

Methicillin-Resistant *Staphylococcus aureus* (MRSA), Vancomycin-Resistant *Staphylococcus aureus* (VRSA), and Vancomycin-Resistant *Enterococci* (VRE) Contamination of Food Samples in Iran: A Systematic Review and Meta-Analysis

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ABSTRACT

During the last years, antimicrobial resistance has become one of the greatest challenges in clinical settings. *Staphylococcus aureus* and *Enterococcus* are important nosocomial pathogens worldwide. In different corners of the world, methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant *enterococci* (VRE) have been isolated from various sources including different foods of animal and plant origin; however, information about food-related vancomycin-resistant *Staphylococcus aureus* (VRSA) is limited. The current review describes the presence of MRSA, VRSA and VRE in different food samples of animal and plant origin in Iran. Databases including PubMed, ScienceDirect, Scopus, SID, and Google Scholar were searched and a total of 65 articles were retrieved (published between 2006 and September 2020) reporting antibiotic-resistant *S. aureus* and *Enterococcus* in food in Iran. The overall prevalence meta-analysis was calculated by using "meta prop program" in STATA statistical software. Finally, 42 studies were included in the present review. These reports indicated high rates of MRSA, VRSA and VRE strains in different types of raw and processed meat, raw milk, traditional cheese, restaurant, and hospital foods. The meta-analysis showed that the highest pooled ES of MRSA was for pastry products and the lowest pooled ES of MRSA was for miscellaneous foods. Thus, higher priority should be given to the development of effective surveillance systems which can track the appropriate use of wide-spectrum antibiotics and spread of antimicrobial-resistant organisms throughout food supply chains. Future research should assess the risk of antimicrobial resistance transmission to consumers following intake of MRSA, VRSA, and VRE-contaminated foodstuffs.

Keywords: Antimicrobial resistance, MRSA, VRSA, VRE, Iranian Foodstuffs

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1. Introduction

Staphylococcal food poisoning has been introduced as the third cause of food-borne disease throughout the world (1). *Staphylococcus aureus* is one of the most worrying pathogens in food due to the ability of some strains to produce heat-stable enterotoxins that can produce gastrointestinal disorders (2). Also, as an opportunistic pathogen for both humans and animals, it can cause pneumonia, osteomyelitis, brain abscesses, toxic shock syndrome, wound infections, bacteremia, and meningitis (3).

Enterococcus bacteria are naturally present in the gastrointestinal microflora of warm-blooded animals and humans (4). For many years, *Enterococcus* spp. were believed to be harmless to humans and medically unimportant (5). However, these bacteria can cause hospital-acquired infections such as urinary tract and wound infections, bacteremia, and endocarditis (4).

Besides toxic effect of foodborne bacterial pathogens, their resistance to antibiotics remains a major threat for animal and human health and its prevalence has increased in recent decades (6). The World Health Organization (WHO) introduced the issue of antimicrobial resistance as one of the biggest health challenges of the 21st century (7). Foodstuffs are an important vehicle for transmission of antibiotic-resistant bacterial strains to the gastrointestinal tract of consumers (8-10). Livestock and livestock-derived foods can serve as a reservoir for human infections caused by antibiotic-resistant *Staphylococcus* strains (1) and potentially transfer resistant *Enterococcus* species (11). In fact, the over-use and often irrational use of prophylactic antibiotics accelerate the emergence and spread of drug-resistant strains (12).

A well-known player in the antimicrobial resistance crisis is methicillin-resistant *S. aureus* (MRSA) (13). Multi-drug resistance in pathogenic bacteria was first mentioned in the 1950s (14). After emergence and dissemination of MRSA strains, vancomycin, a glycopeptide antibiotic, was used for the treatment of infections caused by methicillin-resistant *Staphylococci*. In 1996, vancomycin-resistant *S. aureus* strains (VRSA) emerged in Japan. Because vancomycin is not regularly used for treating infections in animals, few reports on VRSA strains in veterinary medicine exist (15). Vancomycin-resistant *Enterococcus* (VRE) (more frequently seen for *E. faecium* and less for *E. faecalis*) has a lower epidemiological impact compared to MRSA and they are almost exclusively restricted to healthcare settings (13).

Methicillin-Resistant *Staphylococcus aureus* (MRSA)

From different parts of the world, MRSA species have shown resistance towards first-line antibiotics

including beta-lactams (16). MRSA is known to be resistant against almost all types of penicillin and β -lactam antibiotics (17). Methicillin resistance in *staphylococci* is probably the most mysterious among all bacterial antibiotic resistance (18). Mechanism of the intrinsic form of penicillin resistance in *S. aureus* is unknown (18, 19). Methicillin resistance in *S. aureus* has been considered the most important clinical resistance trait (20).

