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Molecular study on methicillin-resistant *Staphylococcus aureus* strains isolated from dogs and associated personnel in Jordan

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ABSTRACT

Objective: To determine the prevalence, genetic relatedness, and pattern of antimicrobial susceptibility in methicillin-resistant *Staphylococcus aureus* (*S. aureus*) (MRSA) isolated from household dogs, farm dogs, and stray dogs, compared to isolates from their associated personnel.

Methods: MRSA was isolated from 250 nasal swabs (150 swabs from dogs and 100 swabs from humans). PCR assays were used to detect the presence of both the *nuc* and *mecA* genes, which confirmed the identity of *S. aureus* isolates and the presence of methicillin resistance, respectively. Disk diffusion was used to determine the antibiotic susceptibility against 15 antimicrobial agents along with an *E*-test that determined the minimum inhibitory concentration for oxacillin. Pulsed field gel electrophoresis was conducted to determine the genetic relatedness of MRSA isolates from dogs to those from associated and unassociated personnel.

Results: The prevalence of *S. aureus* in dogs and humans was 12.7% and 10.0% respectively, while the prevalence of MRSA isolates in dogs and humans was 5.3% and 5.0%, respectively. The prevalence of MRSA isolates in household dogs, farm dogs, and stray dogs was 7.8%, 4.7%, and 0.0%, respectively. MRSA isolates demonstrated a significantly higher rate of multi-resistance against three or more antimicrobial agents than methicillin-susceptible *S. aureus* (MSSA). Trimethoprim-sulphamethoxazole and chloramphenicol were the most effective antibiotics against all MRSA isolates. Pulsed field gel electrophoresis revealed a strong association between dog MRSA isolates and MRSA isolates from strongly associated personnel.

Conclusions: MRSA is prevalent in house dogs, as well as in dog rearing centers and among their strongly associated personnel. A strong association was found between the MRSA isolates from dogs and those from humans who are in close contact. In addition, MRSA isolates showed a high rate of multi-resistance compared to MSSA isolates.

1. Introduction

Methicillin-resistant *Staphylococcus aureus* (*S. aureus*) (MRSA) is a major health care-associated pathogen worldwide and has increased in incidence dramatically over the last decade [1,2]. Companion animals have been implicated more frequently as potential reservoirs of MRSA than other livestock [3,4]. In several studies, a 0%–4% prevalence rate of MRSA in dogs has been reported [5–7]. Other reports demonstrated MRSA at a higher prevalence (~9%) in pets and veterinary staff [8,9], and the nasal carriage of MRSA plays a key role in the epidemiology and pathogenesis of community-associated infections [10,11].

In Jordan, MRSA is widely prevalent in Jordanian hospitals and represents a serious public health problem. The nasal carriage rate of *S. aureus* among the Jordanian healthy young population was 40%, and 19% of the nasal *S. aureus*, and 57% of clinical isolates were resistant to oxacillin [12]. A retrospective study conducted at King Abdullah University Hospital in North Jordan showed that 152 *S. aureus* isolates collected from different infections revealed that the overall rate of MRSA

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was 34%, with a rate of 57%–70% in the adult intensive care unit [13]. To our knowledge, no studies on MRSA in companion animals in Jordan were found in the literature that demonstrated its prevalence and health hazard. Therefore, this study was conducted to document the prevalence of MRSA in dogs and their associated personnel, as well as to determine their genetic relatedness and antimicrobial resistant profile.

2. Materials and methods

2.1. Sample collection, transportation, and preparation

In total, 250 nasal swabs were collected from 150 dogs and 100 humans during a period between March and the end of October 2009. The 150 dog samples were collected from household dogs, stray dogs, and farm dogs from the middle and northern parts of Jordan. The total numbers of household dogs, stray dogs, and farm dogs were 77, 30, and 43, respectively, as illustrated in Table 1.

