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ÖZGÜN ARAŞTIRMA/ORIGINAL ARTICLE

Et Örneklerinden İzole Edilen Staphylococcus aureus'un Enterotoksin Üretimi ve Antibiyotik Direnç Profili

Enterotoxin Production and Antibiotic Resistance Profile of Staphylococcus aureus Isolated from Meat Samples

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Anahtar Sözcükler:
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ÖZ

Amaç: Stafilokokal gıda zehirlenmesi, esas olarak enterotoksijenik Staphylococcus aureus tarafından üretilen stafilokokal enterotoksinlerle kontamine olmuş gıdanın yenmesi ile ortaya çıkan önemli bir hastalıktır. Bu çalışmanın amacı, Giresun ve Trabzon illerinde kasaplarda satılan çiğ et örneklerinde Staphylococcus aureus kontaminasyonu, izolatların enterotoksin kapasiteleri ve çeşitli antibiyotiklere direnç profilleri bakımından taşıdığı halk sağlığı risklerini değerlendirmektir.

Gereç ve Yöntem: Toplam 68 çiğ et örneği, mikrokok/stafilokok ve toplam stafilokok varlığı yönünden analiz edildi. İzolatların tanımlanmaları ve antibiyotik duyarlılık testleri, VITEK 2 cihazı ile saptanmıştır. Enzim bağlı immünolojik test tekniği kullanılarak stafilokok izolatlarındaki klasik tip enterotoksin varlıkları belirlenmiş ve tiplendirilmiştir.

Bulgular: Mikrokok/stafilokok ve toplam stafilokok mikrobiyal yükleri sırasıyla 10^1-10^5 kob/g ve 10^1-10^5 kob/g arasında tespit edildi. Çalışılan 68 et örneğinden, toplam 171 adet stafilokok izole edildi. Elde edilen izolatlar içerisinde, çiğ tavuk etinde bir adet S. aureus (%1,03) izolatı bulundu. Bu izolatın enterotoksin üretme ve E tipi enterotoksin üretme kabiliyetine sahip olduğu bulundu. S. aureus izolatı, test edilen diğer antibiyotiklere duyarlı iken sadece benzilpenisiline dirençli olarak bulundu.

Sonuç: Çalışmanın sonuçları, antibiyotiğe dirençli ve enterotoksijenik S. aureus 'un tüketiciler için önemli bir hijyenik risk oluşturduğunu göstermiştir. Stafilokokal gıda zehirlenmesi vakaları açısından risk oluşturması ve zamanla halk sağlığını tehdit etmesi nedeniyle bu konuda kapsamlı çalışmalar yapılmalıdır.

Key Words:
Antibiotic resistance
Enterotoxin
Meat
Staphylococcus aureus

ABSTRACT

Objective: Staphylococcal food poisoning is a major human disease that is causes by the ingestion of food contaminated by staphylococcal enterotoxins produced mainly by enterotoxigenic Staphylococcus aureus. The aim of this study was to evaluate the public health risks of the raw meat samples marketed in Giresun and Trabzon provinces regarding the contamination of S. aureus, enterotoxin capacity of the isolates and their resistance to various antibiotics.

Material and Method: A total of 68 meat samples were analyzed for micrococci/staphylococci and total staphylococci. The identification and antibiotic susceptibility of isolates were determined with the VITEK 2 device. Classical type enterotoxin production of staphylococcal isolates were determined and typed using with Enzyme-Linked Immuno Sorbent Assay technique.

Results: The microbial loads of micrococci/staphylococci were 10^1 to 10^5 cfu/g and total staphylococci 10^1 to 10^5 cfu/g, respectively in 68 meat samples. A total of 171 staphylococci were isolated from the 68 meat samples. Among the isolates obtained, one isolate (1.03%) detected in the raw chicken meat was S. aureus. This isolate was found to have the ability of producing enterotoxin and to produce E type enterotoxin and was resistant only to benzylpenicillin, while it was sensitive to other tested antibiotics.

Conclusion: The results of this study indicated that antibiotic-resistant and enterotoxigenic S. aureus poses a significant hygienic risk for consumers. Due to the posing a risk for staphylococcal food poisoning cases and threatening public health over time, extensive studies should be conducted on this subject.

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Introduction

Staphylococci are gram-positive bacteria characterized by the formation of grape-like clusters. So far more than 60 species have been identified in the staphylococci group with 32 species colonizing the human body (1). Staphylococci are usually differentiated to two groups using the coagulase test. Even if cause asymptomatic colonization in some people, it is usually recognized that coagulase positive staphylococci (CoPS) are pathogenic, coagulase negative staphylococci (CoNS) are saprophytic or lead to opportunistic infections (2).