During the past years, the rate of occurrence of infections caused by MRSA significantly increased in livestock (i.e., livestock-associated MRSA (LA-MRSA)) (3). LA-MRSA may also be transmitted to humans through occupational contact with infected livestock and consumption of foods prepared from contaminated animals (3, 21). Animals are recognized as the main source of new pathogenic strains of *S. aureus* for human, particularly the clones which possess no host specificity (3). The probability of MRSA transmission via food was largely unknown until 1994 (22). MRS isolates were initially reported in dairy animals in 1972 (from mastitis cows) (16).

Vancomycin-Resistant *S. aureus* (VRSA)

Following the advent of MRSA strains and their rapidly increasing resistance to different groups of antibiotics (like penicillin, cephalosporins, aminoglycosides, fluoroquinolones, and macrolides), vancomycin was used for treatment of patients infected with MRSA or other gram-positive bacteria (23). The first strains of *S. aureus* with reduced sensitivity to vancomycin were reported in Japan in 1997 (24). Thereafter, VRSA as well as vancomycin intermediate-resistant *S. aureus* (VISA) were identified in the United States, the United Kingdom, Germany, Portugal, Brazil, China, Bangladesh, and Jordan (23).

Only a few reports were published about vancomycin-resistant strains of *S. aureus* in veterinary medicine because vancomycin is not regularly used in the treatment of infected animals. The emergence of such strains in animals could be due to the contamination of pasture soils or environment with vancomycin-resistant isolates originating from patients who had been to a healthcare facility and resided very close to animals and milching of milch animal (15).

Vancomycin-Resistant *Enterococci* (VRE)

Vancomycin-resistant *enterococci* (VRE) were first recognized in the United Kingdom and France in 1986 (25, 26). The emergence of VRE was reported to be related to severe clinical use in the US and cross

resistance in Europe, and subsequently, the agricultural use of avoparcin as a growth promoter (25).

In developing countries, antibiotics and anthelmintics (compounds such as antibiotics, coccidiostats, and growth promoting hormones) are widely used to control the pathogens and satisfy the increased demand for meat and dairy products. Hence, some studies in Iran focused on the prevalence of MRSA, VRSA, and VRE in foodstuffs. The objective of this review was to collect and thoroughly review the available information regarding the prevalence of MRSA, VRSA, and VRE in different food samples of animal and plant origin in Iran.

2. Materials and Methods

Search Strategy

PubMed, ScienceDirect, Scopus, SID, and Google Scholar were searched for articles published between 2006 and September 2020 using the following keywords: “*Staphylococcus aureus*”, “Food”, “*Enterococcus*”, “Antimicrobial resistance”, “Prevalence”, “Iran”, “MRSA”, “VRSA”, and “VRE”. Initial screening of the retrieved articles was done through reading their titles and/or abstracts. Sixty-five papers were selected, of which 23 articles including duplicate publications, conference abstracts, and studies that did not contain complete information were excluded. The location of the studies was geocoded using the Google My Maps (<https://www.google.com/mymaps>) software. The ArcMap 10.6 software was used to manage the geographic data, create maps, and provide an overview of the geographical distribution of MRSA contamination in six different food categories including milk, cheese, other dairy products, pastry products, meat products, and miscellaneous foods and VRE in miscellaneous foods in different regions of Iran.

Meta-Analysis

The overall prevalence meta-analysis was calculated by using “meta prop program” in STATA statistical software (STATA, College Station, TX, USA, version 15.0). To estimate the pooled prevalence and corresponding 95% confidence interval (CI), the random-effects model was applied. Cochran's Q test and I^2 were used to determine statistical heterogeneity between studies. The meta regression test was used to evaluate possible sources of heterogeneity based on the duration and location of the study. $P < 0.05$ was considered as a statistically significant.

3. Results and Discussion

Presence of MRSA in Various Food Samples

The number of articles about the prevalence of MRSA strains in Iran has increased in recent years. However, the available information is limited to reports from a few cities. Most of these studies are focused on clinical isolates but there are some papers in which strains from various food samples were analyzed. MRSA strains were identified in milk, cheese, butter, yoghurt, ice cream, *Kashk*, pastry, meat, fish, and ready-to-eat food samples in some Iranian studies (Tables 1-6 and Figures 1-6). Several studies done in different regions of Iran examined the presence of MRSA in milk (Table 1). Many of these MRSA strains were identified using molecular typing techniques. The molecular identification (using polymerase chain reaction (PCR)) of MRSA strains was performed by investigating *mecA* gene. In these studies, antibiotic resistance pattern of isolated MRSA strains was also tested (27-32). In addition, some studies found MRSA strains in milk by phenotypic methods (disk diffusion method) (33, 34).