The 100 human nasal swab samples were collected from personnel strongly associated with dogs (5 from dog owners, 50 from employees who feed, take care of, treat, and train dogs daily at the Spana Welfare Center, Humane Center for Animal Welfare, and K-9 Center, 25 from intermediately associated personnel, including veterinarians working in clinics and veterinary students, and 20 from unassociated personnel who have never been in contact with dogs). A sterile cotton swab moistened with normal saline was inserted into the nares and gently rotated to make contact with the nasal septum. For dogs, smaller swabs were inserted to a distance of about 0.5–1.0 cm. All swabs were placed in a transport medium and stored at 4 °C until cultured within 6-h collection at the Microbiology Research Laboratory, Faculty of Veterinary Medicine, Jordan University of Science and Technology.

2.2. Isolation and identification of S. aureus

All nasal swabs were cultured on mannitol salt agar (Oxoid, UK) and incubated aerobically at 37 °C for 24–48 h. The cultures were then examined for the presence of *S. aureus* (yellow colonies) and for a microscopic appearance after Gram staining. The presumptive *S. aureus* isolates were further examined for pigments and coagulase production, by using the tube method [14].

Table 1
Distribution of the dogs' nasal swab samples according to the dogs' locations and type of rearing system in Jordan.

Middle zone	Household dogs	Stray dogs	Farm dogs*
Amman governorates			
Swelieh city	5	4	6
Jaweh town	0	5	5
Sahaab town	0	6	7
Spana Welfare Center	13	0	0
Dogs Police K-9 Center	27	0	0
Humane Center for Animal Welfare	14	0	0
Al-Zarga governorates	0	4	8
Northern zone			
Ramtha dogs Police K-9 Center	18	0	0
Jarash governorates	0	4	6
Ajlune governorates	0	3	4
Irbid governorates	0	4	7
Total	77	30	43

*: Dogs kept with sheep and goat flocks.

2.3. Molecular identification of S. aureus isolates

2.3.1. DNA extraction and identification of the nuc gene

The extraction protocol was done according to the Wizard genomic DNA purification kit (Promega cooperation, Technical manual genomic DNA purification part TM0580, Madson, USA). Then, presumptive *S. aureus* isolates were tested by PCR amplification of the *nuc* gene [15]. PCR amplification was conducted at a final volume of 25 μL [12.5 μL of Go Taq master mix (Promega, USA), 5 μL (2.5 pmol) of each primer F (GCGATTGATGGTGA TACGGTT) as well as R (AGCCAAGCCTTGACGAACT AAAGC), 2.5 μL of a bacterial DNA sample and 5 μL nuclease free water]. The PCR amplification was conducted as follows: 5 min at 94 °C, 35 cycles for 30 s at 94 °C, 45 s at a corresponding annealing temperature of 55 °C, and 45 s at 72 °C, and a final extension of 10 min at 72 °C. The PCR products were observed on 1.5% agarose gels.

2.4. Identification of MRSA by E-test and antimicrobial susceptibility testing

MRSA isolates were identified by *E*-test (Oxoid, UK), which is a gradient antibiotic stabilized on a plastic strip with 30 graduations to provide an accurate minimum inhibitory concentration (MIC) over a range of 256–0.015 μg/mL. This test was conducted for oxacillin only, according to the manufacturers' instructions (Oxoid, UK) and guidelines. Mueller-Hinton agar (Difco, Detroit, MI, USA) supplemented with 2% NaCl was used for this purpose [16]. Samples for the *E*-test were prepared according to Clinical and Laboratory Standards Institute [17]. Isolates showed MICs equaled to or greater than 4 μg/mL, which were considered MRSAs [18].

