Kafkas Universitesi Veterinar Fakultesi Dergisi
ISSN: 1300-6045 e-ISSN: 1309-2251
Journal Home-Page: http://vetdergikafkas.org

Kafkas Univ Vet Fak Derg
27 (3): 323-329, 2021
DOI: 10.9775/kvfd.2021.25370

RESEARCH ARTICLE

Detection of Methicillin Resistant *Staphylococcus aureus* Strains and Typing of Staphylococcal Cassette Chromosome *mec* from Various Foods Originated Different Region from Turkey [1]

Ghassan ISSA [1,a (*)] Ali AYDIN [2,b]

[1] *This study was supported from Technological Research Council of Turkey (TÜBİTAK-2216)*

[1] Medical Laboratory Techniques Program, Avrupa Vocational School, Kocaeli Health and Technology University, TR-34020 Zeytinburnu, İstanbul - TURKEY
[2] Department of Food Hygiene and Technology, Faculty of Veterinary Medicine, İstanbul University-Cerrahpaşa, TR-34320 Avcılar, İstanbul - TURKEY
ORCIDs: [a] 0000-0002-0229-7632; [b] 0000-0002-4931-9843

Article ID: KVFD-2021-25370 Received: 03.01.2021 Accepted: 08.04.2021 Published Online: 09.04.2021

Abstract

Staphylococcus aureus is a microorganism that is highly resistant to environmental conditions and is widely found in environmental sources. It can cause a large number of infections in both humans and animals. Resistance to methicillin in *S. aureus* strains occurs due to the production of low affinity penicillin binding proteins (PBP2a). PBP2a is encoded by the *mec*A gene. The *mec*A gene is located on a mobile, large genetic element called the staphylococcal cassette chromosome mec (SCC*mec*). Until now, 13 SCC*mec* (I-XIII) types have been identified in *S. aureus* strains. In this study, in different 7 regions (Marmara, Aegean, Central Anatolia, the Black Sea, the Mediterranean, Eastern and Southeastern Anatolia) of Turkey obtained from a variety of points of 700 food items in the [(dairy products (n:560), bakery products (n:89), ready meal (n:40), meat product (n:11)], after the isolation of cultural *S. aureus* and verification by PCR, MRSA detection and SCC*mec* typing were aimed. 67 (9.57%) *S. aureus* strains were isolated from 700 food samples analyzed within the scope of the study. Only 1 (0.14%) of the 67 *S. aureus* strains isolated, both phenotypically and genotypically, was found to be MRSA and when SCC*mec* was typed, it was found to be Type IV. Community-acquired MRSA strains can cause clinical cases ranging from skin infections to fatal pneumonia and sepsis, as well as foodborne diseases. As a result, it is considered that MRSA strains can be an important source of contamination for humans with the consumption of food of animal origin.

Keywords: *Staphylococcus aureus, Antibiotic Resistance, mecA, SCCmec, Food, Turkey*

Türkiye'nin Farklı Bölgeleri Kaynaklı Toplanan Çeşitli Gıda Maddelerinden Metisilin Dirençli *Staphylococcus aureus* Suşlarının Tespiti ve Stafilokokkal Kaset Kromozom *mec* Tiplendirilmesi

Öz

Staphylococcus aureus hem insanlarda, hem de hayvanlarda çok sayıda enfeksiyona neden olabilen, ortam şartlarına oldukça dayanıklı ve çevresel kaynaklarda yaygın olarak bulunan bir mikroorganizmadır. *S. aureus* suşlarında metisiline direnç, düşük afiniteli penisilin bağlayan proteinlerin (PBP2a) üretimine bağlı olarak meydana gelmektedir. PBP2a, *mec*A geni tarafından kodlanmaktadır. *mec*A geni ise stafilokokkal kaset kromozom mec (SCC*mec*) adı verilen hareketli, büyük genetik eleman üzerinde bulunmaktadır. *S. aureus* suşlarında şu ana kadar 13 SCC*mec* (I-XIII) tipi tespit edilmiştir. Bu çalışmada, Türkiye'nin 7 farklı bölgesindeki (Marmara, Ege, İç Anadolu, Karadeniz, Akdeniz, Doğu Anadolu ve Güneydoğu Anadolu) çeşitli noktalardan temin edilen 700 adet gıda maddesinde [(süt ürünü (n:560), unlu mamuller (n:89), hazır yemek (n:40), et ürünü (n:11)] kültürel *S. aureus* izolasyonu ve PCR ile doğrulaması yapıldıktan sonra MRSA tespiti ve SCC*mec* tiplendirilmesi amaçlanmıştır. Araştırma kapsamında analiz edilen 700 adet gıda numunesinden 67 (%9.57) adet *S. aureus* suşu izole edilmiştir. İzole edilen 67 *S. aureus* suşundan hem fenotipik, hem de genotipik olarak sadece 1 (%0.14) adedinin MRSA suşu olduğu saptanmış ve SCC*mec* tiplendirilmesi yapıldığında, Tip IV olduğu tespit edilmiştir. Toplumsal kökenli MRSA suşları, gıda kaynaklı hastalıkların yanı sıra deri enfeksiyonlarından ölümcül pnömoni ve sepsise varan klinik tablolara da neden olabilmektedir. Sonuç olarak hayvansal kökenli gıdaların tüketimi ile MRSA suşlarının insanlar için önemli bir bulaşma kaynağı olabileceği değerlendirilmektedir.