Staphylococcus aureus with the ability to produce enterotoxin are also considered important for food-borne poisoning (3). Staphylococcal enterotoxins (SE) are enterotoxins that are members of the pyrogenic toxin superantigen family in terms of biological activities and structural relationships (4,5). So far, based on the antigenic properties of enterotoxins, a total of 23 SEs have been reported, including seven classical serologic types as well as new type SEs and staphylococcal enterotoxin-like toxins (SEl) (6).

Staphylococcal food poisoning (SFP), is a phenomenon in which staphylococci enterotoxins transmit their potential release to the environment after reaching a level of 10^5-10^6 colony forming unit (cfu)/gram (g) and causes symptoms such as, nausea, vomiting, severe abdominal cramps, abdominal pain, diarrhea, headache, dizziness, general weakness, pulse, weakness, shallow breathing, shock and enteritis (7,8). Foodborne outbreaks caused by S.aureus are common worldwide regardless of the country's level of development. However, this is more frequently encountered in the developing and underdeveloped areas. In addition, common species and enterotoxins vary among the geographic regions (9).

It has been proposed that people may be colonized by S.aureus from raw meat products without contact with livestock and the organism is a mediator that can transmit to the house environment. S.aureus has been isolated from raw meat and chicken meat sold in butchers and it has been reported to have potential to colonize humans if hygienic rules are not followed (10-12).

Given the diversity among antimicrobial agents, in the early 1970s physicians abandoned the belief that all bacterial infections are treatable. This has been caused by the increased multiple antibiotic resistance of pathogens such as S.aureus (13). S. aureus is a major pathogen with increasing importance due to the increase in its antimicrobial resistance. The intensive and unconscious use of antibiotics in agricultural production, animal production and human health has led many bacterial species to become resistant to antibiotics. Over the past decade, it has been observed that the multiple resistance properties of pathogenic bacteria, especially isolated from foods, have increased in antibiotics. Increasing resistance of bacteria to antibiotics and the rapid spread of these resistant bacteria among species are reported to be the main threat to human health all over the world (14-16).

The objectives of this study were to isolate and identify S. aureus from raw bovine meat and chicken meat samples using phenotypic methods, to investigate for the presence of five types classical staphylococcal enterotoxin and resistance to 17 different antibiotics.

Material and Method

Sample Collection

A total of 68 food samples were collected from the butchers in Giresun and Trabzon provinces between 2012 and 2013. These samples consisted of 30 raw bovine meat products (liver, minced, kidney, meatball, meat cubes and heart) and 38 raw chicken meat products (buttock, liver, chest, wing, meatball, gizzard). The samples were collected using sterile polyethylene containers and transferred to the laboratory for immediate process. Ethics committee approval is not required for food samples used in this study.

Isolation and Identification of Staphylococci

All collected samples were homogenized in a blender. 25 g was collected from each sample and put into the sterile bags. 225 mL of buffered peptone water (bioMérieux. Marcy l'Etoile, France) was added on the sample that will be used in the isolation and the mixture was homogenized in the blender (Waring, New Hartford, Conn.).

Homogenized food samples were diluted from 10^{-1} to 10^{-3} and 0.1 mL of two-parallel decimal dilutions was transferred to the Baird Parker Agar supplemented with rabbit plasma (BP+RPF; bioMérieux. Marcy l'Etoile, France) and Mannitol Salt Phenol-Red Agar (MSA, bioMérieux. Marcy l'Etoile, France) media for staphylococci isolation and counting. Spreading on the surface was then carried out with Drigalski loop and the mixture was left to incubation at 37°C for 24-48 hours. At the end of the incubation, suspicious black, convex colonies that formed or did not form a surrounding zone Bair Parker Agar and those fermented and did not ferment mannitol in the mannitol salt agar were selected for pure culturing (17,18).

After incubation of subculture, the colonies on the plates were counted as colony forming units per gram.

Stock cultures were then performed for the identification of the selected colonies. The colonies were identified on the basis of gram staining, catalase, oxidase and clumping factor test, rabbit-plasma-tube coagulase test (Bactident Coagulase; Merck, Madrid, Spain) and bacitracin resistance as coagulase-positive staphylococci (CPS) and coagulase-negative staphylococci (CNS). Then, Staphylococci isolates were confirmed with using by VITEK® 2 Gram-Positive (GP) Identification Card (BioMérieux. Marcy l'Etoile, France) in VİTEK 2 device (19).