The agar disk diffusion susceptibility test of 15 antimicrobials [cefoxitin (10 μg), penicillin (10 IU), cephalexin (30 μg), kanamycin (30 μg), gentamicin (10 μg), tobramycin (10 μg), amikacin (30 μg), ciprofloxacin (5 μg), azithromycin (15 μg), erythromycin (15 μg), tetracycline (15 μg), amoxicillin-clavulanic acid (20/10 μg), trimethoprim-sulphamethoxazole (1.25/23.75 μg), nalidixic (30 μg), and chloramphenicol (30 μg)] was carried out by using the Clinical and Laboratory Standard Institute guidelines [19]. The *S. aureus* ATCC 25923 strain was used as a control.

2.5. Molecular identification of MRSA

For further confirmation, MRSA isolates were tested by PCR amplification of the *mecA* gene. PCR amplification was conducted at a final volume of 25 μL [12.5 μL of Go Taq master mix (Promega, USA), 5 μL (2.5 pmol) of each primer F (5′-GCA ATC GCT AAA GAA CTA AG) as well as R (5′-GGG ACC AAC ATA ACC TAA TA) [20], 2.5 μL of a bacterial DNA template and 5 μL nuclease free water]. PCR amplification was conducted as follows: denaturation at one cycle of 94 °C for 3 min, 30 cycles at 94 °C for 45 s, annealing at 53 °C for 2 min, extension at 72 °C for 30 s, and a final extension at 72 °C for 5 min. The PCR products were observed by electrophoresis on 1.5% agarose gels (Nusieve Bioproducts, Maine, USA).

2.6. Pulsed field gel electrophoresis (PFGE)

PFGE was used to study the genetic relatedness between the *S. aureus* isolates from dogs and human *S. aureus* isolates. PFGE was performed according to a Canadian standard protocol

by using the restriction enzyme *Sma*I [21]. Electrophoresis was done with a CHEF-DR III apparatus (Bio-Rad, USA) by using the auto-algorithm program and switch times of 5.3–34.9 for 18 h at a 6.0 V/cm gradient and 14 °C in 0.5 μL Tris-Borate-EDTA. The gel was stained for 20 min with 0.5 mg of ethidium bromide per liter and destained with Millipore-filtered H_2O for at least 30 min with water changes for three times. PFGE gels were viewed under ultraviolet light.

2.7. Data analysis

Gels were photographed and digitized with the FOTO/Analyst Archiver system, and an analysis with BioNumerics version 2.0 software was conducted (Applied Maths, Belgium). The *S. aureus* ATCC 43300 and ATCC 25923 strains were used as reference standards for MRSA and methicillin-susceptible *S. aureus* (MSSA), respectively. A Wilcoxon test (paired) was used to compare the antibiotic susceptibility of MRSA and MSSA isolates against different antimicrobial agents, and it was also used to compare the MICs of MRSA and MSSA isolates against oxacillin.

3. Results

3.1. Conventional and molecular identification of *S. aureus*

In total, 46 isolates were confirmed as *Staphylococci*; among them, 29 isolates (19 from dogs and 10 from humans) were conventionally identified as *S. aureus* and confirmed by amplifying the thermonuclease gene (*nuc*) using PCR (Figure 1). The prevalence rate of *S. aureus* in dogs was 12.7% (19/150) and in humans, it was 10% (10/100).

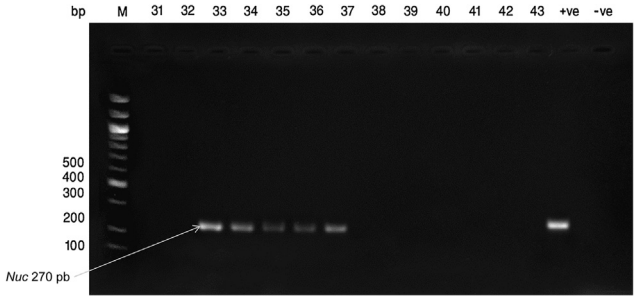

Figure 1. Agarose gel electrophoresis of PCR products (*nuc* gene). Lane M: DNA molecular marker (100 bp); Lane +ve: Positive control (*S. aureus* ATCC 25923); Lane -ve: Negative control (water); Lanes 33, 34, 35, 36, 37: Positive samples showing the 270 bp gene product; Lanes 31, 32, 38, 39, 40, 41, 42, 43: Negative samples.

