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Association of *tsst-1* and *pvl* with *mecA* Genes among Clinical *Staphylococcus aureus* Isolates from a Tertiary Care Hospital

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Abstract

To investigate the association of both *tsst-1* and *pvl* with *mecA* genes in clinical *Staphylococcus aureus (S. aureus)* strains obtained from a tertiary care hospital, Mansoura-Egypt. Besides, relating these genes to antibiotic resistance patterns in such isolates. A prospective study was performed over clinical samples obtained from patients showing evidence of infection at Mansoura University Children Hospital, Egypt. *Staphylococcus aureus* isolates were obtained to test their antibiotic sensitivity pattern by disk diffusion method, E-test strips besides oxacillin agar screen test followed by screening such isolates for *mecA, pvl* and *tsst-1* genes by polymerase chain reaction. Out of 180 isolated *S. aureus* strains, 88 isolates (48.9%) were methicillin resistant. The *tsst-1* gene was positive in 65.9% of methicillin resistant *S. aureus* (MRSA) and 55.4% of methicillin sensitive *S. aureus* (MSSA) strains. Carriage of *mecA* and *tsst-1* genes was not significantly associated (P=0.69). The *pvl* gene was positive in 23.9% of MRSA and 9.8% of MSSA isolates showing a significant association with *mecA* gene (P=0.04*). The *mecA* gene was significantly associated with multidrug resistant isolates (p= 0.041*). There was no significant association between antibiotic resistance and the presence of *tsst-1* or *pvl* genes. The *tsst-1* gene had a high prevalence among *S. aureus* isolates and it was similarly distributed in both MRSA and MSSA groups. The *pvl* gene was significantly associated with MRSA group. Antibiotic resistance pattern of *S. aureus* was not significantly associated to the presence of *tsst-1* or *pvl* genes in any way.

Keywords: *Staphylococcus aureus*, MRSA, *mecA* gene, *pvl* gene, *tsst-1* gene, PCR.

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INTRODUCTION

Staphylococcus aureus (S. aureus) is a major bacterial human pathogen held responsible for both community-acquired as well as hospital-acquired related infections[1], that might cause a wide variety of clinical manifestations including sepsis, respiratory tract, surgical site and urinary tract infections[2]. *Staphylococcus aureus* infection remains a tough challenge to manage due to the emergence of multi-drug resistant strains that led to drag its clinical importance under the spot light over the past decades[3]. Methicillin resistant *S. aureus* (MRSA) strains show resistance to a large group of beta-lactam antibiotics including penicillins and cephalosporins[4]. The MRSA strains produce an altered penicillin-binding protein-2, referred to as PBP2a encoded by *mecA* gene[5], such protein has more resistance to beta-lactam antibiotics when compared to the original PBP2[6].

Many virulence factors are responsible for the unique pathogenicity of *S. aureus* such as toxins, enzymes and adhesion factors. The toxins produced by *S. aureus* include Panton-Valentine leukocidin (PVL), hemolysin, toxic shock syndrome toxin-1 (TSST-1), exfoliative toxins and staphylococcal enterotoxins[7]. The PVL is a pore forming leukotoxinen coded into by *pvl* gene and has the ability to target and kill host leukocytes through perforation of plasma membrane together with intracellular organelle membranes. The PVL producing *S. aureus* strains can cause various infections ranging from as simple as infections of skin and soft tissue up to serious life endangering conditions such as severe necrotic pneumonia[8,9].

Nevertheless, another toxin known as TSST-1 encoded by *tsst-1* gene is also produced by *S. aureus* strains[10], and its expression can lead to an acute life threatening toxic shock syndrome (TSS) that can present itself with rash, fever and multi-organ dysfunction[11]. A strong relation was found between the TSS and tampon use by women due to toxin released by colonizing *S. aureus* strains in the vagina. However, the TSS can also occur in other different conditions such as surgical site complications, skin and respiratory tract infections[12]. Having said that, the secretion of TSST-1 into blood also increases the rate of TSS-like neonatal exanthematous lesion, sudden infant death syndrome and Kawasaki syndrome in pediatric age group[11]. The improper timing of prescribing the necessary antimicrobial treatment for patients with TSS may lead to fatal shock within hours after the onset of clinical presentations[12]. Therefore, to improve the outcome of such patients it is extremely important to have sufficient data about the *tsst-1* gene prevalence and its relation to antibiotic susceptibility pattern.