Detection and Typing of Staphylococcal Enterotoxin

The presence of five type (A-E) classical staphylococcal enterotoxins (SEs) were detected with Ridascreen® SET A, B, C, D and E Kit (R-Biopharm AG, Darmstadt Germany, Art. No. R4101). Staphylococci were cultered in tubes containing 10 ml Brain Heart Infusion Broth (Oxoid, CM0225) and incubated at 37 °C for 18-24 hour. The culture suspensions were then centrifuged at 15 °C and 3500 rpm for 5 minutes, filtered through a sterile filter with a pore diameter of 0.2 µm and filtrates were obtained. 100 µL of the filtrates of each isolate was put into the wells A-G of the microtiter plate so that each strip used for one isolate and 100 µL of positive control plate to the well H. The plate was manually mixed by carefully shaking and left to incubation at 35-37°C for one hour. Then the liquid was dumped out of the wells into a sink. The conjugate was added 100 µl to each well and incubated for 1 h at 35-37 °C. The conjugate 2 was added 100 µl to each well and incubated for 30 min at 35-37 °C. The subsrate/chromogen was added 100 µl to each well and incubated for 15 min at 35-37 °C. After the conjugate, conjugate 2 and subsrate/chromogen steps, the wells were filled with 300 µl wash buffer per well and washing step was repeated 4 times. After adding 100 µL of stop solution on each well, measurements were made with ELISA (ELx800) device at 450-630±10 nm absorbance within 30 minutes. Interpretation of the results was made based on the cut-off value. Accordingly, the samples with an OD ≥ cut-off value were considered as positive, and those with an OD<cut-off value as negative (20,21).

Determination of Antibiotic Resistance

Preparations and application procedures of the isolates were made according to the operating manual of Vitek analyzer (VITEK Microbiology Reference Manual, bioMerieux, Inc. Box 15969 Durham, North Carolina 27704-0969, USA). The isolates were inoculated in 5% sheep blood agar for antibiogram tests of staphylococci. Three-four fresh colonies of the same genus were selected and put into a tube containing 3 mL sterile physiologic water (0.45%-0.50% NaCl, pH 4.5-7.0). Concentration of bacteria suspension in the tube was set to 0.5-0.63 McFarland standart and pipetted to a second tube containing 280 µL physiologic water. The prepared suspension tubes were inserted into the cassette that was put into the device and VITEK 2 AST P592 Gram positive antibiotic sensitivity cartridge was inserted to the cassette as to correspond the tubes. Barcodes of the kits in the cassette were read by the device from analysis entry screen. The results were read from the monitor of the device after the necessary incubation period was completed.

VITEK 2 antibiogram sensitivity kit P592 (bioMerieux Ref 22287) used in the study includes benzylpenicillin (BEN), fusidic acid (FA), gentamicin (CN), oxacillin (OX), imipenem (IPM), ciprofloxacin (CIP), clindamycin (DA), linezolid (LNZ), moxifloxacin (MXF), teicoplanin (TEC), vancomycin (VA), tetracycline (TE), rifampicin (RA), tigecycline (TIG), erythromycin (E), fosfomycin (FF) and trimethoprim/sulfamethoxazole (SXT) antibiotics. S.aureus ATCC-29213 was used as the reference strain (19).

Statistical Analysis

Descriptive statistical parameters and percentage were conducted using SPSS v22 (IBM Corp., Armonk, NY, USA) statistical software. Count data obtained were expressed as the mean ± standard deviation (SD).

Results

In the study, 68 raw meat sample presented for consumption in the butchers within Giresun and Trabzon province were analyzed. Distribution of the samples collected from the provinces is listed in Table 1.

Table 1. Distribution of the meat samples.

Sample	Sample type	Number of samples	Total (n)
Raw bovine meat products	kidney	5	30
	liver	5	
	minced	5	
	meatball	5	
	meat cubes	5	
	heart	5	
Raw chicken meat products	buttock	4	38
	liver	15	
	breast	5	
	wing	4	
	meatball	5	
	gizzard	5	

Thirty raw bovine meat sample and 38 raw chicken meat sample were examined for micrococci/staphylococci and total staphylococci. A mean of 4.90 log cfu/g micrococci/staphylococci and 4.86 log cfu/g total staphylococci were counted in the bovine meat samples. Whereas a mean of 4.91 log cfu/g micrococci/staphylococci and 4.85 log cfu/g total staphylococci were counted in the chicken meat samples. The mean and standard deviation of count results is given in Table 2.