Anahtar sözcükler: *Staphylococcus aureus, Antibiyotik direnci, mecA, SCCmec, Gıda, Türkiye*

How to cite this article?

Issa G, Aydin A: Detection of methicillin resistant *Staphylococcus aureus* strains and typing of staphylococcal cassette chromosome *mec* from various foods originated different region from Turkey. *Kafkas Univ Vet Fak Derg,* 27 (3): 323-329, 2021.
DOI: 10.9775/kvfd.2021.25370

(*) Corresponding Author

Tel: +90 850 450 2828; Fax: +90 212 547 0068
E-mail: ghassan.issa@kocaelisaglik.edu.tr (G. İssa)

Introduction

Staphylococcus aureus is a microorganism that is highly resistant to environmental conditions and *S. aureus* widely found in environmental sources and can cause a large number of infections in both humans and animals. In addition, *S. aureus* is also widely found in food and food facilities, staff involved in food production, hospital staff and hospital settings. Personnel who carry and prepare food with their hands, especially in the food sector, are considered to be an important source of staphylococcal intoxications due to the risk of possible contamination [1-6].

Methicillin resistance in *S. aureus* strains occurs due to the production of low affinity penicillin binding proteins. Responsible for resistance to all beta-lactam antibiotics, PBP2a is encoded by the *mec*A gene. The *mec*A gene is located on a mobile, large genetic element called the staphylococcal cassette chromosome *mec*. The SCC*mec* structure of *S. aureus* consists of three elements: *mec* gene complex *(mec*A gene and its regulators *mec*I and *mec*R1), cassette chromosome recombinase *(ccr)* gene complex, and J (junkyard). SCC*mec*; they are divided into types according to *mec* and *ccr* gene complexes and subtypes according to differences in J regions. 13 SCC*mec* (I-XIII) types have been identified in *S. aureus* strains so far. Type I, II, III and VIII hospital-acquired methicillin-resistant *S. aureus* (HA-MRSA), types IV, V, VI and VII are CA-MRSA. While Type II and III cause multiple antibiotic resistance other than beta-lactam, Type I, IV and V do not carry multiple antibiotic resistance genes other than beta-lactam [7-15].

Turkey is located in the transition zone of the position as Asia, Europe and Africa. It has a very important geopolitical location. It is possible not only for humans or animals but also for microorganisms to pass between regions. This study was carried out in order to reveal the regional incidence of MRSA strains in Turkey, which is currently on the world agenda and draw attention in terms of public health for our country, and to make SCC*mec* typing in food-borne MRSA strains.

Material and Methods

Sampling

In this study, 560 dairy products (cheese), 89 bakery products (turkish raw flatbread [n:49], turkish handmade noodles [n:40]), 40 Ready To Eat Foods (raw meatballs [n:21], entrees [n:19]) and 11 meat products (sausages [n:6], salami [n:5]) to a total of 700 different food items in Turkey's seven regions (Black Sea region [n:100], Marmara region [n:100], Aegean region [n:100], Mediterranean region [n:100], Central Anatolia region [n:100], Southeastern Anatolia region [n:100], Eastern Anatolia region [n:100]) obtained from various points. Approximately 100 g of sample taken under aseptic conditions, depending on the type of sample, was brought to the laboratory under cold storage conditions (4°C) and microbiological analysis was performed as soon as possible.

Isolation and Identification of S. aureus from Food Samples

Food samples brought to the laboratory were weighed 10 g each in sterile stomacher bag and 90 mL peptone salt water (Maximum Recovery Diluent) solution (Oxoid CM 0733) was added and homogenized in stomacher (Seward 400, England) for about 2 min. From the obtained homogenization and dilutions, they were planted with the spreading technique on Baird Parker Agar (Oxoid, CM 0275) plates with tellurite added egg yolk (Oxoid SR 0054) and incubated for 24-48 h at 37°C. *S. aureus* colonies which ocur due to the lecithinase activity on BP agar with a diameter of 2-3 mm 'gray-black' zones around were considered suspicious. *S. aureus* isolates growing on BP agar were inoculated into Mannitol Salt Agar (Oxoid CM 0085) to evaluate mannitol fermentation at a later stage. Then, biochemical confirmation was performed on *S. aureus* strains by Gram staining, catalase test, DN*ase* test (Oxoid CM 321) and latex agglutination test (Oxoid, DR 100M) [16-20].

Determination of Methicillin Resistant S. aureus Strains by Phenotypic Methods

Kirby-Bauer disc diffusion method was carried out in accordance with the recommendations of CLSI (2009) in determining the methicillin resistance of the strains defined as *S. aureus*. Conventionally, cefoxitin disk diffusion (FOX 30 µg, Oxoid CT0119B), oxacillin disk diffusion (OX 1 µg, Oxoid CT0159B) and oxacillin resistance screening agar (ORSAB, Oxoid CM 1008) tests were used to determine methicillin resistant *S. aureus* strains [21-24].

Verification of S. aureus strains by Molecular Methods

DNA extraction of *S. aureus* strains was performed according to the instruction of Genomic DNA Isolation from Bacteria kit (RTA, 09005100). For PCR mix 30 µL Sterile dH_2O, 5 µL $(NH_4)_2SO_4$ containing 10x buffer (Fermentas, EP0402), 3 µL $MgCl_2$ (25Mm, Fermentas, EP0402), 1 µL Forward primer (10 mM), 1 µL Revers primer (10 mM), 5 µL dNTP mix (2 mM each, Fermentas, R0241), 0.24 µL *Taq* DNA polymerase (500 U, Fermentas, EP0402) and 5 µL Genomic DNA template after preparation to 50 µL containing PCR tubes were placed in the Thermal cycler (Biorad, Italy).