In literature, many studies have reported an alarming increase in the MRSA strains positive for *tsst-1* and *pvl* genes[7,13]. Similarly, previous studies conducted in Egypt detected *tsst-1* and *pvl* genes among MRSA strains[14,15]. Toxins encoded by *tsst-1* and *pvl* genes can cause severe infections such as TSS and necrotic pneumonia. Treatment of such infections is even more challenging when caused by MRSA because of the *mecA* gene- associated antimicrobial resistance. Therefore, investigating the prevalence of these toxins-encoding genes among MRSA isolates in different geographical locations is necessary for proper treatment and control of these infections. Moreover, these data will enable us to set up a proper antibiotic policy to improve the outcome of these cases. In our study, we investigated the association of both *tsst-1* and *pvl* with *mecA* genes among clinical *S. aureus* isolates obtained from a tertiary health care center, Mansoura-Egypt. In addition, the relation of these genes to antibiotic resistance patterns was evaluated.

MATERIALS and METHODS

This prospective study was undertaken at the Medical Microbiology and Immunology Department, Faculty of Medicine, Mansoura University-Egypt. The protocol was revised and proved to be fulfilling ethical criteria by our institutional review board.

Sample Collection

Various clinical samples were collected from inpatients and outpatients showing evidence of infection at Mansoura University Children Hospital, Mansoura-Egypt from January to October 2018. Suggestive signs and symptoms differed according to the site of infection such as fever, dysuria, suprapubic tenderness, dyspnea, tachypnea, purulent sputum, wound pain or tenderness, purulent wound drainage, purulent eye secretions, etc. Samples included blood, urine, endotracheal aspirates, wound drainages, sputum, eye secretions and abscesses. Enrolled

samples were sent to the Medical Microbiology and Immunology laboratory for processing.

Microbiological Processing

All samples were processed by standard microbiological techniques. Resulting *S. aureus* isolates were fully identified by colonial morphology, Gram-staining and biochemical testing including catalase, coagulase and mannitol fermentation. API Staph (bioMerieux, France) was also performed in order to confirm the identification of *S. aureus* isolates.

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility pattern of *S. aureus* isolates was tested by disk diffusion method on Mueller-Hinton agar (MHA) using the following antibiotic disks (Oxoid, UK): penicillin (10 units), cefazolin (30mg), ciprofloxacin (5mg), erythromycin (15mg), clindamycin (2mg), Trimethoprim-sulfamethoxazole (1.25/23.75mg), gentamicin (10mg), linezolid (30mg) and tetracycline (30mg). Disk diffusion procedure was carried out according to the guidelines of clinical and laboratory standard institute (CLSI)[16]. Determination of the minimum inhibitory concentration for vancomycin was achieved by E-test strips (bioMerieux, France) on MHA plates following the manufacturer's guidelines. The breakpoints for resistance as implemented by the CLSI were used for results interpretation[16].

Detection of Methicillin Resistance

Methicillin resistance in previously identified *S. aureus* isolates was phenotypically detected by cefoxitin disk diffusion and oxacillin agar screen tests following the CLSI guidelines. Cefoxitin disk diffusion test was conducted by standard disk diffusion procedure using cefoxitin (30mg) as a surrogate for oxacillin[16]. As recommended by the CLSI, oxacillin agar screen test was conducted using MHA containing 4% NaCl and 6mg/mL oxacillin. A half McFarland suspension was prepared from the *S. aureus* isolates. Then, a sterile swab was dipped in the prepared bacterial suspension. After expressing the excess liquid, an entire quadrant of the MHA plate was streaked to be incubated at 35°C for 24 hours and then examined with transmitted light. Growth of > 1 colony on this culture medium indicated methicillin resistance[16]. *Staphylococcus aureus* ATCC 29213 was used as negative control (methicillin sensitive) while *S. aureus* ATCC 33591 was used as a positive control (methicillin resistant). Isolates were then kept in tryptic soy broth supplemented by 30% glycerol at -70°C until further molecular testing. Being resistant to at least 3 or more antibiotic classes in addition to beta-lactams, define multidrug resistance[17].

