 (n: 21)
Aerobic bacteria ^a	5.92	5.70	5.83
Presumptive <i>E. coli</i> ^b	1.07 (1/93)	0.25 (1/25)	0.21 (1/21)
Pseudomonads ^a	4.79	4.83	4.59
Yeasts and Moulds ^a	3.36	3.59	4.38
Psychrotrophs ^a	4.61	4.55	4.77
Coagulase positive <i>S. aureus</i> ^b	nd ^c	20	nd ^c

^a : (Mean values log CFU/g)

^b : percentage

^c : Not detected

In boneless sheep meat samples coagulase positive *S. aureus* was detected in 7 samples from 25 local markets (20%). All of the 7 samples were from the same local market.

The bacterial counts of food surfaces and tools

The results of the mean values of aerobic bacteria, yeasts and moulds and presumptive *E. coli* and *S. aureus* (coagulase positive) from cutting tables, chopping boards, workbenches, knives, balances and workers' aprons from supermarkets, local markets, and butchers are shown in Table 3. The mean values of aerobic bacteria from cutting tables, chopping boards, workbenches, knives, balances and aprons were 6.67, 6.47, 7.62, 6.76, 7.0 and 6.88 log CFU/ml, respectively. The mean values of yeast and moulds were as follows: cutting tables 6.27 log CFU/ml; chopping boards, 5.0 log CFU/ml; workbenches, 5.51 log CFU/ml; knives, 5.56 log CFU/ml; balances, 6.32 log CFU/ml; and aprons, 6.36 log CFU/ml. *E. coli* was not detected on the food surfaces or tools. Coagulase positive *S. aureus* was detected on knives (1.9%) and aprons (3.4%).

Discussion

During deboning there is a dilution of bacteria on the meat surface as a result of contact with freshly exposed sterile tissue and care should be taken to ensure that cutting boards are regularly cleaned during the process (14,15). There were significant differences in APCs in

boneless meat samples within the 3 groups ($P < 0.001$). The higher APCs (group 1; 5.92 log CFU/g; group 2; 5.70 log CFU/g; and group 3; 5.83 log CFU/g) suggested that an unusual amount of contamination and/or growth of natural flora occurs during deboning. The findings of Vanderlinde et al. (14) were similar to those of the present study. High numbers of bacteria could be transmitted from the fleece of sheep to the carcass surface during hide removal (16). The areas of highest contamination were those sites where cuts were made through the skin (17).

In 5 samples from local markets (n: 25) coagulase positive *S. aureus* was detected. However, coagulase positive *S. aureus* was not determined from the supermarkets or butchers. Regarding the tools, out of 255 knife samples 5 were determined to contain coagulase positive *S. aureus*. *S. aureus* is usually transmitted through humans, as reported by Heinzel (18) and De Wit and Kampelmacher (19). The existence of staphylococci on food surfaces and tools suggested contamination, either from animals or from humans, of the meat. Hansson (20) found that coagulase positive *S. aureus* was detected in 9% of beef carcasses from high-capacity slaughter houses and in 16% of beef samples from low-capacity slaughterhouses.

Presumptive *E. coli* was found in 34% of the samples from beef carcasses. This higher level could be explained by the evisceration technique used. This technique is probably the main reason for the higher level of aerobic

Table 3. The mean values of bacterial counts found from cutting tables, chopping boards, workbenches, knives, balances and aprons.

Control plan	Aerobic bacteria ^a	Yeasts and moulds ^a	presumptive <i>E. coli</i> ^b	<i>S. aureus</i> ^{b,c}
Cutting tables (n: 139)	6.67	4.27	nd ^d	nd
Chopping Boards (n: 139)	6.47	4.0	nd	nd
Workbenches (n: 139)	7.62	4.51	nd	nd
Knives (n: 255)	6.76	5.56	nd	1.9
Balances (n: 139)	7	4.32	nd	nd
Aprons (n: 278)	6.88	4.36	nd	3.4

^a : (mean values log CFU/ml)

^b : (percentage)

^c : Coagulase positive *Staphylococcus aureus*

^d : not detected

bacteria (20). To avoid contamination during hide removal, it is essential to keep hides from the dehided surfaces. The great majority of the mesophilic or psychrotrophic bacteria on cattle carcasses after slaughtering and dressing derive from the hide (21). Moist and unclean hides from cattle may be the source of strong contamination of the dehided carcasses (22).

E. coli was detected in one each of supermarkets, local markets and butchers. *E. coli* was found in 1 sample from the 21 butchers. *E. coli* is an indicator of faecal contamination. It is likely that the observed incidence of faecal bacteria is due to problems associated with removal of the fleece and its coming into contact with the surface of the carcass. The low counts of *E. coli* in the 3 groups of boneless sheep meat suggest that temperatures during transportation and holding were adequate to limit the growth of the bacteria. *E. coli* was not detected from the samples of food surfaces and tools. In a previous study, 7 out of 45 samples were found to contain *E. coli* (23).

The counts of yeasts and moulds of supermarkets, local markets and butchers were 3.36 3.59 and 3.38 log CFU/g, respectively. There were no significant differences

within the supermarkets and local markets in terms of yeast and moulds ($P > 0.05$). There was a significant difference within groups 1-3 and 2-3 in terms of yeast and moulds ($P < 0.001$). The values displayed a similarity to the illustrative values of Nebraska (24).

The counts of pseudomonads and psychrotrophs were significantly correlated ($P < 0.001$). In cooler climates psychrotrophic bacteria constitute a higher percentage of hide flora than they do in warmer climates (25). The higher proportion of psychrotrophic bacteria found in the present study indicates that surface drying and/or the temperature were not adequate to control their growth.

In conclusion, from the beginning of the 1990s, HACCP was mandated as a prerequisite for the international meat trade (26). Food safety systems based on HACCP principles have to be applied in food processing plants, retail food stores and food service operations. Education of food handlers and information for consumers about microbial risks associated with the consumption of meat and how to control them are needed.

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