Following the amplification process, PCR products were analyzed by horizontal agarose gel electrophoresis. While protein A *(spa)*, thermonuclease *(nuc)* and coagulase *(coa)* genes were examined for molecular confirmation of strains, the presence of *mec*A gene was investigated for the determination of methicillin resistance. Primers and thermal cycling conditions used for the verification of *S. aureus* strains and detection of *mec*A are given in *Table 1* [25-27].

Table 1. *Primers and thermal cycling conditions used for the verification of S. aureus isolates and detection of mecA*

Target	Amplicon Size (bp)	Primer; Oligonucleotide Ssequence (5'-3')	Thermal Cycling Conditions
spa	100-450	1113F; TAAAGACGATCCTTCGGTGAGC 1514R; CAGCAGTAGTGCCGTTTGCTT	Initial Denaturation 80°C 5 min **Denaturation 94°C 45 s** **Annealing 60°C 45 s = 35 Cycles** **Extension 72°C 90 s** Final Extension 72°C 10 min
nuc	416	F; GGCAATTGTTTCAATATTAC R; TTTTATTTGCATTTTCTACC	Initial Denaturation 80°C 5 min **Denaturation 94°C 45 s** **Annealing 60°C 45 s = 35 Cycles** **Extension 72°C 90 s** Final Extension 72°C 10 min
coa	500-650	F; ATAGAGATGCTGGTACAGG R; GCTTCCGATTGTTCGATGC	Initial Denaturation 80°C 5 min **Denaturation 94°C 45 s** **Annealing 60°C 45 s = 35 Cycles** **Extension 72°C 90 s** Final Extension 72°C 10 min
mecA	162	*mec*A P4; TCCAGATTACAACTTCACCAGG *mec*A P7; CCACTTCATATCTTGTAACG	Initial Denaturation 80°C 5 min **Denaturation 94°C 45 s** **Annealing 60°C 45 s = 35 Cycles** **Extension 72°C 90 s** Final Extension 72°C 10 min

Table 2. *Primers and thermal cycling conditions used in SCCmec typing*

Primer	Oligonucleotide Sequence (5'-3')	Amplicon Size (bp)	Specificity (SCC*mec* type)	Thermal Cycling Conditions
CIF2F2 CIF2R2	TTCGAGTTGCTGATGAAGAAGG ATTTACCACAAGGACTACCAGC	495	I	Initial Denaturation 94°C 4 min **Denaturation 94°C 30 s** **Annealing 53°C 30 s = 30 Cycles** **Extension 72°C 1min** Final Extension 72°C 4 min
KDPF1 KDPR1	AATCATCTGCCATTGGTGATGC CGAATGAAGTGAAAGAAAGTGG	284	II	
MECIP2 MECIP3	ATCAAGACTTGCATTCAGGC GCGGTTTCAATTCACTTGTC	209	II, III	
DCSF2 DCSR1	CATCCTATGATAGCTTGGTC CTAAATCATAGCCATGACCG	342	I, II, IV	
RIF4F3 RIF4R9	GTGATTGTTCGAGATATGTGG CGCTTTATCTGTATCTATCGC	243	III	
RIF5F10 RIF5R13	TTCTTAAGTACACGCTGAATCG GTCACAGTAATTCCATCAATGC	414	III	

Mutiplex PCR was performed using primer sequences and protocol suggested by Oliveira and Lencastre [26] for SCC*mec* typing in *mec*A positive samples. Primers and thermal cycling conditions used in SCC*mec* typing are given in *Table 2*.

Results

The distribution of the sample types provided in our research by product groups and regions is given in *Table 3*. A total of 67 (9.57%) *S. aureus* strains were isolated from 700 food samples analyzed within the scope of the project. Of the 67 *S. aureus* strains; 50 (74.62%) were obtained from dairy products, 16 (23.88%) from bakery products, and 1 (1.50%) from RTE food samples. On the other hand, *S. aureus* was not found in any meat product samples. Accordingly, the distribution and percentages of *S. aureus* strains by product groups and regions are shown in *Table 4*. *spa*, *nuc* and *coa* genes were verified by PCR in 67 *S. aureus* strains isolated by cultural methods. The gel image of the detected genes *spa* (100-450 bp) is shown in *Fig. 1-A*, *nuc* (416 bp) in *Fig. 1-B* and *coa* (500-600 bp) in *Fig. 1-C*. Only 1 (0.14%) of 67 *S. aureus* strains isolated was identified as both phenotypically and genotypically MRSA. The detected MRSA strain was isolated from a cheese (dairy product) collected from the Marmara region. While the growth image of the MRSA positive strain in ORSAB medium is given in *Fig. 1-D*, the disk diffusion test image of oxacillin (1 µg) and cefoxitin (30 µg) is given in *Fig. 1-E*. *mec*A gene responsible for methicillin resistance was also detected by genotypic (PCR) methods in *S. aureus* strain isolated as MRSA by phenotypic methods. SCC*mec* typing of dairy derived MRSA strain detected by both phenotypic and genotypic methods and SCC*mec* Type IV (community-acquired) was found in our study. The detected *mec*A (162 bp) and SCC*mec* Type IV (342 bp) gel image is shown in *Fig. 1-F*.