DNA Extraction and Polymerase Chain Reaction

Extraction of Genomic DNA was conducted by using an available commercial Qia -AmpDNA Mini Kit (QIAGEN-UK) through applying the manufacturer's guidelines. For detection of *mecA, pvl* and *tsst-1* genes among the isolated *S. aureus* strains, polymerase chain reaction (PCR) was performed using specific primers as shown in Table 1 [18]. The PCR was performed in a final 50mL volume reaction containing Taq PCR Master Mix (QIAGEN-UK), template DNA and specific primers.

PCR identification of the *mecA* gene was performed with the thermo-cycling conditions set at 5 min at 94°C for initial denaturation, followed by 40 amplification cycles: 94°C for 30s, 57°C for 45 s, 72°C for 30 s then final extension at 72°C for 5 min [18]. Multiplex PCR for detection of *pvl* and *tsst-1* genes was performed with the following cycling settings: 5 min at 94°C for initial denaturation followed by 40 cycles of amplification of successive

Table 1. Primers used in the study

Gene	Primer	Sequence (5'→3')	Product size (bp)
mecA	F	AGAAGATGGTATGTGGAAGTTAG	583
	R	ATGTATGTGCGATTGTATTGC	
pvl	F	GGAAACATTTATTCTGGCTATAC	502
	R	CTGGATTGAAGTTACCTCTGG	
tsst-1	F	TTATCGTAAGCCCTTTGTTG	398
	R	TAAAGGTAGTTCTATTGGAGTAGG	

steps at 94°C for 40 s, 60°C for 40 s and 72°C for 1 min with a final extension at 72°C for 5 min [18]. To determine the presence of the desired genes, PCR products were visualized by electrophoresis in agarose gel stained with ethidium bromide followed by UV lights examination. Reference strains of *S. aureus* ATCC 33591, *S. aureus* ATCC 49775 and *S. aureus* ATCC BAA-2094 were used as positive control for *mecA, pvl* and *tsst*-1 genes respectively while distilled water was used as the negative control.

Statistical Analysis

Chi-square test was applied to compare variables and the results were presented as percentages (%). When P value was <0.05 the difference was statistically significant. We used SPSS 22 for Windows (SPSS Inc., Chicago, IL, USA).

RESULTS

A total of 180 *S. aureus* strains were isolated from the processed 375 clinical samples (48%) [88 Male (48.9%) and 92 Females (52.1%)], the mean± SD age was 6.1 ±2.8 years. Out of the *S. aureus* isolates, 75 (41.7%), 43 (23.9%) and 19 (10.6%) were recovered from blood, urine and endotracheal aspirates respectively as demonstrated in Table 2. Phenotypic evaluation of methicillin resistance by cefoxitin disk diffusion and oxacillin agar screen tests revealed that 88 out of 180 (48.9%) *S. aureus* isolates were methicillin resistant. The *mecA* gene was further identified in all of these isolates that confirmed their primary identification as MRSA by phenotypic methods.

In the current study, we determined the distribution of *mecA, tsst-1* and *pvl* genes among the recovered *S. aureus* strains by PCR. Out of the 180 isolated *S. aureus* strains, 88 (48.9%), 109 (60.6%) and 30 (16.7%) isolates had the *mecA, tsst-1* and *pvl* genes respectively (Table 3). Fig. 1 demonstrated a representative example of multiplex PCR for detection of *pvl* and *tsst-1* genes.

Table 2. Different sources of *Staphylococcus aureus* isolates

Sample	Number of isolates (%)
Blood	75 (41.7%)
Urine	43 (23.9%)
Endotracheal aspirates	19 (10.6%)
Wound drainages	13 (7.2%)
Sputum	12 (6.7%)
Eye secretions	11 (6.1%)
Abscesses	7 (3.9%)
Total	180 (100%)

Table 3. Distribution of *mecA*, *tsst-1* and *pvl* genes among *Staphylococcus aureus* isolates

Gene	*Staphylococcus aureus* isolates (No=180)
mecA positive	88 (48.9%)
mecA negative	92 (51.1%)
tsst-1 positive	109 (60.6%)
tsst-1 negative	71 (39.4%)
pvl positive	30 (16.7%)
pvl negative	150 (83.3%)

Values are expressed as No (%)