Table 2. Microorganism counts of meat samples

	Micrococci/Staphylococci			Total Staphylococci		
Sample group	Min.	Max.	Mean±SD	Min.	Max.	Mean±SD
Raw bovine meat products	1.17	5.39	4.90±1.07	1.14	5.39	4.86±1.41
Raw chicken meat products	1.17	5.30	4.91±1.38	1.17	5.34	4.85±1.71
Mean	1.17	5.34	4.88±1.23	1.15	5.36	4.89±1.56

The species identity of 171 staphylococcus strains isolated from 68 meat samples was determined. A total of 75 coagulase-negative staphylococci (19 S.saprophyticus, 17 S.vitulinus, 9 S.warneri, 7 S.equorum, 6 S.xylosus, 5 S.sciuri, 4 S.hominis spp. hominis, 3 S.lentus, 2 S.epidermidis, 2 S.klossii and 1 S.hominis spp. novobiosepticus) were isolated from the raw meat samples. Of the 95 coagulase-negative (24 S.vitulinus, 18 S.equorum, 18 S.saprophyticus, 16 S.warneri, 10 S.sciuri, 3 S.xylosus, 3 S.haemolyticus, 1 S.simulans, 1 S.lentus, 1 S.cohnii spp. urealyticus) and 1 coagulase-positive (S. aureus) staphylococci were isolated from the studied raw chicken samples (Table 3).

Table 3. Distribution of *Staphylococcus* strains in bovine and chicken meat samples.

	Raw bovine meat samples		Raw chicken meat samples	
Staphylococcus strains	n	%	n	%
S. aureus	-	-	1	1
S. cohnii spp. Urealyticus			1	1
S. epidermidis	2	2 6	-	-
S. equorum	7	15 5	18	18 7
S. haemolyticus	-	-	3	3 1
S. hominis spp. Hominis	4	5 3	-	-
S. hominis spp. novobiosepticus	1	1 3	-	-
S. klossii	2	2 6	-	-
S. lentus	3	4	1	1
S. saprophyticus	19	25 3	18	18 7
S. sciuri	5	6 6	10	10 4
S. simulans	-	-	1	1
S. vitulinus	17	22 6	24	25
S. warneri	9	12	16	16 6
S. xylosus	6	8	3	3 1
Total	75	100	96	100

n= no of sample, %= sample rate

One isolate (1.03%) isolated from the raw chicken meat was found to be S.aureus, which belongs to the coagulase positive staphylococcus group. According to enterotoxin analyses; S. aureus isolate was found to have the ability of producing enterotoxin and to produce E type enterotoxin (SEE). The optic dansity and ELISA image of enterotoxigenic S. aureus is presented in Figure 1 and Figure 2.

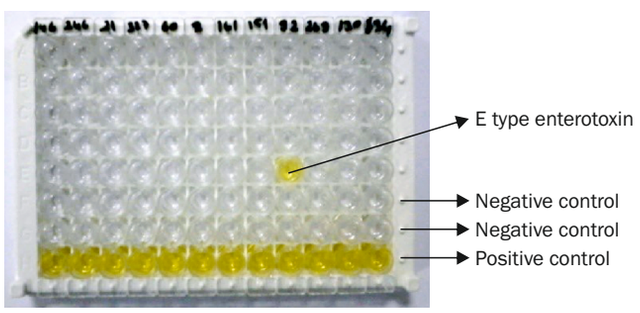

Figure 1. Image of S. aureus E type enterotoxin.

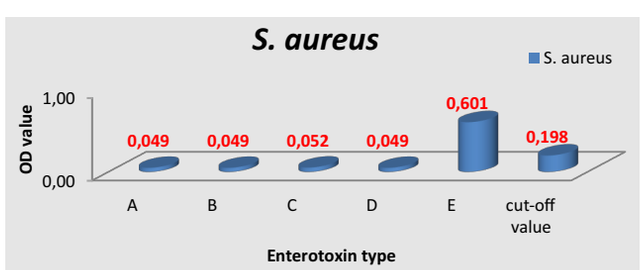

Figure 2. Optic density value of enterotoxigenic S. aureus.