Table 3. *The distribution of the sample types provided by product groups and regions*

Product Groups	n	Marmara Region n (%)	Aegean Region n (%)	Black Sea Region n (%)	Central Anatolia Region n (%)	Eastern Anatolia Region n (%)	Southeastern Anatolia Region n (%)	Mediterranean Region n (%)
Meat Products	11	11 (100)	-	-	-	-	-	-
Dairy Products	560	58 (10.35)	92 (16.45)	82 (14,64)	67 (11.96)	95 (16,96)	88 (15.71)	78 (13.93)
Bakery Products	89	15 (16.85)	2 (2.25)	17 (19.11)	18 (20.22)	3 (3.37)	12 (13.48)	22 (24.72)
Ready Meals	40	16 (40)	6 (15)	1 (2.5)	15 (37.5)	2 (5)	-	-
Total (%)	700	100 (14.29)	100 (14.29)	100 (14.29)	100 (14.29)	100 (14.29)	100 (14.29)	100 (14.29)

Table 4. *The distribution and percentages of S. aureus isolates by product groups and regions*

Product Groups	S. aureus n (%)	Marmara Region n (%)	Aegean Region n (%)	Black Sea Region n (%)	Central Anatolia Region n (%)	Eastern Anatolia Region n (%)	Southeastern Anatolia Region n (%)	Mediterranean Region n (%)
Meat Products	-	-	-	-	-	-	-	-
Dairy Products	50 (74.62)	5 (10)	7 (14)	4 (8)	7 (14)	7 (14)	11 (22)	9 (18)
Bakery Products	16 (23.88)	2 (12.5)	-	8 (50)	3 (18.75)	-	-	3 (18.75)
Ready Meals	1 (1.49)	1	-	-	-	-	-	-
Total (%)	67 (100)	8 (11.94)	7 (10.45)	12 (17.91)	10 (14.93)	7 (10.45)	11 (16.41)	12 (17.91)

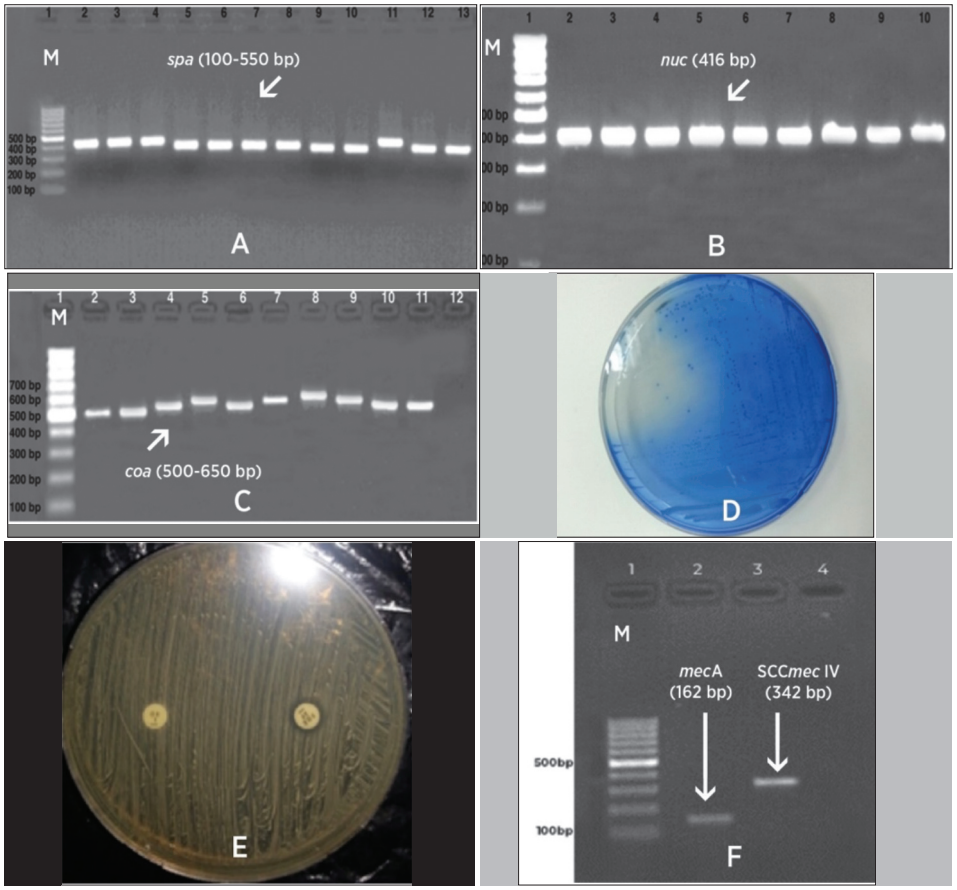

Fig 1. A- *spa* gene positive gel image (lane 1: DNA ladder [100 bp], lanes 2-13: *spa* gene positive samples); **B-** *nuc* gene positive gel image (lane 1: DNA ladder [100 bp], lanes 2-10: *nuc* gene positive samples); **C-** *coa* gene positive gel image (lane 1: DNA ladder [100 bp], lanes 2-11: positive samples of the *coa* gene, lane 12: negative control)**; D-** MRSA development on ORSAB agar**; E-** Disk diffusion test of oxacillin (1 µg) and cefoxitin (30 µg) in MHA; **F-** *mec*A gene positive and SCC*mec* Type IV gel image (lane 1: DNA ladder [100 bp], lane 2: *mec*A gene positive sample, lane 3: SCC*mec* Type IV positive sample, lane 4: negative control)