Out of 88 isolated MRSA strains, 37 (42.0%) were positive only for the *tsst-1* gene and 21 (23.9%) were positive for both *tsst-1* and *pvl* genes. Among the 92 methicillin sensitive *S. aureus* (MSSA) isolates,44 (47.8%) were positive only for the *tsst-1* gene, 2 (2.2%) were positive only for the *pvl* gene and 7 (7.6%) were positive for both genes as demonstrated in Table 4. Totally, the *tsst-1* gene was detected in 65.9% (58/88) of MRSA and 55.4% (51/92) of MSSA strains. No significant association was found between the carriage of *mecA* and *tsst-1* genes (P=0.69). Regarding the *pvl* gene, it was found in 23.9% (21/88) of MRSA and 9.8% (9/92) of MSSA isolates. The *pvl* gene was significantly associated with the presence of *mecA* gene (P=0.04).

Among the 88 *mecA* gene positive *S. aureus* isolates, 39(44.3%), 20(22.7%) and 8(9.1%) were recovered from blood, urine and endotracheal aspirates respectively. Of the 109 *tsst-1* gene positive *S. aureus* strains, 41 (37.6%) were isolated from blood and 29 (26.6%) from urine. As regard the 30 *pvl* positive *S. aureus* isolates, 11(36.7%) and 7 (23.3%) were obtained from blood and urine respectively. No significant association was found between the source of clinical sample and the presence of *mecA, tsst-1* or *pvl* genes (Table 5).

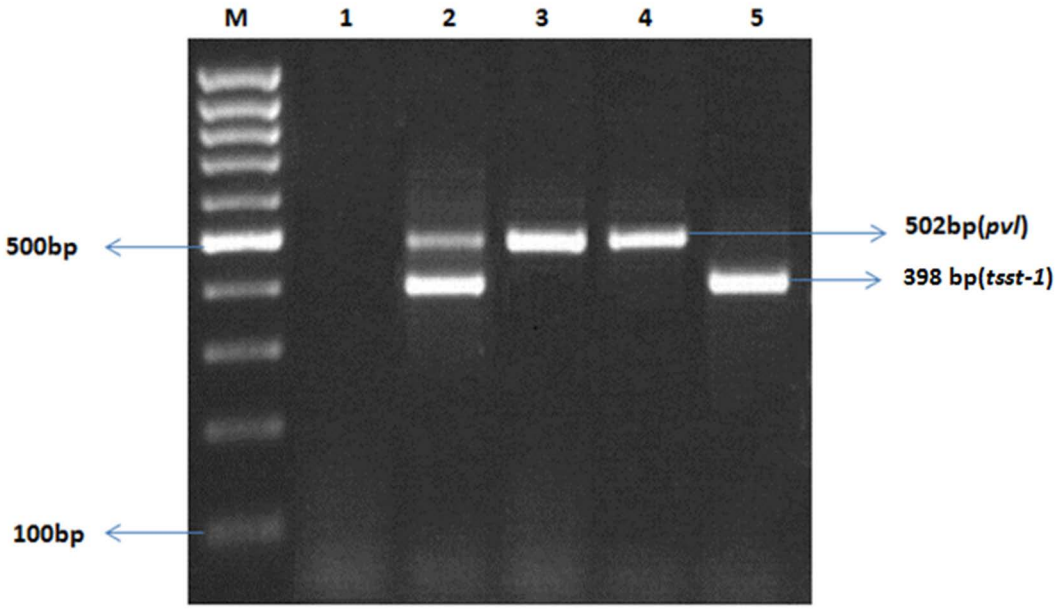

Fig. 1. Multiplex PCR for *pvl* (502 bp) and *tsst-1* (398 bp) genes. M: Marker (100 bp); 1: Negative control, 2-3: Test isolates, 4: Positive control for *pvl* gene, 5: Positive control for *tsst-1* gene

In the present study, we determined the antibiotic resistance patterns of the recovered *S. aureus* isolates. Of the 180 *S. aureus* isolates, 108 (60.0%) were resistant to penicillin, 105 (58.3%) to cefazolin, 93 (51.7%) to tetracycline, 79 (43.9%) to ciprofloxacin, 63 (35.0%) to erythromycin, 59 (32.8%) to gentamicin, 62 (34.4%) to clindamycin and 45 (25.0%) to trimethoprim-sulfamethoxazole. The highest resistance observed was to penicillin while all isolates were sensitive to vancomycin and linezolid. Antibiotic resistance patterns in relation to the existence or absence of *mecA*, *tsst-1* and *pvl* genes were given in Table 6.