S.aureus isolate was resistant to benzylpenicillin, while it was sensitive to oxacillin, imipenem, gentamicin, ciprofloxacin, moxifloxacin, erythromycin, clindamycin, linezolid, teicoplanin, vancomycin, tetracycline, tigecycline, fosfomycin, fusidic acid, rifampicin, and trimethoprim/sulfamethoxazole antibiotics.

Discussion

Staphylococcal enterotoxins produced during bacterial growth in foods play a role in staphylococcal food poisoning. S.aureus is the most enterotoxigenic strain among staphylococci. S.aureus is a pathogenic bacterium that has been intensively studied worldwide due to diseases caused by the microorganism. The presence of S.aureus has been well-established in commercial products (2,12,13,22).

A mean of micrococci/staphylococci and total staphylococci were counted 4.90, 4.86 log cfu/g in the bovine meat samples and 4.91, 4.85 log cfu/g in the chicken meat samples, respectively. The detection of micrococci/staphylococci counts in this study was lower than the 6.21

log cfu/g reported by Tascı et al. (23). Similar micrococci/ staphylococci and total staphylococci count results were also 5.14 log cfu/g and 3.7-6.15 log cfu/g reported by Can ve Çelik (24), Martins et al. (25) respectively.

There are numerous studies in the literature reporting foodborne outbreaks originated from S.aureus. However, very variable results have been reported by these studies regarding the occurrence of enterotoxigenic S.aureus in foods. In 2014, 393 foodborne outbreaks caused by staphylococcal toxins have been reported (26). In a study by Pu et al. (27); the prevalence of enterotoxigenic S.aureus in retail meats was reported as 85% in Louisiana, USA. In another study by Argudin et al. (28), Park et al. (29), Savariraj et al. (30) enterotoxigenic S.aureus was isolated from 31 (48.4%) of 64 isolates in Spain, 104 (74.8%) of 139 isolates in Korea and 20 (66.67%) of 80 isolates in India respectively. In a study by Avsaroglu et al. (31) from Turkey, enterotoxigenic S.aureus was detected in 6 (5%) of 23 isolates. In another studies from Turkey, enterotoxin contamination was reported as 3.02% by Ertas et al. (32) in Kayseri 2010, 60.1% by Guven et al. (33) 2010 in Kutahya and Eskisehir provinces, 25% by Can and Celik (24) in Ankara and 13.7% by Gucukoglu et al. (34) in Samsun. Similar results of low prevalence of staphylococci was also determined by Pereira et al. (35) in whose study, out of 148 staphylococci, nine S. aureus isolate produced entero-toxin from raw meat samples. In Slovakia, Medvedova et al. (36) observed that 2 of 28 S. aureus isolates were posi-tive for genes that encode one or more enterotoxins. The variation of the contamination level may be due to poor hygiene practiced during the slaughter and processing of foods in different countries.

Foodborne infection and intoxication cases arise mainly from raw milk and dairy products, but also from meat and meat products. In addition to the presence of pathogenic microorganisms in foodstuffs, their active numbers are also among the determining factors in terms of food poisoning (37). Significant cases of staphylococcal food poisoning occurred as a result of cooking or heat treatment of food following the formation of enterotoxins in food (4). According to enterotoxin analyses in this study; S. aureus isolate was found to produce E type enterotoxin (SEE) isolated from chicken liver (Figure 1). In similar lines, Wang et al. (38) have analyzed 23 methicillin-resistance S. aureus isolates from retail foods and found positive isolates for see (staphylococcal enterotoxin gene A) (8.7%), followed by seb (staphylococcal enterotoxin gene B) (52.2%), sec (staphylococcal enterotoxin gene C) (4.3%), sed (staphylococcal enterotoxin gene D) (43.5%), seg (staphylococcal enterotoxin gene G) (56.5%), sei (staphylococcal enterotoxin gene I) (4.3%). Li et al. (39) have found pvl (Panton-Valentine leukocidin gene) to be the most frequent enterotoxin encoding gene (26.6%), followed by sej (staphylococcal enterotoxin gene J) (12.5%), sea (9%), seh (staphylococcal enterotoxin gene H) (8.3%), seb (6.9%), sec (6.9%), sed (4.8%), sei (3.1%) and see (staphylococcal enterotoxin gene E) (2.4%) in 133 S. aureus strains isolated from retail raw chicken meat. Ali et al. (40) have noted that 48.6% of the examined S. aureus isolates recovered from raw chicken meat and giblets harbored 21 isolates see enterotoxin gene types. In contrast, in India the most frequent SE genes of S. aureus isolates from retail chicken meat were seb (80.95%) through seg (66.67%), sei (66.67%), sec (14.29%), sed (9.53%) and sej (9.53%) (30). Differences between the studies are resulted from the differences in number of the samples and food groups. In addition, the rate of contamination shows a great diversity according to geographic areas, season when the samples were collected and echologic origin of the species.