3.2. MRSA and susceptibility test

Out of 29 *S. aureus* strains, 13 were MRSA; among them, 8 were from dogs (prevalence rate was 5.3%) and 5 were from humans (prevalence rate was 5.0%). Six MRSA isolates were from 77 household dogs [one from each of the Swelieh city, Spana Welfare Center, Humane Center for Animal Welfare, and K-9 Center/Ramtha and two from the K-9 Center/Amman (prevalence rate was 7.8%)]. The other two MRSA isolates were isolated from 43 farm dogs from the Al-Zarqa and Irbid governorates (prevalence rate was 4.7%). No MRSA was isolated from stray dogs. One human MRSA isolate was from a dog owner whose dog had MRSA, and the other four MRSA isolates were from strongly associated personnel (two from K-9, one from Spana Welfare Center, and one from Humane Center for Animal Welfare).

All MRSA isolates were found to be resistant to cefoxitin. In contrast, all MRSA isolates were susceptible to trimethoprim-sulphamethoxazole and chloramphenicol. MRSA was also found to be susceptible to azithromycin, cephalexin, aminoglycoside, kanamycin, gentamicin, tobramycin, and amikacin in a range of 62%–85%. The antibiotics resistant to all MRSA and MSSA isolates are shown in Table 2, and the MRSA isolates were found to be significantly more resistant to the tested antibiotic than MSSA isolates ($P < 0.001$). In addition, the MRSA isolates of humans were significantly more resistant than the MRSA isolates of dogs against erythromycin, tetracycline, and amoxicillin-clavulanic acid (Table 3).

3.3. MIC

An *E*-test was used to evaluate the MIC of oxacillin. Out of 29 *S. aureus* isolates, all MRSA (13) isolates showed oxacillin resistance (oxacillin MIC $\geq$ 4 μg/mL).

In addition, MRSA MICs were significantly higher than MSSA MICs in both humans and dogs ($P \leq 0.001$), and MRSA

Table 2
Antibiotic resistance pattern of 13 methicillin resistant and 16 susceptible *S. aureus* isolates. *n* (%).

Antibiotics	MRSA[r]			MSSA[s]		
	R	I	S	R	I	S
Cefoxitin	13 (100)	0	0	1 (6)	1 (6)	14 (88)
Penicillin	12 (92)	1 (8)	0	3 (19)	2 (13)	11 (69)
Cephalexin	3 (23)	1 (7)	9 (70)	1 (6)	1 (6)	14 (88)
Kanamycin	3 (23)	0	10 (77)	2 (13)	2 (13)	12 (75)
Gentamicin	3 (23)	0	10 (77)	1 (6)	1 (6)	14 (88)
Tobramycin	3 (23)	1 (7)	9 (70)	1 (6)	1 (6)	14 (88)
Amikacin	3 (23)	2 (15)	8 (62)	2 (13)	2 (13)	12 (75)
Ciprofloxacin	3 (23)	0	10 (77)	0	2 (13)	14 (88)
Azithromycin	2 (15)	0	11 (85)	0	1 (6)	15 (94)
Erythromycin	7 (54)	1 (7)	5 (39)	2 (13)	2 (13)	12 (75)
Tetracycline	5 (39)	2 (15)	6 (46)	2 (13)	3 (19)	11 (69)
Amoxicillin-clavulanic acid	6 (46)	0	7 (54)	2 (13)	0	14 (88)
Trimethoprim-sulphamethoxazole	0	0	13 (100)	0	0	16 (100)
Nalidixic	9 (69)	1 (7)	3 (23)	3 (19)	2 (13)	11 (69)
Chloramphenicol	0	0	13 (100)	1 (6)	1 (6)	14 (88)

[r,s]: MRSA are significantly resistant to the tested antibiotic than MSSA ($P < 0.001$); R: Resistant; I: Intermediate; S: Susceptible.