Resistance patterns within MRSA strains was found to penicillin (100%), cefazolin (100%) followed by tetracycline (65.9%), erythromycin (48.9%), clindamycin (48.9%), ciprofloxacin (47.7%), gentamicin (45.5%) and trimethoprim-sulfamethoxazole (34.1%). Apart from ciprofloxacin, linezolid and vancomycin, resistance to evaluated antibiotics were significantly higher with the presence of *mecA* gene. Furthermore, the existence of *mecA* gene was significantly associated with multidrug resistant isolates (P=

Table 4. Distribution of *tsst-1* and *pvl* genes among methicillin resistant and methicillin sensitive *Staphylococcus aureus* isolates

Gene	MRSA isolates (*mecA* positive)	MSSA isolates (*mecA* negative)	*Staphylococcus aureus* isolates
Only *tsst-1* positive	37 (42.0%)	44 (47.8%)	81 (45.0%)
Only *pvl* positive	0	2 (2.2%)	2 (1.1%)
Both *tsst-1* and *pvl* positive	21 (23.9%)	7 (7.6%)	28 (15.6%)
Both *tsst-1* and *pvl* negative	30 (34.1%)	39 (42.4%)	69 (38.3%)
Total	88 (100%)	92 (100%)	180 (100%)

Values are expressed as No (%); MRSA= Methicillin resistant *Staphylococcus aureus;*
MSSA= Methicillin sensitive *Staphylococcus aureus*

Table 5. Distribution of *mecA, tsst-1* and *pvl* genes among *Staphylococcus aureus* isolates recovered from different clinical samples

Sample	*mecA* gene			*tsst-1* gene			*pvl* gene		
	Positive	Negative	P value	Positive	Negative	P value	Positive	Negative	P value
Blood	39 (44.3)	36 (39.1)	0.72	41 (37.6)	34 (47.9)	0.56	11 (36.7)	64 (42.7)	0.68
Urine	20 (22.7)	23 (25.0)	0.88	29 (26.6)	14 (19.7)	0.68	7 (23.3)	36 (24.0)	0.89
Endotracheal aspirates	8 (9.1)	11 (12.0)	0.49	13 (11.9)	6 (8.5)	0.72	4 (13.3)	15 (10.0)	0.69
Wound drainages	6 (6.8)	7 (7.6)	0.76	9 (8.3)	4 (5.6)	0.64	3 (10.0)	10 (6.7)	0.43
Sputum	5 (5.7)	7 (7.6)	0.66	7 (6.4)	5 (7.0)	0.65	2 (6.7)	10 (6.7)	1.0
Eye secretions	6 (6.8)	5 (5.4)	0.59	6 (5.5)	5 (7.0)	0.57	2 (6.7)	9 (6.0)	0.89
Abscesses	4 (4.5)	3 (3.3)	0.77	4 (3.7)	3 (4.2)	0.69	1 (3.3)	6 (4.0)	0.71
Total	88	92		109	71		30	150	

Values are expressed as No (%)

Table 6. Relation of *mecA*, *tsst-1* and *pvl* genes to antibiotic resistance patterns of *Staphylococcus aureus* isolates

Antibiotic resistance	*mecA* gene			*tsst-1* gene			*pvl* gene		
	Positive	Negative	P value	Positive	Negative	P value	Positive	Negative	P value
Penicillin	88 (100)	20 (21.7)	0.012 *	68 (62.4)	40 (56.3)	0.60	21 (70.0)	87 (58.0)	0.59
Cefazolin	88 (100)	17 (18.5)	0.001 *	63 (57.8)	42 (59.2)	0.88	21 (70.0)	84 (56.0)	0.48
Gentamicin	40 (45.5)	19 (20.7)	0.048 *	41 (37.6)	18 (25.4)	0.08	9 (30.0)	50 (33.3)	0.66
Erythromycin	43 (48.9)	20 (21.7)	0.039 *	41 (37.6)	22 (31.0)	0.49	12 (40.0)	51 (34.0)	0.58
Tetracycline	58 (65.9)	35 (38.0)	0.04 *	55 (50.5)	38 (53.5)	0.51	14 (46.7)	79 (52.7)	0.7
Ciprofloxacin	42 (47.7)	37 (40.2)	0.62	52 (47.7)	27 (38.0)	0.22	16 (53.3)	63 (42.0)	0.42
Clindamycin	43 (48.9)	19 (20.7)	0.046 *	40 (36.7)	22 (31.0)	0.47	11 (36.7)	51 (34.0)	0.79
Trimethoprim-sulfamethoxazole	30 (34.1)	15 (16.3)	0.044 *	30 (27.5)	15 (21.1)	0.37	8 (26.7)	37 (24.7)	0.65
Linezolid	0	0	-	0	0	-	0	0	-
Vancomycin	0	0	-	0	0	-	0	0	-
Total	88	92		109	71		30	150	