Determination of S.aureus variants resistant to antibiotics has been especially underlined in the literature (10,11). Particularly, resistance of S.aureus to methicillin and clindamycin has been widely studied (41,42). Studies in the literature have mostly focused on some antibiotics to which S.aureus showed the highest resistance (43,44). Unlike these studies, in this study the resistance of S.aureus against 17 different antibiotics was investigated. In this study, S.aureus was found to be resistant only to benzylpenicillin and it was sensitive against OX, IPM, CN, CIP, MXF, E, DA, LNZ, TEC, VA, TE, TIG, FF, FA, RA, and SXT antibiotics. The same high penicillin resistance rate of S. aureus among meat samples has also been reported in Turkey and other countries including Bangladesh, USA, China, Egypt (27, 38, 45, 46, 47). On the other hand, other researchers have found resistance levels to TE (67%), E (30%), DA (18%), OX with 2% NaCl (14%), CIP (13%), CN (3%), and MXF (1%) (27), to TE (30%), E (20%), CIP (12.5%) in Turkey (45), to CIP (21.43%), E (21.43%), OX (28.57%) and SXT (14.28%) (46), to OX (92.3%), TE (66.7%), CIP (25.6%), E (59.0%), SXT (5.1%), DA (25.6%) and VA (7.7%) in methicillin-sensitive S. aureus in Bangladesh (47). Different results among the studies might be resulted from the source of the S. aureus isolates and the frequency and type of antimicrobial agents prescribed for treating Staphylococcal infections, e.g. in food-producing animals in different geographical areas.

Conclusion

According to the results of this study, we demonstrated that the presence of enterotoxigenic and antimicrobial-resistant S.aureus poses a risk for food poisoning cases and seriously threatens public health. This emphasizes the importance of hygienic measures in the consumption chain for food safety. Comprehensive studies on this issue should be continuously conducted because of the increasing antibiotic resistance of S.aureus over time.

Yazarlık Katkısı: Fikir/Hipotez: TC, NG. Tasarım: TC, NG. Veri toplama/Veri işleme: TC. Veri analizi: TC, NG. Makalenin hazırlanması: TC, NG.