Table 3
Comparison of antibiotic resistance pattern of 8 MRSA isolates from dogs and 5 MRSA from humans. *n* (%).

Antibiotics	MRSA in dogs			MRSA in humans		
	R	I	S	R	I	S
Cefoxitin	8 (100)	0	0	5 (100)	0	0
Penicillin	5 (63)	1 (12)	2 (25)	3 (60)	0	2 (40)
Cephalexin	2 (25)	0	6 (75)	1 (20)	1 (20)	3 (60)
Kanamycin	3 (37)	0	6 (75)	2 (40)	0	3 (60)
Gentamicin	1 (12)	0	7 (88)	2 (40)	0	3 (60)
Tobramycin	2 (25)	1 (12)	5 (63)	1 (20)	0	4 (80)
Amikacin	0	2 (25)	6 (75)	2 (40)	0	3 (60)
Ciprofloxacin	0	0	8 (100)	3 (60)	0	2 (40)
Azithromycin	1 (12)	0	7 (88)	1 (20)	0	8 (80)
Erythromycin	3 (37)^{S}	0	5 (67)	4 (80)^{S}	1 (20)	0
Tetracycline	2 (25)^{S}	1 (12)	5 (67)	3 (60)^{S}	1 (20)	1 (20)
Amoxicillin-clavulanic acid	2 (25)^{S}	0	6 (75)	3 (60)^{S}	0	2 (40)
Trimethoprim-sulphamethoxazole	0	0	8 (100)	0	0	5 (100)
Nalidixic	5 (63)	1 (12)	2 (25)	4 (80)	1 (20)	0
Chloramphenicol	0	0	8 (100)	0	0	5 (100)

S: Significant difference ($P < 0.001$); R: Resistant; I: Intermediate; S: Susceptible.

Table 4
MIC of MRSA and MSSA isolates in dogs and humans against oxacillin by *E*-test. μg/mL.

Sample code	MRSA^{r} MICs	Sample code	MSSA^{r} MICs
D8	8^{r1}	D14	0.250
D9	6	D19	0.500
D20	6	D23	2.000
D58	16	D28	0.012
D71	6	D53	1.000
D82	8	D61	1.000
D143	12	D66	0.060
D147	16	D92	1.000
H34	8^{r1}	D103	0.500
H37	32	D109	0.120
H92	48	D137	0.030
H15	8	D84	2.000
H13	6	H25	0.500
		H67	0.120
		H73	1.000
		H76	0.500

^{r}: MRSA MICs are significantly higher than MSSA MICs in both human and dogs ($P < 0.001$); ^{r1}: MRSA MICs are significantly higher in humans than MRSA MICs in dogs ($P < 0.001$).

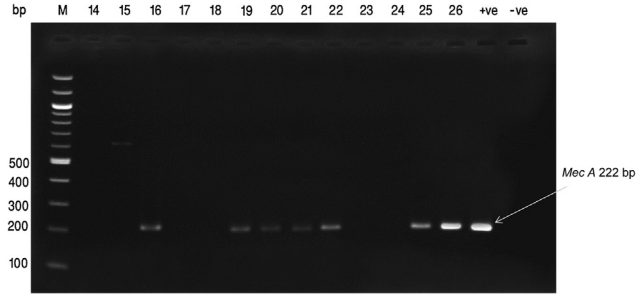

Figure 2. Agarose gel electrophoresis of PCR products (*mecA* gene).
Lane M: DNA molecular marker (100 bp); Lane +ve: Positive control (*S. aureus* ATCC 43300); Lane -ve: Negative control (*S. aureus* ATCC 25923); Lanes 16, 19, 20, 21, 22, 25, 26: Positive samples showing the 222 bp gene product; Lanes 14, 15, 17, 18, 23, 24: Negative samples.