Based on the literature, so far, MRSA strains were reported in milk samples in 6 provinces and in 4 of them (Chaharmahal and Bakhtiari, East Azerbaijan, Hamadan, and Mazandaran) MRSA strains were found. The MRSA prevalence identified in the different provinces was highly variable (from 0.00% to 13.17%; Figure 1). Chaharmahal and Bakhtiari province showed the highest prevalence (11.51%-13.7%). Nonetheless, MRSA strains were not identified in milk samples from Fars province.

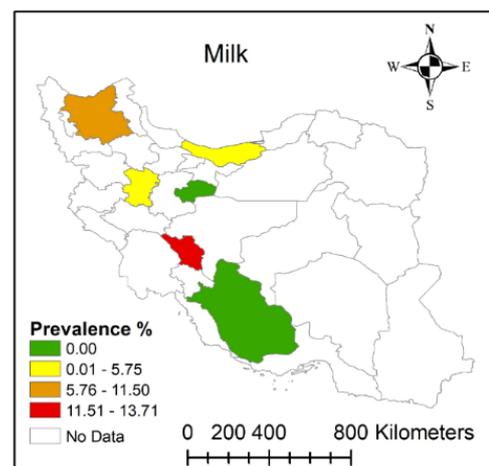


Figure 1. Distribution of methicillin-resistant *Staphylococcus aureus* (MRSA) prevalence in milk samples in Iran during 2006-2020.

Table 1. Reports on the presence of methicillin-resistant *Staphylococcus aureus* (MRSA) in milk samples in Iran during 2006-2020.

Sample type	Location	No. of samples	Milk		Year	Reference
			No. of <i>S. aureus</i> positive samples (Prevalence)	No. of MRSA positive samples (Prevalence)		
Raw cow milk	Chaharmahal and Bakhtiari (Shahr-e Kord)	152	89 (58.55%)	68 (44.73%)	2016	(27)
Raw sheep milk	Chaharmahal and Bakhtiari (Shahr-e Kord)	32	18 (56.25%)	10 (31.25%)	2016	(27)
Raw goat milk	Chaharmahal and Bakhtiari (Shahr-e Kord)	52	16 (30.76%)	11 (21.15%)	2016	(27)
Pasteurized cow milk	Chaharmahal and Bakhtiari (Shahr-e Kord)	65	ND	ND	2016	(27)
Raw milk	Hamadan	131	NS	2 (1.52%)	2013-2014	(28)
Pasteurized milk	East Azerbaijan (Tabriz)	100	1 (1%)	ND	2010	(32)
Raw milk	East Azerbaijan (Tabriz)	100	45 (45%)	14 (14%)	2010	(32)
Raw cow milk	Mazandaran	1035	162 (15.7%)	21 (2.02%)	2006-2013	(29)
Raw sheep milk	Mazandaran	895	86 (9.6%)	11 (1.23%)	2006-2013	(29)
Raw milk	Mazandaran	120	NS	44 (36.67%)	2017-2018	(30)
Raw milk	Hamadan	271	29 (10.7%)	3 (1.107%)	2013-2014	(31)
Raw milk (cow, sheep and goat)	Fars, Chahar Mahal va Bakhtiari and Ghom	348	46 (13.2%)	ND	2010-2011	(33)
Raw milk	NS	100	60 (60%)	31 (31%)	NS	(34)

ND = Not Detected (it means that the referred bacteria was not detected in samples); NS = Not Specified (it means that the referred item was not specified in the article by the author).

Numerous studies evaluated the prevalence of MRSA in cheese samples in different provinces of Iran (Table 2). In most studies, MRSA detection was done using specific *mecA* primers, by PCR method (27, 28, 31, 35-40). The MRSA prevalence was identified in different provinces including Mazandaran, Hamadan, Khorasan Razavi, East Azerbaijan, West Azerbaijan, and Chaharmahal and Bakhtiari (Table 2 and Figure 2). A high variation in MRSA prevalence in cheese samples among different provinces and even in cheese samples from the same province was observed (Table 2). As

shown in Figure 2, the prevalence of MRSA in cheese samples varied from 0.01 to 22.22 with Chaharmahal and Bakhtiari province having the highest prevalence (17.26% -22.22%) and Hamadan having the lowest prevalence (0.01-5.75).

Differences in results of different studies can be attributed to variations in cheese production technologies, number of samples, milk source (raw/pasteurized), and hygiene measures which impact the rate of contamination.

Table 2. Reports on the presence of methicillin-resistant *Staphylococcus aureus* (MRSA) in cheese in Iran during 2006-2020.