Values are expressed as No (%); *Statistically significant

0.041). No significant association was detected between antibiotic resistance and the presence of *tsst-1* or *pvl* genes (Table 6).

DISCUSSION

Staphylococcus aureus is increasingly recognized globally as a common and serious cause of various community and healthcare associated infections with a significant increase in not only the morbidity but also in mortality. They are known producers of different types of toxins and other virulence factors that shape up their clinical significance. Molecular studies have shown that some MRSA strains that carry *tsst-1* and *pvl* genes[7] have the ability to produce toxins encoded by these genes responsible for severe infections such as TSS and necrotic pneumonia with a mortality rate up to 75%[19]. However, treating such infections is considered a challenge to physicians in view of their antimicrobial resistant properties. Therefore, having knowledge about virulence factors of *S. aureus* and local prevalence of MRSA is beneficial for control and treatment of *S. aureus* infections.

In our study, out of the 180 *S. aureus* strains investigated, a predominance of 88 (48.9%) were found to be MRSA, similar to the results reported by Barakat and his colleagues who conducted a study in Egypt with a 45.3% of *S. aureus* isolates were MRSA[20]. Nevertheless, the reported prevalence of MRSA has varied widely dependent on location and the socioeconomic status of the region. The MRSA prevalence was found to be as low as 34% in European countries[21,22], 18% in Russia[23] and 41.6% in USA[24]. On the other hand, studies from developing countries have reported MRSA prevalence of as high as 72% in Lebanon[25], 87.6% in Iran[26] and 80% in China[27] which is considerably higher than our findings. We have reported that the source of 44.3% of MRSA isolates was the blood, going through the same stream with other studies who reported that MRSA strains were responsible for majority of bloodstream infections among hospitalized patients[28,29].

In the current study, phenotypic detection of methicillin resistance correlated well with genotypic detection of *mecA* gene. However, Motamedi and his colleagues have reported that only 30% of the phenotypically identified as MRSA isolates had the *mecA* gene[30], that was attributed to different mechanisms of methicillin resistance other than *mecA* gene. Nevertheless, our results have shown that methicillin resistance is primarily mediated by *mecA* gene among the studied population.

In our study, the multiplex PCR has detected *tsst-1* gene in 60.6% of the recovered *S. aureus* isolates, while Xie and his colleagues have reported that *tsst-1* gene prevalence of 48% which was lower than our findings[31]. However, other studies have reported lower prevalence of *tsst-1* gene as less as 20%[24, 32-35]. This variation could be attributed to the geographical location and also to different methodology encountered in these studies as their detection of the *tsst-1* gene was carried out at the protein level only[32,33]. The expression of *tsst-1* gene does not grant the production of the toxin. Therefore, despite the presence of the gene, the toxin might not been expressed among the tested group which could explain the differences in the reported *tsst-1* gene prevalence.

We reported that the *tsst-1* gene existed in 65.9% of MRSA and 55.4% of MSSA strains. The presence of *tsst-1* gene was not significantly associated with either MRSA or MSSA groups. This was concordant with reports from Zarei Koosha and his colleagues who detected *tsst-1* gene among 69.8% of MRSA isolates[26]. Other studies have also reported that up to 58% of MRSA strains were *tsst-1* gene positive[33,36,37]. Similar to our findings, the prevalence of *tsst-1* gene was reported up to 56% among the MSSA isolates in other studies[24-26, 38].