MICs were significantly higher in humans than MRSA MICs in dogs ($P \leq 0.001$), as shown in Table 4.

3.4. PCR for the identification of the mecA gene

The results of PCR showed that 13 (45%) of 29 *S. aureus* isolates were positive for the presence of the *mecA* gene (Figure 2). Five MRSA isolates were from humans and eight were from dogs.

3.5. PFGE

The *Sma*I macrorestriction fragment profiles of 29 *S. aureus* isolates were determined by PFGE (Figure 3). A dendrogram of a percent similarity and DNA relatedness were calculated based on the Dice coefficient, which revealed five major clusters of isolates: Jordan A, Jordan B, Jordan C, Jordan D, and Jordan E (Figure 4). All isolates are grouped into the above-mentioned five clusters, as shown in Table 5.

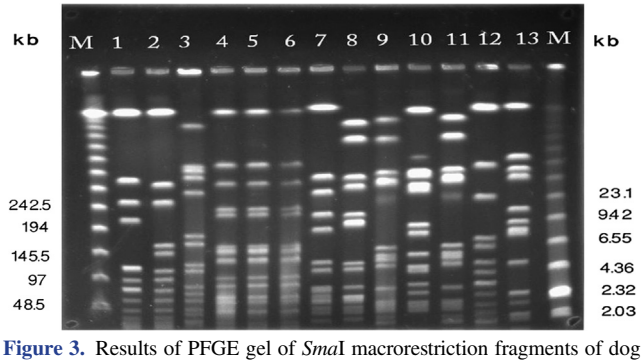

Figure 3. Results of PFGE gel of *Sma*I macrorestriction fragments of dog and human *S. aureus* isolates.
Lane M: DNA molecular marker (100 bp); Lane 1: Positive control of MRSA (*S. aureus* ATCC 43300); Lane 2: Positive control of MSSA (*S. aureus* ATCC 25923): Lanes 3 to 13: *S. aureus* isolates; M: Molecular sizes.

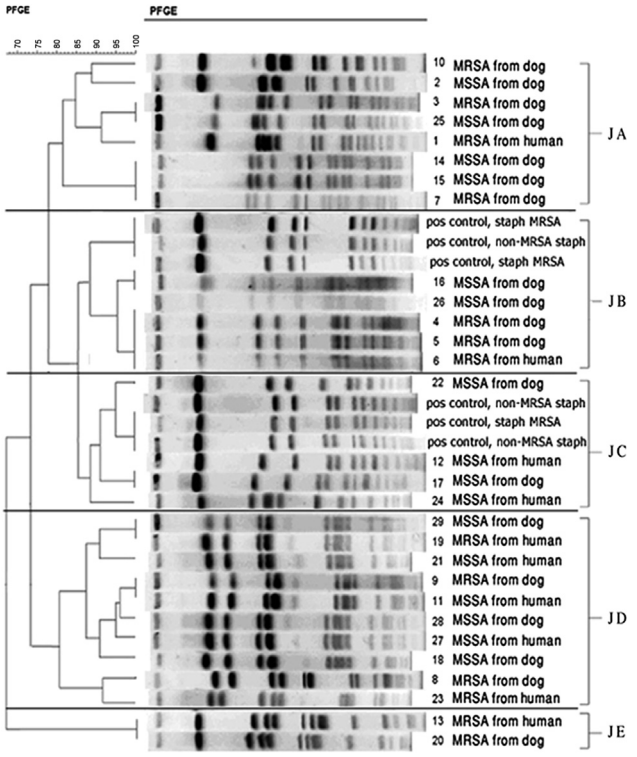

Figure 4. A PFGE dendogram of 29 *S. aureus* isolates collected from humans and dogs in Jordan with positive control of MRSA and non-MRSA.
JA: Jordan A cluster; JB: Jordan B cluster; JC: Jordan C cluster; JD: Jordan D cluster; JE: Jordan E cluster.

