

## Prevalence of Methicillin Resistant Staphylococcus Aureus (MRSA) and Antimicrobial Susceptibility Patterns at a Private Hospital in Sana'a, Yemen.

**Background:** Methicillin Resistant Staphylococcus Aureus (MRSA) strains are common causes of nosocomial infections and are associated with increased morbidity and mortality. In addition, the antibiotic resistance for MRSA is a major concern in clinical practice.

**Objective:** To study the prevalence and the antimicrobial susceptibility pattern of MRSA isolates from patients in a private hospital.

**Methods:** The study was performed at a private hospital in Sana'a, Yemen. All the patients' samples from January, 2017 to December, 2017 were included. All isolates from inpatients' clinical specimens (mainly respiratory secretion, pus, urine, and blood) were collected and standard isolation procedures were applied to all the samples. The records were taken from the microbiology department. Antibiotic susceptibility testing was done by Kirby Bauer's Disc diffusion technique, following the national committee for clinical laboratory standards. The antimicrobial susceptibility patterns of all the MRSA strains were studied against several antibiotics.

**Results:** A total of 2079 samples were gathered during the study period. Among them, 199 strains of *staphylococcus aureus* were isolated. A majority of *staphylococcus aureus* isolates were from pus specimen (n=81/199; 40.7%). MRSA prevalence among the patients was 17.6 % (n=35/199; 17.6). Highest proportion of MRSA was in ICU and surgical departments about 29% of all MRSA isolates. A majority of MRSA isolates were from sputum specimen (n=18/35; 51.4%)

The study findings showed that MRSA isolates had the highest frequency of resistant (100%) to levofloxacin and amoxicillin/clavulanic acid, followed by ciprofloxacin 97%, gentamicin and ampicillin/ sulbactam 94 %, cefuroxime 91 %, moxifloxacin 76%, erythromycin 71.5%, clindamycin 70.5%, and imipenem 55%. The highest frequency of sensitivity (100%) was observed with linezolid, vancomycin, and trimethoprim/sulfamethoxazole.

**Conclusion:** MRSA isolates were highly susceptible to newer drugs such as linezolid and to vancomycin which is not a commonly prescribed drug due to the higher nephrotoxic antibiotic. A more careful monitoring for use of broad-spectrum antibiotics should be instituted.

**Keywords:** MRSA, Prevalence, Antimicrobial susceptibility, Vancomycin.

### 1. Introduction:

*Staphylococcus aureus* is one of the most important pathogens affecting humans. Penicillin is the drug of choice to treat infections caused by *Staphylococcus aureus*. Because of the penicillin was commonly prescribed drugs, the prevalence of penicillin resistant strains increased, especially in hospitals<sup>(1)</sup>. MRSA was first described in 1961. It was reported after one year of introduction of methicillin. Recently, it was defined as one of the most important nosocomial pathogens. New studies suggest that the infection due to MRSA is not only hospital-acquired but community acquired as well<sup>(2)</sup>. Also MRSA has become a major public health problem worldwide<sup>(3)</sup>. Some large outbreaks of MRSA have been documented from different regions of the world, where it had caused severe infections including septicemia, endocarditis and meningitis<sup>(4)</sup>.

A study by Dickinson in England and Wales has approved an increase in the death rate of MRSA infection<sup>(5)</sup>. The risk factors that increase in MRSA prevalence are antibiotics abuse, prolonged hospitalization, intravascular instrumentation and hospitalization in an intensive care unit. There is significant variation in prevalence of clinical infections among units, hospitals and countries<sup>(4)</sup>. MRSA is of serious concern because of resistance to many other antimicrobials that are commonly used in hospitals. Another important element was that emergence of resistance to vancomycin, although at a low level has been documented<sup>(6)</sup>. Significantly, the Netherlands and countries in the Scandinavian region, had maintained low rates of MRSA infection due to strict infection control practices<sup>(7)</sup>. The problem of MRSA continues to increase and the rising colonization rates lead to the increasing of infection rates in the public and in hospitals. This can increase hospital stays and greater costs of the health care system<sup>(8)</sup>.

Appropriate selection of empiric antibiotics for infections, depending on local circulating pathogens, can lead to good patient outcomes. So, epidemiologic information collected through ongoing surveillance is important to support clinicians and infection control committees to prevent and treat infection<sup>(7)</sup>. The hospitals of different sizes were facing the problem of MRSA. In addition, the problem of MRSA appears to be increasing regardless the size of hospital and its control measures for MRSA<sup>(8)</sup>. The aim of the present study is the detection of MRSA from various clinical departments at a local private hospital in Sana'a, Yemen and determining their susceptibility to some antimicrobial agents.