Cheese						
Sample type	Location	No. of samples	No. of <i>S. aureus</i> positive samples (Prevalence)	No. of MRSA positive samples (Prevalence)	Year	Reference
Traditional cheese	Mazandaran	360	224 (62.2%)	119 (33.05%)	2016-2017	(36)
Traditional cheese	Mazandaran	450	49 (10.9%)	15 (3.33%)	2006-2013	(29)
Traditional and industrial white cheese	Hamadan	120	19 (15.8%)	4 (3.33%)	2015	(64)
Cheeses (white and feta)	Khorasan Razavi (Mashhad)	100	25 (25%)	8 (8%)	NS	(35)
Traditional cheeses	East Azerbaijan (Tabriz)	100	19 (19%)	9 (9%)	NS	(38)
Traditional cheese	West Azerbaijan (Qotur of khoy)	80	43 (53.57%)	4 (5%)	2011	(65)
Traditional cheese	Chaharmahal and Bakhtiari (Shahr-e Kord)	27	11 (40.74%)	6 (22.22%)	2016	(27)
Traditional cheeses	Azerbaijan	100	16 (16%)	3.36 (3.36%)	2012	(39)
Traditional cheese	West Azarbaijan and East Azarbaijan	73	123** (NS)	19** (15.44%)	NS	(40)
Local cheese	East Azerbaijan	100	22 (22%)	1 (1%)	2017	(37)
Cheese	Hamadan	170	19 (11.2%)	2 (1.17%)	2013-2014	(31)
Cheese	Hamadan	84	NS	1 (1.19%)	2013-2014	(28)

NS = Not Specified (it means that the referred item was not specified in the article by the author); ** Numbers of isolates.

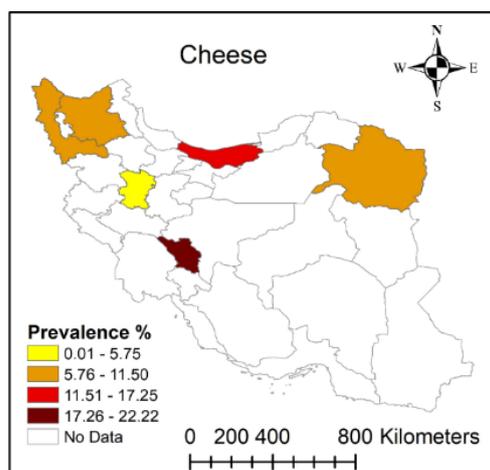


Figure 2. Distribution of methicillin-resistant *Staphylococcus aureus* (MRSA) prevalence in cheese samples in Iran during 2006-2020.

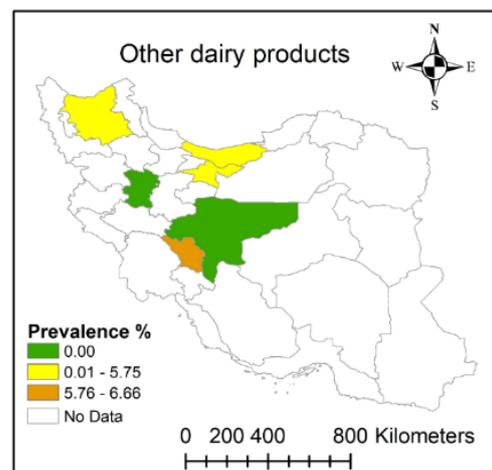


Figure 3. Distribution of methicillin-resistant *Staphylococcus aureus* (MRSA) prevalence in other dairy products in Iran during 2006-2020.

Several studies surveyed the prevalence of MRSA in milk and cheese while few studies considered other dairy products (Table 3). MRSA identification was done through *mecA* gene detection (27, 29, 31, 32, 38, 41, 42). Different dairy products including butter, yoghurt, cream/ top of the milk, ice cream, and *Kashk* were evaluated in Chaharmahal and Bakhtiari, Mazandaran, Hamadan, East Azerbaijan, Tehran, and Isfahan (Table 3). The prevalence of MRSA varied between 0.00 and 6.66 (Figure 3). A high prevalence

of MRSA was reported in Chaharmahal and Bakhtiari while MRSA strains were not detected in different dairy product samples from Hamadan (Figure 3). MRSA strains were identified in ice cream samples in Tehran but were not found in ice cream samples from Chaharmahal and Bakhtiari, Hamadan, and Isfahan. Different sampling methods, seasonal effects, and laboratory techniques could have caused the observed differences in the results among different studies.

Table 3. Reports on the presence of methicillin-resistant *Staphylococcus aureus* (MRSA) in other dairy products in Iran during 2006-2020.