Discussion

Staphylococcus aureus causes significant infections in animals and humans. It is a source of nosocomial infection in humans. One of the most important hospital and community-acquired factors that lead to unsuccessful treatment and deaths is MRSA [28]. In addition, the increase in the frequency of MRSA strains in recent years has revealed the need to develop effective strategies to control staphylococcal infections and the development of resistance in microorganisms against antibiotics. Therefore, it is of great importance to know the epidemiology, pathogenesis and population genetics of *S. aureus*. Misuse and abuse of antibiotics emerges as an important factor that increases MRSA colonization. In our country, misuse of antibiotics and related resistance problems are frequently encountered in both veterinary and human medicine. CA-MRSA infections can be transmitted by direct contact as well as by consumption of animal-based foods. Poor personal hygiene increases the potential for contamination. In addition to the use of antibiotics in our country, there are also important problems in ensuring food hygiene and personal hygiene. Normanno et al.[29], isolated 160 (9.80%) *S. aureus* strains from 1634 animal origin food they collected, and reported that only 6 (3.75%) of the strains were MRSA. Meemken et al.[30], examined 687 swab samples taken from pig nose for MRSA and reported that 85 (12.37%) were positive. De Neeling et al.[31], reported that 209 (38.70%) of the 540 pig samples taken from the slaughterhouse were MRSA positive. Qudduomi et al.[32] evaluated 6 (0.84%) out of 717 meat samples as MRSA positive. Kwon et al.[11] detected the presence of *S. aureus* in 292 (31.40%) of the 930 food samples (cattle, chicken and pig) they collected from slaughterhouses and markets. They reported that 766 *S. aureus* strains isolated from these 292 foodstuffs, 4 of the isolated strains carried *mec*A and phenotypically only 3 were MRSA. They stated that 3 MRSA strains from 2 (0.22%) chicken samples detected and all 3 were SCC*mec* Type III. On the other hand, similar to our study, Uçan and Aslan [33] detected methicillin resistance in only 1 (1.33%) of 75 *S. aureus* strains. In the study conducted by İssa and Aksu [2], 119 (29.31%) *S. aureus* strains were detected from 406 raw milk samples, and only 1 (0.84%) of these 119 strains was reported to be MRSA. In another study, Kwon et al.[34], found that 14 (0.018%) of the 75335 milk samples they examined in Korea were MRSA. SCC*mec* typing of 14 isolated MRSA strains was made and it was revealed that all strains were Type IV. A total 67 (9.57%) strains of *S. aureus* were isolated from 700 food samples analyzed in our study, and it was found that only 1 (0.14%) strain was MRSA, and SCC*mec* Type IV (community acquired) were found.

Several studies [2,4,35-38] reported that the prevalence of *S. aureus* is relatively high in milk and dairy products. These reports are consistent with our results. Normanno et al.[37] examined the swabs taken from 11384 foodstuffs and food contact surfaces they bought from the markets in Italy and isolated 1971 (17.3%) coagulase positive *S. aureus*. At the same study, coagulase positive *S. aureus* was detected from 1245 (23.1%) samples of 5369 meat products and 168 (38.4%) of 437 raw milk samples. Juhasz-Kaszanyitzky et al.[38] examined 595 milk samples taken from cows with subclinical mastitis and isolated 375 (63.02%) of *S. aureus*. Of these 375 *S. aureus* strains, only 27 (7.20%) were found to be MRSA. Argudin et al.[4] found in a study they conducted that 2 (3.12%) of the 64 *S. aureus* strains they isolated from food and food processors were MRSA and when SCC*mec* was typed, and both were Type IV. Stastkova et al.[39], reported that they isolated 34 (22.22%) *S. aureus* strains out of 153 goat milk samples, 5 (3.26%) of the isolates were MRSA and these 5 strains were Type IV as a result of SCC*mec* typing. Similarly, Vanderhaeghen et al.[40] stated that 11 (9.30%) of 118 *S. aureus* strains isolated from mastitis cows were MRSA, 5 of them were SCC*mec* IV, 5 were SCC*mec* V and the other 1 strain could not be typed. Huber et al.[36] isolated 142 *S. aureus* strains from mastitis milk and stated that only 2 (1.40%) of the strains were MRSA and these 2 strains belonged to Type V as a result of SCC*mec* typing. In another study, Gülbandılar [17] took samples from the nasal mucosa of 3048 people from food processors (cooks, bakers, pastry makers, etc.) and tradesmen (barbers, hairdressers, etc.) who had direct contact with the public, and isolated a total 217 (7.12%) *S. aureus* strains. Gülbandılar [17] reported that only 12 (5.30%) of the 217 *S. aureus* strains they isolated were MRSA strains. Similarly, in a study conducted by Lee [22], they isolated 421 (22%) *S. aureus* from 1913 samples taken from cattle, pigs and chickens, and 28 (6.65%) of these 421 *S. aureus* strains were phenotypically MRSA. However, they found that only 15 (3.56%) of them were MRSA in genotypic evaluation.