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Kaynaklar

1. Khorshed A, Ozbal Y. Identification of coagulase negative stephylococci isolated from blood cultures and determination and investigation of antibiotic susceptibility. Journal of Health Sciences 2012;21:153-163.
2. Gonzalez-Martına M, Corberaa JA, Suarez-Bonnet A, Tejedor-Juncoa MT. Virulence factors in coagulase-positive staphylococci of veterinary interest other than Staphylococcus aureus. Veterinary Quarterly 2020;40:118-131.
3. Erol, İ. Gıda Hijyeni ve Mikrobiyolojisi. 1. Baskı. Ankara: Pozitif Matbaacılık, 2007;135-144.
4. Bergdoll MS. Enterotoxins: Staphylococci and Staphylococcal Infections (SEA-E). New York: Academic Press, 1983:559-598.
5. Omoe K, Ishıkawa M, Shımoda Y, Hu DL, Ueda S, Shınagawa K. Detection of seg, seh, and sei genes in Staphylococcus aureus isolates and determination of the enterotoxin productivities of S. aureus isolates harboring seg, seh, or sei genes. Journal of Clinical Microbiology 2002;40: 857-862.
6. Liu D. Enterotoxin-Producing Staphylococcus aureus. In: Tang YW, Sussman M, Liu D, Poxton I, Schwartzman J editors. Molecular Medical Microbiology. Second Edition, USA: Elsevier, 2015:979-995.
7. Tang J, Tang C, Chen J et al. Phenotypic characterization and prevalence of enterotoxin genes in Staphylococcus aureus isolates from outbreaks of ilness in Chengdu City. Foodborne Pathogens and Disease 2011;8:1317-1320.
8. Yılmaz, S. Kayseri Bölgesinde Tüketime Sunulan Çiğ Sütlerde Staphylococcus aureus ve Enterotoksin Varlığının Araştırılması. Yüksek Lisans Tezi, Erciyes Üniversitesi Sağlık Bilimleri Enstitüsü, Kayseri, 2009;1-35.
9. Hennekinne JA, De Buyser ML, Dragacci S. Staphylococcus aureus and its food poisoning toxins: characterization and outbreak investigation. FEMS Microbiol Rev 2012;36:815- 836.
10. De Boer E, Zwartkruis-Nahuis JTM, Wit B, Huijsdens XW. Prevalence of methicillin-resistant Staphylococcus aureus in meat. International journal of food microbiology 2009;134: 52-56.
11. Hanson BM, Dressler AE, Harper AL et al. Prevalence of Staphylococcus aureus and methicillin-resistant Staphylococcus aureus (MRSA) on retail meat in Iowa. Journal of Infection and Public Health 2011;4:169-174.
12. Waters AE, Content-Cuomo T, Buchhagen J et al. Multidrug-resistant Staphylococcus aureus in US meat and poultry. Clinical Infectious Diseases 2011;52:1227-1230.
13. Swartz MN. Use of antimicrobial agents and drug resistance. N. Engl. J. Med 1997;337:491–492.
14. Pesavento G, Ducci B, Comodo N et al. Antimicrobial resistance profile of Staphylococcus aureus isolated from raw meat: A research for methicillin resistant Staphylococcus aureus (MRSA). Food Control 2007;18:196-200.
15. Normanno G, La Salandra G, Dambrosio A et al. Occurence, characterization and antimicrobial resistance of enterotoxigenic Staphylococcus aureus isolated from meat and dairy products. International Journal of Food Microbiology 2007: 115: 290-296.
16. Morandi S, Brasca M, Andrighetto C, Lombardi A, Lodi R. Phenotypic and genotypic characterization of Staphylococcus aureus strains from Italian dairy products. International Journal of Microbiology 2009;501362: 1-7.
17. Baird RM, Lee WH. Media used in the detection and enumeration of Staphylococcus aureus. Journal of Food Protection 1995;26:15-24.
18. Korpysa-Dzirba W, Osek J. Detection of classical genes and enterotoxins of Staphylococcus aureus isolated from raw milk in the south-east region of Poland. Bulletin of the Veterinary Institute in Pulawy 2014; 58:559-561.
19. Nonhoff C, Rottiers S, Struelens MJ. Evaluation of the Vitek 2 system for identification and antimicrobial susceptibility testing of Staphylococcus spp. Clinical Microbiology and Infection 2005;11:150-153.

20. Kılıç S, Küplülü Ö. Detection the enterotoxin producing capacity of coagulase positive Staphylococcus by EIA (Enzyme Immuno Assay) isolated from turkey meat. Ankara University Faculty of Veterinary Medicine Journal 2009;56:183-186.

21. Bingöl EB, Çetin Ö, Çolak H, Hampikyan H. Presence of enterotoxin and verotoxin in Turkish cheeses sold in İstanbul. Turkish Journal of Veterinary Animal Science 2012;36:424-432.

22. Kelman A, Soong YA, Dupuy N et al. Antimicrobial susceptibility of Staphylococcus aureus from retail ground meats. Journal of food protection 2011;74:1625-1629.

23. Taşçı F, Şahindokuyucu F, Öztürk D. Detection of Staphylococcus species and staphylococcal enterotoxins by ELISA in ice cream and cheese consumed in Burdur Province. African Journal of Agricultural Research 2011;6:937-942.

24. Can HY, Çelik TH. Detection of enterotoxigenic and antimicrobial resistant S. aureus in Turkish cheeses. Food Control 2012;24:100-103.

25. Martins I M, Kabuki DY, Miya NTN, Pereira JL. Occurrence and Characterization of Enterotoxigenic Potential of Staphylococcus Isolated from Dairy Products. Journal of Food Safety 2014; 34:185-192.

26. European Food Safety Authority (EFSA). The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2014 and 2015;13: 4329.

27. Pu S, Wang F, Ge B. Characterization of toxin genes and antimicrobial susceptibility of Staphylococcus aureus isolates from Louisiana retail meats. Foodborne Pathog Dis 2011;8: 299306.

28. Argudín MA, Mendoza MC, González-Hevia MA, Bances M, Guerra B, Rodicioa MR. Genotypes, Exotoxin Gene Content, and Antimicrobial Resistance of Staphylococcus aureus Strains Recovered from Foods and Food Handlers. Applied and Environ-mental Microbiology 2012;78:2930-2935.

29. Park KM, Oh SK, Cha JO, Lee YS, Koo M. Characterization of Antibiotic Resistant Staphylococcus aureus Isolates from Ready-to-Eat Foods in Korea. Journal of the Korean Society for Applied Biological Chemistry 2014;57:387-395.