Other Dairy Products						
Sample type	Location	No. of samples	No. of <i>S. aureus</i> positive samples (Prevalence)	No. of MRSA positive samples (Prevalence)	Year	Reference
Traditional butter	Chaharmahal and Bakhtiari (Shahr-e Kord)	30	7 (23.33%)	3 (10%)	2016	(27)
Traditional yoghurt	Chaharmahal and Bakhtiari (Shahr-e Kord)	15	2 (13.33)	1 (6.66%)	2016	(27)
Traditional ice cream	Chaharmahal and Bakhtiari (Shahr-e Kord)	18	6 (33.33%)	ND	2016	(27)
Traditional "Kashk" **	Chaharmahal and Bakhtiari (Shahr-e Kord)	12	2 (16.66%)	1 (8.33%)	2016	(27)
"Kashk"	Mazandaran	270	31 (11.5%)	6 (2.22%)	2006-2013	(29)
Cream	Hamadan	66	4 (6.1%)	ND	2013-2014	(31)
Traditional yoghurt	Hamadan	45	2 (4.4%)	ND	2013-2014	(31)
Traditional cream	Hamadan	47	2 (4.3%)	ND	2013-2014	(31)
Butter	Hamadan	72	16 (8.3%)	ND	2013-2014	(31)
Traditional butter	East Azerbaijan (Tabriz)	150	11 (7.33%)	2 (1.33%)	NS	(38)
Ice cream	East Azerbaijan (Tabriz)	100	23 (23%)	6 (6%)	2010	(32)
Cream	Hamadan	32	NS	ND	2013-2014	(28)
Yoghurt	Hamadan	21	NS	ND	2013-2014	(28)
Top of the milk	Hamadan	22	NS	ND	2013-2014	(28)
Butter	Hamadan	35	NS	ND	2013-2014	(28)
Ice cream	Tehran	241	41 (17%)	2 (0.83%)	2006-2007	(66)
Traditional and commercial dairy products	Isfahan	347	20 (5.8%)	ND	2010-2011	(67)

ND = Not Detected (it means that the referred bacteria was not detected in samples); NS = Not Specified (it means that the referred item was not specified in the article by the author); ** "Kashk" is fermented and dried cow's and/or sheep's milk used in Iranian diets with a salty or sour taste.

Studies on the isolation of MRSA from pastry products are shown in [Table 4](#). These studies reported MRSA in pastry cream in Mazandaran and Tehran provinces and in sweet samples in Hamadan province ([Table 4](#)) as examined by PCR to detect *mecA* (Azizkhani and Tooryan 2018) or *nuc* genes (Fatahi et al. 2018) and disc diffusion method (Soltan Dallal et al. 2008). Prevalence of MRSA in pastry products varied among different regions (0.01%-9.46%; [Figure 4](#)). The highest prevalence of MRSA in pastry products was reported in Hamadan followed by Mazandaran and Tehran provinces ([Table 4](#) and [Figure 4](#)). According to a study conducted in Mazandaran (Amol), remarkable differences in the prevalence of MRSA may be due to the providing optimum temperature for *S. aureus* growth in warm seasons ([36](#)).

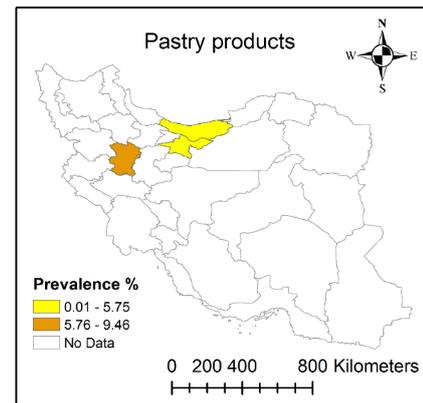


Figure 4. Distribution of methicillin-resistant *Staphylococcus aureus* (MRSA) prevalence in pastry products in Iran during 2006-2020.

Table 4. Reports on the presence of methicillin-resistant *Staphylococcus aureus* (MRSA) in pastry products in Iran during 2006-2020.

Pastry Products						
Sample type	Location	No. of samples	No. of <i>S. aureus</i> positive samples (Prevalence)	No. of MRSA positive samples (Prevalence)	Year	Reference
Pastry cream	Mazandaran (Amol)	360	150 (41.6%)	11 (3.05%)	2016-2017	(36)
Sweet	Hamadan	370	100 (27.02%)	35 (9.46%)	2017-2018	(68)
Pastry cream	Tehran	214	37 (17.3%)	1 (0.46%)	2006-2007	(66)

NS = Not Specified (it means that the referred item was not specified in the article by the author).