Yang et al.[10], isolated 69 (12.54%) *S. aureus* strains out of 550 RTE food samples from the markets in their study between 2011-2014, and only 6 (1.09%) of these strains were MRSA, as a result of SCC*mec* typing. They found that 2 of them MRSA were evaluated as Type IV and 2 of them MRSA were evaluated as Type V, but the other 2 MRSA strains could not be typed. Wang et al.[9], isolated 455 (23%) *S. aureus* strains in a study they conducted with 1979 food samples collected from the markets between 2008-2012, only 17 (0.90%) of these strains were MRSA, and confirmed that as a result of SCC*mec* typing; 4 of them MRSA were Type II and 9 MRSA were Type IV, 2 of them MRSA strains were evaluated as Type V and the remaining strains could not be typed. The fact that both *S. aureus* and methicillin resistant *S. aureus* strains have been reported at different rates in studies conducted in our country and in the world may be caused by the differences in geographical regions, the difference of the samples from which the strains were isolated and the increase in resistant strains over time. Depending on the season, different rates of agent isolation may be in question. In this context, Stastkova et al.[39] reported that the rates of *S. aureus* and MRSA isolated from milk samples can be different according to the seasons.

CA-MRSA strains can cause clinical cases ranging from skin infections to fatal pneumonia and sepsis, as well as food-borne diseases. Several virulent genes have been identified, such as Panton-Valentin Leukocidin (PVL), which cause degradation of leukocytes and tissue necrosis in CA-MRSA strains and are not found in hospital-acquired MRSA strains [41-44]. The problem of antimicrobial resistance, which has reached serious level in recent years, causes adverse effects such as increased mortality rates, prolonged treatment periods, loss of work force due to prolonged hospital stay, and the necessity to use newly developed expensive antibiotics as a result of nosocomial and community-acquired bacterial infections in developed and developing countries.

As a result, consumption of animal origin foods is an important source of contamination of MRSA strains for humans. There is need for studies to determine the source of antibiotic resistant bacteria by using molecular methods. Based on these researches, it should be a correct policy regarding the use of antibiotics in human infections, veterinary medicine and animal husbandry. It is considered that generalizing the studies on antibiotic resistance and determining the types of MRSA will be beneficial for the protection of public health in our country.

Acknowledgement

This study was supported from Technological Research Council of Turkey (TUBITAK-2216).

Conflict of Interest

The authors declare that they have no conflict of interest.

Author Contributions

Gİ planned and designed the research, carried out experiments. Gİ and AA discussed the results. All authors contributed to writing of the final manuscript.

References

1. Wang W, Baloch Z, Jiang T, Zhang C, Peng Z, Li F, Fanning S, Ma A, Xu J: Enterotoxigenicity and antimicrobial resistance of *Staphylococcus aureus* isolated from retail food in China. *Front Microbiol*, 8:2256, 2017. DOI: 10.3389/fmicb.2017.02256

2. İssa G, Aksu H: Detection of methicillin-resistant *Staphylococcus aureus* in milk by PCR-based phenotyping and genotyping. *Acta Vet Eurasia*, 46, 120-124, 2020. DOI: 10.5152/actavet.2020.20011

3. Erol İ: Gıda Hijyeni ve Mikrobiyolojisi. 135-144, Pozitif Matbaacılık, Ankara, 2007.

4. Argudin MA, Mendoza MC, Gonzalez-Hevia MA, Bances M, Guerra B, Rodicio MR: Genotypes, exotoxin gene content, and antimicrobial resistance of *Staphylococcus aureus* strains recovered from foods and food handlers. *Appl Environ Microbiol*, 78 (8): 2930-2935, 2012. DOI: 10.1128/AEM.07487-11

5. Vural H, Öztan A: Effects of starter culteres on growth of *Staphylococcus aureus* in fermented meat products. *Gıda*, 18 (4): 259-263, 1993.

6. Sergelidis D, Angelidis AS: Methicillin-resistant *Staphylococcus aureus*: A controversial food-borne pathogen. *Lett Appl Microbiol*, 64, 409-418, 2017. DOI: 10.1111/lam.12735

7. Singh-Moodley A, Strasheim W, MogokotlengR, Ismail H, Perovic O: Unconventional SCC*mec* types and low prevalence of the Panton-Valentine Leukocidin exotoxin in South African blood culture *Staphylococcus aureus* surveillance isolates, 2013-2016. *PLoS ONE*, 14 (11): e0225726, 2019. DOI: 10.1371/journal.pone.0225726

8. Chen L, Mediavilla JR, Oliveira DC, Willey BM, Lencastre HD, Kreiswirth BN: Multiplex real-time PCR for rapid staphylococcal casette chromosome *mec* typing. *J Clin Microbiol*, 47, 3692-3706, 2009. DOI: 10.1128/JCM.00766-09

9. 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 Pathog Dis*, 11 (4): 281-286, 2014. DOI: 10.1089/fpd.2013.1643

10. Yang X, Zhang J, Yu S, Wu Q, Guo W, Huang J, Cai S: Prevalence of *Staphylococcus aureus* and methicillin-resistant *Staphylococcus aureus* in retail ready-to-eat foods in China. *Front Microbiol*, 7:816, 2016. DOI: 10.3389/fmicb.2016.00816