Table 5
Groups of the 29 *S. aureus* isolates in different clusters determined by PFGE dendogram.

Clusters	*S. aureus* groups			
	MRSA/dogs	MRSA/humans	MSSA/dogs	MSSA/humans
Jordan A	3	1	4	0
Jordan B	2	1	2	0
Jordan C	0	0	2	2
Jordan D	2	2	3	3
Jordan E	1	1	0	0

4. Discussion

In this study, the prevalence of the *S. aureus* nasal carriage isolates in dogs was 12.7% and of MRSA, it was 5.3%. To the best of our knowledge, this is the first report about the nasal carriage of *S. aureus* and MRSA in dogs in Jordan. The *S. aureus* nasal carriage of dogs in Jordan is more prevalent than 8.8% which was reported in a Hong Kong study [22], similar to the other study in Austria [23]. The human nasal carriage rate was 10%, which is similar to other reports in Jordan [11], but it is not in agreement with others [12], which reported a 40% nasal carriage rate in Jordan. However, the variable in the nasal carriage of *S. aureus* may be different due to populations, geographical locations, and the influence of genetic and environmental factors [10]. In addition, the cell-wall lipoteichoic acid, hormonal status, and antimicrobial activity of nasal secretions play a role [24].

In this study, the prevalence rate of MRSA in dogs was 5.3% and most dogs carrying MRSA are either household dogs or reared at rearing centers (prevalence rate was 7.9%), where they are exposed daily to close contact with their personal associates. In contrast, stray dogs did not have MRSA, and this could be explained by that those dogs are not exposed to the human population and do not receive veterinary care. Two MRSA isolates were from farm dogs and a study may be needed to demonstrate the prevalence of MRSA in farm animals in Jordan. The MRSA prevalence in dogs was similar to other studies [7,8,25].

The MRSA nasal carriage rate in humans was 5%, and that represents 50% of the 10 *S. aureus* strains isolated from 100 human nasal swabs, which may be explained by daily close contact with dogs that transmit MRSA between owners and personnel strongly associated with dogs easily. It may also be due to the continued use of antibiotics [9,22,26]. The close contact between household pets and humans offers favorable conditions for the transmission of MRSA by direct contact (petting, licking, and physical injuries) or through the domestic environment (contamination of food, water, and plates) and physical contact with dogs, as well as through contact with household environments contaminated by pets (floors, furniture, and carpets) [27].

In the current study, the oxacillin resistance in human isolates was significantly higher than that in dog isolates. This may indicate the unwise use of antibiotics in humans, wherein people obtain antibiotics without a prescription. In other countries, this is not always the case, as a higher oxacillin resistance in canine isolates compared to that in human isolates was reported [22].

The present investigation also demonstrated MRSA isolates were significantly more resistant to the tested antibiotics than those of MSSA. However, all MRSA isolates were susceptible to trimethoprim-sulphamethoxazole and chloramphenicol. The results of the current work showed similar findings to that of Al-Zu'bi *et al.* [12], where all nasal isolates were susceptible to chloramphenicol, while clinical isolates showed some resistance.