Studies that examined meat and meat products for MRSA contamination are shown in [Table 5](#). In these studies, molecular detection of specific target of MRSA, *mecA* gene, was performed for all *S. aureus* strains isolated from meat and meat products in Tehran, Isfahan, and Hamadan provinces ([28, 31, 41-49](#)). The prevalence of MRSA in meat and meat products varied from 0.01% to 19.44% ([Figure 5](#)). Isfahan had the highest prevalence of MRSA (17.26%-19.44%). Prevalence of MRSA in these products varied between 0.01% and 5.75%, while MRSA strains were not identified in meat samples from Hamadan province ([Figure 5](#)). The upper line differences observed among studies may be related to the type of meat processing, human hygiene-related factors, sample preparation, and analysis techniques.

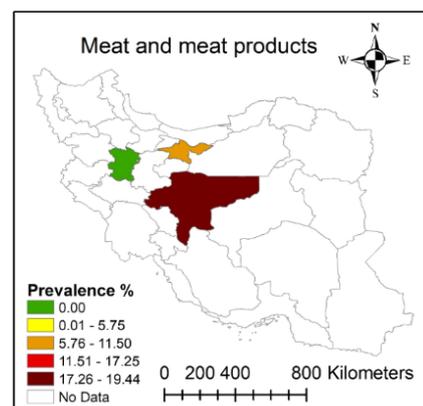


Figure 5. Distribution of methicillin-resistant *Staphylococcus aureus* (MRSA) prevalence in meat and meat products in Iran during 2006-2020.

Table 5. Reports on the presence of methicillin-resistant *Staphylococcus aureus* (MRSA) in meat and meat products in Iran during 2006-2020.

Meats						
Sample type	Location	No. of samples	No. of <i>S. aureus</i> positive samples (Prevalence)	No. of MRSA positive samples (Prevalence)	Year	Reference
Packaged hamburgers	Tehran	256	64 (25%)	58 (22.65%)	2010	(48)
Raw meat (beef, chicken, turkey)	Tehran	131	NS	49 (37.4%)	2016	(47)
Chicken meat	Isfahan	36	NS	25 (69.4%)	2014	(42)
Raw chicken meat	Isfahan	360	82 (22.77%)	68 (18.8%)	2011-2012	(46)
Raw meat (beef, sheep, goat, and camel)	Isfahan	900	NS	160 (17.7%)	2011-2012	(46)
Raw red meat	Hamadan	243	25 (10.3%)	ND	2013-2014	(31)
Raw poultry	Hamadan	136	11 (8.1%)	ND	2013-2014	(31)
Red meat	Hamadan	119	NS	ND	2013-2014	(28)
Chicken meat	Hamadan	66	NS	ND	2013-2014	(28)
Protein food (raw and cooked)	Tehran	481	18 (3.7%)	ND	2006-2007	(66)
Shrimp (fresh and frozen)	Tehran	300	84 (28%)	24 (8%)	2013-2014	(49)
Fish and shrimp (fresh and frozen, marine and farmed)	Tehran	600	206 (34.3%)	49 (8.16%)	2013-2014	(43)

ND = Not Detected (it means that the referred bacteria was not detected in samples); NS = Not Specified (it means that the referred item was not specified in the article by the author).

Studies that examined MRSA in miscellaneous foods are listed in [Table 6](#). Some of these studies tested for the presence of MRSA strains using PCR to detect

mecA gene ([41, 50-54](#)). In a study done in Isfahan, a high prevalence of MRSA strains was found in the restaurant food samples. Cooked chicken samples had

the highest prevalence of MRSA strains. No study reported MRSA strains in raw fish. Low prevalence of MRSA in fish meat samples was attributed to the low ability of *S. aureus* to compete with specific primary bacterial flora of fish (53). In another study, MRSA was most prevalent in meat barbecue, chicken barbecue, soup, and salad followed by raw chicken meat, raw read meat, and rice. Also, MRSA strains were not found in raw fish and grilled fish (50). According to Table 6, the prevalence of MRSA in provinces of Isfahan, Tehran, and Hamadan was within the range of 0.01%-8.30% (Figure 6). The highest prevalence of MRSA was identified in restaurant food in Isfahan province (55) followed by *Samosa* and *Falafel* in Hamadan province (56) and hospital food in Isfahan (57) (Table 6). So, it seems that foods prepared in public places including restaurants, hospitals, and sandwich shops are among the most common sources of foodborne MRSA. As shown in Figure 6, the prevalence of MRSA in samples from Isfahan and

Hamadan provinces was higher (5.76%-8.30%) compared to Tehran province (0.01%-5.75%).