11. Kwon NH, Park KT, Jung WK, Youn HY, Lee Y, Kim SH, Bae W Lim JY, Kim JY, Hong SK, Park YH: Chracteristics of methicillin resistant *S. aureus* isolated from chicken meat and hospitalized dogs in Korea and their epidemiological relatedness. *Vet Microbiol*, 117, 304-312, 2006. DOI: 10.1016/j.vetmic.2006.05.006

12. Saber H, Jasni AS, Jamaluddin TZMT, Ibrahim R: A review of staphylococcal cassette chromosome *mec* (SCC*mec)* types in coagulase-negative staphylococci (CoNS) species. *Malays J Med Sci*, 24 (5): 7-18, 2017. DOI: 10.21315/mjms2017.24.5.2

13. Reichmann NT, Pinho MG: Role of SCC*mec* type in resistance to the synergistic activity of oxacillin and cefoxitin in MRSA. *Sci Rep*, 7:6154, 2017. DOI: 10.1038/s41598-017-06329-2

14. Afshari SG, Sepahi AA, Goudarzi H, Tabrizi MS, Goudarzi M, Hajikhani B, Heidary M, Azimi H: Distribution of SCC*mec* types in methicillin-resistant *Staphylococcus aureus* isolated from burn patients. *Arch Clin Infect Dis*, 12 (2): e62760, 2017. DOI: 10.5812/archcid.62760

15. Ghanbari F, Saberianpour S, Zarkesh-Esfahani FS, Ghanbari N, Taraghian A, Khademi F: Staphylococcal cassette chromosome *mec* (SCC*mec*) typing of methicillin-resistant *Staphylococcus aureus* strains isolated from community- and hospital-acquired infections. *Avicenna J Clin Microbiol Infect*, 4 (2): 42244, 2017. DOI: 10.5812/ajcmi.42244

16. ISO 6888-1: 1999/A1: 2003, Microbiology of food and animal feeding stuffs - Horizontal method for the enumeration of coagulase-positive staphylococci *(Staphylococcus aureus* and other species) - Part 1: Technique using Baird-Parker Agar medium (includes amendment A1: 2003).

17. Gülbandilar A: Investigation of *Staphylococcus aureus* carriage and antibiotic sensitivity in nose mucous membrane in Kütahya Province. *DPÜ Fen Bil Enst Derg*, 18, 1-6, 2009.

18. Amirkhiz MF, Rezaee MA, Hasani A, Aghazadeh M, Naghili B: SCC*mec* typing of methicillin-resistant *Staphylococcus aureus*: An eight year experience. *Arch Pediatr Infect Dis*, 3 (4): e30632, 2015. DOI: 10.5812/pedinfect.30632

19. APHA (American Publich Health Association): Compendium methods for the microbiological examination of foods. 4, 387-404, Washington DC, 2001.

20. Oxoid Manual: 8, Oxoid Ltd., Basingstok, UK, 1998.

21. Wikler MA, Cockerill FR, Craig WA, Dudley MN: Performance standards for antimicrobial susceptibility. Clinical and Laboratory Standards Institute. 17th ed., Informational Supplement, 27, 44-51, 2007.

22. Lee JH: Methicillin (oxacillin)-resistant *Staphylococcus aureus* strains isolated from maor food animals and their potential transmission to humans. *Appl Environ Microbiol*, 69 (11): 6489-6494, 2003. DOI: 10.1128/AEM.69.11.6489-6494.2003

23. Clinical and Laboratory Standards Institute (CLSI): Performance standards for antimicrobial susceptibility testing. 23rd Informational Supplement. CLSI document M100-S23, 33 (1): USA, 2013.

24. Nahimana I, Francioli P, Blanc DS: Evaluation of three chromogenic

media (MRSA-ID, MRSA-Select and CHROMagar MRSA) and ORSAB for surveillance cultures of methicillin-resistant *Staphylococcus aureus*. *Clin Microbiol Infect*, 12, 1168-1174, 2006. DOI: 10.1111/j.1469-0691.2006.01534.x

25. Hashemizadeh Z, Hadi N, Mohebi S, Kalantar-Neyestanaki D, Bazargani A: Characterization of SCC*mec, spa* types and multi drug resistant of methicillin-resistant *Staphylococcus aureus* isolates among inpatients and outpatients in a referral hospital in Shiraz, Iran. *BMC Res Notes*, 12:614, 2019. DOI: 10.1186/s13104-019-4627-z

26. Oliveira DC, De Lencastre H: Multiplex PCR strategy for rapid identification of structural types and variants of the *mec* element in methicillin resistant *Staphylococcus aureus*. *Antimicrob Agents Chemother*, 46 (7): 2155-2161, 2002. DOI: 10.1128/AAC.46.7.2155-2161.2002

27. Sudagidan M, Aydin A: Screening virulence properties of staphylococci isolated from meat and meat products. *Wien Tieraerztl Mschr*, 95, 128-134, 2008.

28. Bahrami N, Motamedi H, Tofighi SER, Akhoond MR: SCC*mec* typing and Panton-Valentine leukocidin occurrence in methicillin resistant *Staphylococcus aureus* isolates from clinical samples of Ahvaz, southwest of Iran. *Microbiol Medica*, 34:8050, 2019. DOI: 10.4081/mm.2019.8050

29. Normanno G, Corrente M, La Salandra G, Dambrosio A, Quaglia NC, Parisi A, Greco G, Bellacicco AL, Virgilio S, Celano GV: Methicillin-resistant *Staphylococcus aureus* (MRSA) in foods of animal origin product in Italy. *Int J Food Microbiol*, 117, 219-222, 2007. DOI: 10.1016/j.ijfoodmicro.2007.04.006

30. Meemken D, Cuny C, Witte W, Blaha T: Occurrence of MRSA in pıgs in the northwest of Germany. *Proceedings of the International Pig Veterinary Society Congress,* Durban, South Africa, 22-26, June-2008.