In our study, we found that 16.7% of isolated *S. aureus* strains were positive for the *pvl* gene, putting in consideration that in previous studies, the prevalence of *pvl* gene has shown a wide variation ranged from 2% up to 38.9% [39,40]. Moreover, we have detected the *pvl* gene in 23.9% of MRSA and 9.8% of MSSA isolates. This was similar to the findings of Goudarzi and his colleagues, who reported the prevalence of *pvl* gene of 21.4% within MRSA isolates[37], and Abiri and his colleagues who detected *pvl* gene among 21.5% of *mecA* positive MRSA strains[7]. We found that *pvl* gene was significantly prevalent in MRSA group (P=0.04). Nevertheless, these findings were contrary to what been reported by Motamedi and

his colleagues, that *mecA* and *pvl* genes did not show any association[30].

In our study, antimicrobial susceptibility patterns of *S. aureus* isolates have shown higher rate of resistance to many antimicrobials that are routinely prescribed on daily bases in clinical practice for case of staphylococcal infections with the highest resistance observed toward penicillin as previously reported in other studies[23,24]. Such findings highlight the urge for the availability of an accurate and geographically targeted data about local antibiotic susceptibility patterns for appropriate antibiotic selection. This measure will hinder the rapid spreading of antimicrobial resistant *S. aureus* strains and it will give a solution for major challenge faced by healthcare providers in their clinical practice while treating infections caused by such isolates.

We found that 35.0% of *S. aureus* isolates were resistant to erythromycin which was similar to previous reports from Iran (51.3%)[26] and China (97.8%)[33], others have found the resistance to erythromycin was as low as 0%[23]. On the other hand, the prevalence of tetracyclin resistant strains was 51.7% close to the 48% reported by Tokajian and his colleagues in Lebanon[25]. Gentamicin resistance was found to be 32.8% that is nearly similar to the reports from China (28.1%)[24] and higher than the reports from Russia (19%)[23]. In accordance with other reports, none of our isolates demonstrated resistance to vancomycin or linezolid[24,25]. The reported MRSA resistance patterns in our work were in line with the findings of Goudarzi and his colleagues[36].

It has been shown that *S. aureus* strains isolated from different locations had different virulence genetic profiles. Therefore, the variations between other studies and our findings in terms of prevalence of the studied genes and antimicrobial resistance patterns might be attributed to different geographical locations. Nevertheless, different study design, successful execution of infection control measures and implementation of properly designed antibiotic stewardship policy might offer another explanation as well. Moreover, the variations in antimicrobial resistance reported from different regions emphasize the importance of tailoring antibiotic policy based on local antibiotic sensitivity patterns and international guidelines. To the best of our knowledge, this is the first research conducted in our locality to find the association of both *tsst-1* and *pvl* genes with *mecA* gene in *S. aureus* isolates and to study their effect over the antibiotic susceptibility pattern of such isolates.

CONCLUSION

The *pvl* and *tsst-1* genes positive *S. aureus* strains could lead to severe infections with high morbidity and mortality. Therefore, frequent monitoring of these strains and their virulence factors are substantially important. We reported a high prevalence of *tsst-1* gene among *S. aureus* isolates. The *tsst-1* gene was not significantly associated with either MRSA or MSSA groups. On the other hand, the *pvl* gene was significantly associated with MRSA group. The isolated *S. aureus* strains demonstrated elevated antibiotic resistance to many of the routinely used antimicrobials and the *mecA* gene was significantly associated with multidrug resistant isolates. Hence, implementation of antibiotic stewardship policy based on local antibiotic resistance patterns would help the physicians in selecting the choice antibiotics for their patients.

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CONFLICTS OF INTEREST

The authors declare that there are no conflicts of interest.

AUTHORS' CONTRIBUTION

AS developed the concept and designed the study. AS and YN did sample processing. AS collected and analysed data and did the interpretation of results. YN carried out protocol development and contributed in designing and writing of the methodological part of the article. AS wrote the manuscript. AS and YN did critical revision of the article. AS reviewed and approved the manuscript.

FUNDING

None.

DATA AVAILABILITY

All datasets generated or analyzed during this study are included in the manuscript.

ETHICS STATEMENT

All procedures performed in this study were in accordance with the ethical standards of the institutional research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Informed consent was obtained from at least one of parents of participants included in the study. The study protocol has been approved by Faculty of Medicine - Mansoura University ethical committee.

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