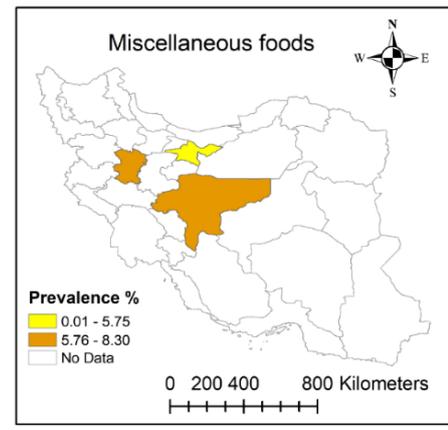


Figure 6. Distribution of methicillin-resistant *Staphylococcus aureus* (MRSA) prevalence in miscellaneous foods in Iran during 2006-2020.

Table 6. Reports on the presence of methicillin-resistant *Staphylococcus aureus* (MRSA) in miscellaneous foods in Iran during 2006-2020.

Miscellaneous Foods						
Sample type	Location	No. of samples	No. of <i>S. aureus</i> positive samples (Prevalence)	No. of MRSA positive samples (Prevalence)	Year	Reference
Ready to eat foods	Isfahan	384	4 (1.042%)	4 (1.04%)	2015	(59)
Restaurant food	Isfahan	580	119 (20.51%)	83 (14.31%)	2015-2016	(53)
Dairy and meat products	Isfahan	139	9 (6.48%)	0.86 (0.61%)	2015-2016	(51)
Hospital food	Isfahan	485	47 (9.69%)	37 (7.62%)	2015-2016	(50)
Hospital food	Tehran	44	2 (4.54%)	1 (2.27%)	NS	(69)
Fruit juice	Tehran	32	3 (9.4%)	ND	2006-2007	(66)
Salad	Tehran	79	1 (1.3%)	ND	2006-2007	(66)
Dairy and meat products	Tehran	913	93 (10.18%)	5 (0.54%)	2010	(54)
<i>Samosa</i> and <i>Falafel</i>	Hamadan	120	56 (46.67%)	10 (8.3%)	2015-2016	(52)

ND = Not Detected (it means that the referred bacteria was not detected in samples); NS = Not Specified (it means that the referred item was not specified in the article by the author).

Presence of VRSA in Various Food Samples

Only one study in Iran has investigated the prevalence of VRSA strains in food samples. Totally, 119 samples of chicken and turkey raw meat were analyzed for the presence of *Staphylococcus aureus*. Based on this report, 29 strains of *S. aureus* were isolated; out of them, 14 (48.5%) strains were found to be resistant to vancomycin as shown by phenotypic

methods. The presence of *vanA* gene using molecular typing techniques (PCR) was identified in chicken and turkey raw meat samples. The prevalence of *vanA* gene in the tested food samples was 43% (6 strains) (58). In some studies, performed in Iran, vancomycin-resistant strains were identified by phenotypic methods but the presence of *van* gene was not studied (32, 36, 38, 48, 53, 59).

Presence of VRE in Various Food Samples

Iranian studies that assessed the prevalence of VRE in different food samples are presented in [Table 7](#). In a study conducted in Tehran province, vancomycin-resistance was most-commonly observed for *E. faecalis* isolates followed by *E. faecium*, *E. gallinarum*, and *E. durans*. The prevalence of five of the *van* (*vanA*, *B*, *C*, *D*, and *E*) genes was investigated by PCR technique. *VanA*, *vanB* and *vanC* genes were the most common phenotypes observed in the isolates, respectively, while *vanD* and *vanE* genes were not identified. The findings of this study suggest that the presence of *Enterococci* in packed and unpacked dried vegetables is due to application of contaminated organic fertilizers and irrigation with wastewater ([45](#)). In another study in Tehran, PCR-based assay showed that all of the VRE isolates were *E. faecium*. All of the isolates were positive for *vanA*, but *vanB* was not detected. Isolates displaying positive results for VRE were observed in meat, chicken, and cheese samples, respectively. The results of this study indicated a high prevalence of VRE in meat and chicken possibly due to extensive use of glycopeptide antibiotics including vancomycin in veterinary practices ([60](#)). In a study performed in Hamadan, VRE strains were found in chicken meat, raw ground beef (14 and 27 strains), milk, and cheese (6 and 1 strains). *VanA*, *vanB*, and *vanC* were detected in VRE isolates ([61](#)). In a study conducted in Lorestan province, *vanC* type was not

identified by PCR ([62](#)). The highest prevalence of VRE was found in Ilam province, *E. faecalis* was recovered from all food samples. Also, all *E. faecalis* isolates showed resistance to vancomycin ([63](#)). As shown in [Figure 7](#), variable prevalence of VRE (0.00% to 100.0%) was reported from different provinces of Iran. Ilam and Lorestan provinces had the highest prevalence of VRE (75.01-100.00). In Tehran, VRE prevalence was 25.01-50.00 and in Hamadan province it was 0.001-25.00. No VRE strain was detected in dairy product samples from Urmia and Tabriz (East Azerbaijan and West Azerbaijan provinces, respectively).