31. De Neeling AJ, Van Den Broek MJM, Spalburg EC, Van Santen-Verheuvel MG, Dam-Deisz WDC, Boshuizen HC, Van De Giessen AW, Van Duijkeren E, Huijsdens XW: High percentage of methicillin-resistant *Staphylococcus aureus* in pigs. *Vet Microbiol*, 122, 366-372, 2007. DOI: 10.1016/j.vetmic.2007.01.027

32. Qudduomi SS, Bdour SM, Mahasneh SM: Isolation and characterisation of methicillin-resistant *Staphylococcus aureus* from livestock and poultry meat. *Ann Microbiol*, 56, 155-161, 2006.

33. Uçan US, Aslan E: İnek mastitisislerinden izole edilen koagülaz pozitif stafilokok suşlarının penisilin direnci ve bazı antibiyotiklere duyarlılıkları. *Vet Bil Derg*, 18, 19-22, 2002.

34. Kwon NH, Park KT, Moon JS, Jung WK, Kim SH, Kim JM, Hong SK, Koo HC, Joo YS, Park YH: Staphylococcal casette chromosome *mec* (SCC*mec*) characterization and molecular analysis for methicillin-resistant *Staphylococcus aureus* and novel SCC*mec* subtype IVg isolated from bovine milk in Korea. *J Antimicrob Chemother*, 56, 624-632, 2005. DOI: 10.1093/jac/dki306

35. Aydin A, Sudagidan M, Muratoglu K: Prevalence of staphylococcal enterotoxins, toxin genes and genetic-relatedness of foodborne *Staphylococcus aureus* strains isolated in the Marmara Region of Turkey. *Int J Food Microbiol*, 148, 99-106, 2011. DOI: 10.1016/j.ijfoodmicro.2011.05.007

36. Huber H, Koller S, Giezendanner N, Stephan R, Zweifel L: Prevalence and characteristics of methicillin-resistant *Staphylococcus aureus* in human in contact with farm animals, in livestock, and in food of animal origin, Switzerland, 2009. *Euro Surveill*, 15 (16): pii=19542. 2010.

37. Normanno G, Firinu A, Virgilio S, Mula G, Dambrosio A, Poggiu A, Decastelli L, Mioni R, Scuota S, Bolzoni G, Di Giannatale E, Salinetti AP, La Salandra G, Bartoli M, Zuccon F, Pirino T, Sias S, Parisi A, Quaglia NC, Celano GV: Coagulase-positive *Staphylococci* and *Staphylococcus aureus* in food products marketed in Italy. *Int J Food Microbiol*, 98, 73-79, 2005. DOI: 10.1016/j.ijfoodmicro.2004.05.008

38. Juhasz-Kaszanyitzky E, Janosi S, Somogyi P, Dan A, Van Der Graaf-Von Bloois L, Van Duijkeren E, Wagenaar JA: MRSA transmission between cows and humans. *Emerg Infect Dis*, 13 (4): 630-632, 2007.

39. Stastkova Z, Karpiskova S, Karpiskova R: Occurrence of methicillin-resistant strains of *Staphylococcus aureus* at a goat breeding farm. *Vet Med*, 54 (9): 419-426, 2009. DOI: 10.17221/88/2009-VETMED

40. Vanderhaeghen W, Cerpentier T, Adriaensen C, Vicca J, Hermans K, Butaye P: Methicillin-resistant *Staphylococcus aureus* (MRSA) ST398 associated with clinical and subclinical mastitis in Belgian cows. *Vet Microbiol*, 144 (1-2): 166-171, 2010. DOI: 10.1016/j.vetmic.2009.12.044

41. Funaki T, Yasuhara T, Kugawa S, Yamazaki Y, Sugano E, Nagakura Y, Yoshida K, Fukuchi K: SCC*mec* typing of PVL positive community-acquired *Staphylococcus aureus* (CAMRSA) at a Japanese hospital. *Heliyon 5*:e01415, 2019. DOI: 10.1016/j.heliyon.2019.e01415

42. Ul Bashir Y, Bali N, Sharma S, Jahan T, Shafi H: SCC*mec* Type IV and V methicillin resistant *Staphylococcus aureus* intrusion in healthcare settings. *Am J Clin Microbiol Antimicrobial*, 2 (1): 1032, 2019.

43. Abdurahman MA, Tosun İ, Durukan İ, Khorshidtalab M, Kılıç AO: Four temperate bacteriophages from methicillin-resistant *Staphylococcus aureus* show broad bactericidal and biofilm removal activities. *Kafkas Univ Vet Fak Derg*, 27 (1): 29-36, 2021. DOI: 10.9775/kvfd.2020.24680

44. Tsilochristou O, Toit G, Sayre PH, MD, Roberts G, Lawson K, Sever ML, Bahnson HT, Radulovic S, Basting M, Plaut M, Lack G: Association of *Staphylococcus aureus* colonization with food allergy occurs independently of eczema severity. *J Allergy Clin Immunol*, 144 (2): 494-503, 2019. DOI: 10.1016/j.jaci.2019.04.025