In the current study, the MRSA isolates showed 92% resistance to penicillin, which is similar to another study in Jordan that showed all human MRSA isolates were resistant to penicillin and 23% of MRSA isolates were resistant to cephalexin, which is not in agreement with other reports [11,28]. MRSA in the current study showed 23% resistance to aminoglycoside, gentamicin, kanamycin, tobramycin, and amikacin, in contrast to the studies of others where higher resistance to kanamycin (98%), tobramycin (97%), and amikacin (89%) was reported [11,28,29]. In total, 77% and 23% of MRSA isolates were susceptible to ciprofloxacin and nalidixic acid, respectively, which is not in agreement with other reports that demonstrated 100% and 93% of MRSA isolates were susceptible to ciprofloxacin and nalidixic acid, respectively [9,11]. The MRSA in the current work revealed a 54% resistance to erythromycin, which is in agreement with other reports [11,28,30]. The MRSA susceptibility to tetracycline was 46%, which is not in agreement with other studies that reported all MRSA isolates were susceptible to tetracycline [9,11]. The lowest resistance was observed against azithromycin (15%) and 46% of MRSA isolates were resistant to amoxicillin-clavulanic acid, which is in contrast with another study in Korea, wherein 97% and 98% of MRSA isolates were resistant to azithromycin and amoxicillin-clavulanic acid, respectively [29]. Gentamicin, ciprofloxacin, azithromycin, amoxicillin-clavulanic acid, and tetracycline are human medicines, and the emergence of strains

resistant to these antibiotics demonstrates the potential public health risk of MRSA.

The resistance patterns of MSSA are significantly less than those of MRSA ($P < 0.001$), except for trimethoprim-sulphamethoxazole, which is similar. All MSSA isolates were susceptible to all antimicrobial agents tested in this study in a range of 69%–100%, and this is in agreement with another study wherein multi-drug resistance was found to be less common amongst MSSA isolates [30]. In the current study, the MSSA isolates showed no resistance to ciprofloxacin, azithromycin, or trimethoprim-sulphamethoxazole, followed by aminoglycoside and chloramphenicol. These results are similar to the findings of another report in India and are in contrast to the results of a UK study that stated all MSSA isolates were susceptible to gentamicin and tetracycline [30,31].

The discrepancies of MRSA antibiotic susceptibility results may be due to the use of different methods for susceptibility testing, different breakpoints for the evaluation of the results, and the misuse of antimicrobial agents, resulting in microbial fitness [27].

Two of five human MRSA isolates showed significantly higher oxacillin MICs than those in dog MRSA isolates (Table 4). This may be due to the high and uncontrolled use of antimicrobial agents in human medicines in Jordan. In addition, the authors' observations indicated that dogs included in this study were less exposed to antimicrobial treatment. In case of MRSA with oxacillin MIC $\geq$8 μg/mL, multiple-drug resistance was observed.

PFGE revealed five major clusters designated as pulsed-field types: Jordan A, Jordan B, Jordan C, Jordan D, and Jordan E. These clusters showed a percentage of similarities among different isolates. Isolates in clusters Jordan A, Jordan B, Jordan D, and Jordan E showed that the genetic relatedness between dog and human MRSA isolates were as follows. In cluster Jordan A, the relatedness between MRSA isolates from dogs and those from humans was 80%–90%, and in clusters Jordan B and Jordan E, the relatedness was 100%. In cluster Jordan D, the relatedness was 80%–90%. This may be because most human and dog MRSA isolates were collected from the same rearing centers where dogs and personnel are in daily close contact for months or even years. MRSA isolates in the Jordan E cluster are from one dog and his owner in Swelieh city. These results agree with other study findings that MRSA isolate relatedness is high when dogs and humans share the same place [32]. The MRSA isolates from either dogs or humans shared mostly similar antibiotic resistance patterns, and this agreed with another study [32]. Based on these data, we assume that the dogs are colonized with the same strain or clone as their owners or strongly associated personnel. This is in agreement with other reports about the possibility of MRSA transmission between dogs and humans [3,9,32,33]. The current study concludes and confirms the presence of MRSA in the nasal cavities of dogs, dog owners, and personnel strongly associated with dogs in Jordan, and that MRSA is highly prevalent in household dogs rather than in stray dogs. A genetic relatedness among the MRSA isolates of dogs and those of humans was demonstrated, suggesting a strong possibility of MRSA transmission between them, and MRSA isolates showed a high rate of multi-resistance compared to MSSA isolates.

Conflict of interest statement

We declare that we have no conflict of interest.

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