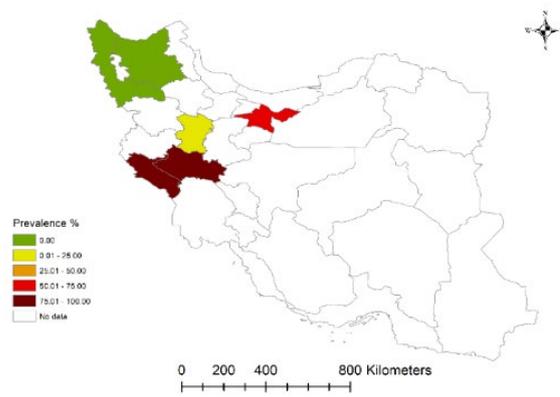


Figure 7. Distribution of vancomycin-resistant *enterococci* (VRE) prevalence in miscellaneous food samples in Iran during 2006-2020.

Table 7. Reports on the presence of vancomycin-resistant *enterococci* (VRE) in miscellaneous food samples in Iran during 2008-2020.

Miscellaneous Food Samples						
Sample type	Location	No. of samples	No. of <i>Enterococcus</i> positive samples (Prevalence)	No. of VRE positive samples (Prevalence)	Year	Reference
Packed and unpacked dried vegetables	Tehran	140	84 (60%)	41 (29.28%)	2015	(45)
Meat, chicken, cheese	Tehran	30	NS	NS	2010	(60)
Dairy products and meat	Hamadan	200	135 (67.5%)	48 (24%)	2012-2014	(61)
Ground meat, raw meat, kebab	Ilam	34	34 (NS)	34 (NS)	NS	(63)
Red meat	Lorestan (Borujerd)	181**	181**	68** (83.95%)	2014-2015	(62)
Traditional cheese	Urmia and Tabriz	50	48 (96%)	ND	2014-2015	(70)

NS = Not Specified (it means that the referred item was not specified in the article by the author); ** Numbers of isolates.

Meta-Analysis Results

Forest plot diagram of the current systematic review and meta-analysis based on overall prevalence of MRSA, VRSA, and VRE in foods are shown in Supplementary file. In addition, meta-regression plot of the current systematic review and meta-analysis in foods based on year of studies were indicated in Supplementary file. Funnel plot and Sensitivity of the current systematic review and meta-analysis in foods were also displayed in Supplementary file. Moreover, the pooled ES of MRSA in milk, cheese, other dairy product, pastry product, meat product, and miscellaneous foods were as follows, respectively: 0.26% (95% CI: 0.19%, 0.34%), 0.25% (95% CI: 0.13%, 0.36%), 0.12% (95% CI: 0.08%, 0.16%), 0.29% (95% CI: 0.15%, 0.42%), 0.21% (95% CI: 0.13%, 0.29%), and 0.11% (95% CI: 0.06%, 0.17%) (Supplementary file). Accordingly, the highest pooled ES of MRSA was for pastry products and the lowest pooled ES of MRSA was for miscellaneous foods. Furthermore, the pooled ES of VRE in miscellaneous foods was 0.80% (95% CI: 0.62%, 0.98%) (Supplementary file).

5. Conclusion

Antimicrobial resistance is one of the most important threats to the public health and food safety and security. Since food plays a critical role in development of antimicrobial resistance, identification of food sources and geographical distribution patterns is necessary to control and manage antimicrobial resistance. This review demonstrated that antimicrobial resistance varies among different foodstuffs. Such disparities can be attributed to differences in food handling practices, hygiene practices during processing, traditional food-processing and preparation methods, geographical area and use of

antimicrobials in plant and animal agriculture. Based on the published reports, MRSA, VRSA and VRE strains are widely present in different types of raw and processed meat, raw milk, traditional cheese, restaurant, and hospital foods. Of note, preventing excessive use of antimicrobials can be helpful in guaranteeing the product safety from farm to fork. Good agricultural practices, preventive controls, surveillance system in agriculture, and livestock farming sectors, improved on-farm prevention strategies, sufficient knowledge about MRSA, VRSA, and VRE transmission routes to the food chain, and determination of high-risk sources of food contamination and the prevalence of mentioned microorganisms in plant- and animal-originated foods are highly needed specially in hot spot regions.

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Authors declare that there is no conflict of interest in this work.

Authors' Contribution

The authors confirm contribution to the paper as follows: study conception and design: A.A. and M.H.; data collection: R.R. and B.K.; interpretation of results: S.G.H., S.M., and A.A.; writing the manuscripts: A.A., S.M; meta-analysis performance: MA.S.

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Conflict of Interest

None.

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