30. Savariraj WR, Ravindran NB, Kannan P, Rao, VA. Occurrence and enterotoxin gene profiles of Staphylococcus aureus isolated from retail chicken meat. Food Science and Technology International 2020;1-7.

31. Avsaroglu MD. Prevalence of Staphylococcus aureus Isolated From Various Foods of Animal Origin in Kırşehir, Turkey and Their Enterotoxigenicity. Turkish Journal of Agriculture - Food Science and Technology 2016;4:1179-1184.

32. Ertas N, Gonulalan Z, Yildirim Y, Kum E. Detection of Staphylococcus aureus enterotoxins in sheep cheese and dairy desserts by multiplex PCR technique. Int J Food Microbiol 2010;142:74-77.

33. Guven K, Mutlu MB, Gulbandilar A, Cakir P. Occurrence and characterization of Staphylococcus aureus isolated from meat and dairy products consumed in Turkey. J Food Safety 2010;30:196212.

34. Gücükoğlu A, Kevenk TO, Uyanik T, Çadirci Ö, Terzi G, Alişarli M. Detection of enterotoxigenic Staphylococcus aureus in raw milk and dairy products by multiplex PCR. J Food Sci 2012; 77:620-623.

35. Pereira V, Lopes C, Castro A, Silva J, Gibbs P, Teixeira P. Characterization for enterotoxin production, virulence factors, and antibiotic susceptibility of Staphylococcus aureus isolates from various foods in Portugal. Food Microbiology 2009;26: 278-282.

36. Medveďová A, Studeničová A, Valík L, Horváthová Z. Prevalence and Growth Dynamics of Enterotoxinogenic Staphylococcus aureus Isolates in Slovakian Dairy Products. Czech Journal of Food Science, 2014;32:337-341.

37. Uğur M, Nazlı B, Bostan K. Gıda Hijyeni. İstanbul: Teknik Yayınevi, 2001;57-58.

38. Wang X, Li G, Xia X, Yang B, Xi M, Meng J. Antimicrobial Susceptibility and Molecular Typing of Methicillin-Resistant Staphylococcus aureus in Retail Foods in Shaanxi, China. Foodborne Pathogens and Disease 2014;11:281-286.

39. Li S, Wang P, Zhao J et al. Characterization of toxin genes and antimicrobial susceptibility of Staphylococcus aureus from retail raw chicken meat Journal of Food Protection, 2018;8: 528-533.

40. Ali SS, Moawad SMS, Hussein MA et al. Efficacy of metal oxide nanoparticles as novel antimicrobial agents against multidrug and multi-virulent Staphylococcus aureus isolates from retail raw chicken meat and giblets. International journal of food microbiology 2021;344:109116.

41. Karamatsu ML, Thorp AW, Brown L. Changes in community-associated methicillin-resistant Staphylococcus aureus skin and soft tissue infections presenting to the pediatric emergency department: comparing 2003 to 2008. Pediatr Emerg Care 2012;28:131-135.

42. Vicetti Miguel CP, Mejias A, Leber A, Sanchez PJ. A Decade of Antimicrobial Resistance in Staphylococcus Aureus: A Single Center Experience. PLoS One 2019:12;e0212029.

43. Foster TJ. Antibiotic resistance in Staphylococcus aureus. Current status and future prospects. FEMS Microbiology Reviews 2017;41:430-449.

44. Yunlei G, Guanghui S, Meiling S, Juan W, Yi W. Prevalence and Therapies of Antibiotic-Resistance in Staphylococcus aureus. Front Cell Infect Microbiol 2020;10:107.

45. Can HY, Elmalı M, Karagöz A. Molecular typing and antimicrobial susceptibility of Staphylococcus aureus strains isolated from raw milk, cheese, minced meat, and chicken meat samples. Korean Journal for Food Science of Animal Resources 2017;37:175-180.

46. Darwish WS, Atia AS, Reda LM, Elhelaly AE, Thompson LA, Eldin WFS. Chicken giblets and wastewater samples as possible sources of methicillin-resistant Staphylococcus aureus Prevalence, enterotoxin production, and antibiotic susceptibility. J Food Safety 2018;38:e12478.

47. Parvin S, Ali Y, Talukder S et al. Prevalence and Multidrug Resistance Pattern of Methicillin Resistant S. aureus Isolated from Frozen Chicken Meat in Bangladesh. Microorganisms 2021; 9:636.