## **2. Method:**

The current retrospective study based on electronic laboratory records of *Staphylococcus aureus* isolates and MRSA isolates from clinical specimens analyzed at microbiology laboratory of university of sciences and technology (UST) hospital in Sana'a, Yemen.

The records were taken from the microbiology department. Staphylococcus strains were identified based on Gram's stain morphology, colony characteristics, and biochemical identification tests. All isolates were identified as *S. aureus* according to standard methods<sup>(9)</sup>. A total of 2079 isolates were collected from the hospitalized patients in hospital of UST from January 2017 to September 2017. These isolates were obtained through conventional clinically oriented ordered cultures. From 2079 isolates, a total of 199 isolates were *S. aureus* strains. Methicillin resistance was determined using methicillin screening.

All isolates were from inpatients' clinical specimens (mainly respiratory secretion, pus, urine, and blood) and Standard isolation procedures were applied to all the samples.

Antibiotic susceptibility testing was done by Kirby Bauer's Disc diffusion technique, following Clinical & Laboratory Standards Institute (CLSI)<sup>(9)</sup>. The antimicrobial susceptibility patterns of all the MRSA strains were determined against the following antibiotics: vancomycin, linezolid, imipenem, levofloxacin, ciprofloxacin, Co-Trimoxzole, Erythromycin, amoxicillin/ clavulanic Acid, cefuroxime, doxycycline, clindamycin, ampicillin/ sulbactam, gentamicin, moxifloxacin, and Lincomycin.

Full ethical clearance was obtained from the qualified authorities who approved the study design. All data were analyzed using SPSS Statistics 21. Data was presented in tables and graphs. All data were analyzed using SPSS Statistics 21. Data was presented in tables and graphs.

#### 4. Results:

The figure 1 showed highest numbers of staphylococcus aureus were obtained from pus (n=81/199; 40.7%) and least from urine (n=11/199; 5.5%). In this study, Overall MRSA prevalence was 17.6 % (n=35/199; 17.6).

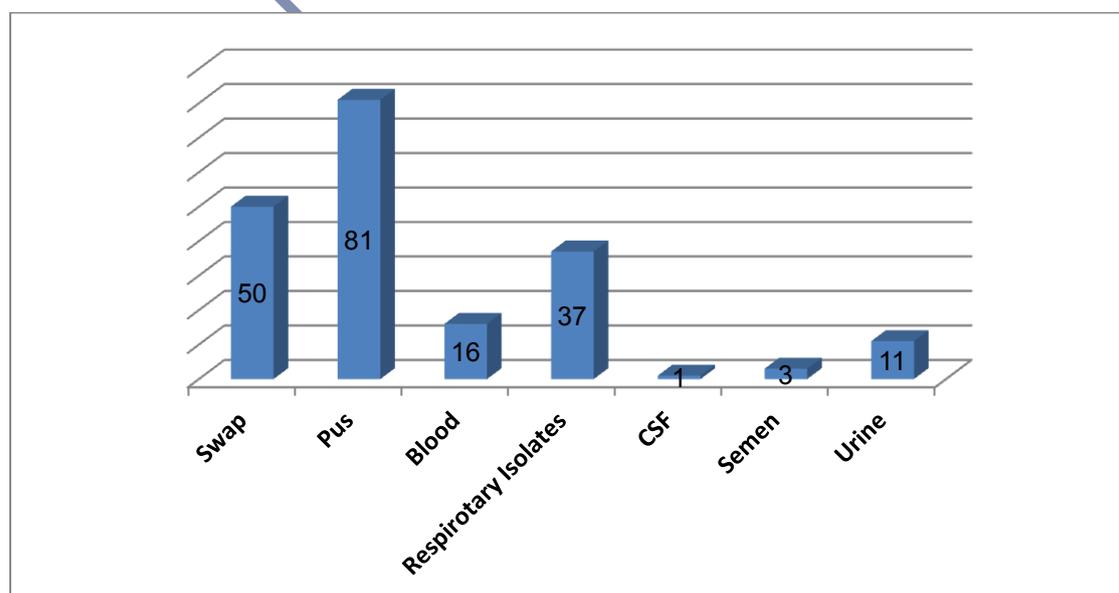


Figure 1. Distribution of *S. aureus* according to sample types

The study results showed that the most of the MRSA isolates prevalence in age group between 46 to 60 years about 12(34%), followed by the age more than 60 years in second rank about 8 (23%), and finally the age between 1 to 15 or years only about 2(6%).

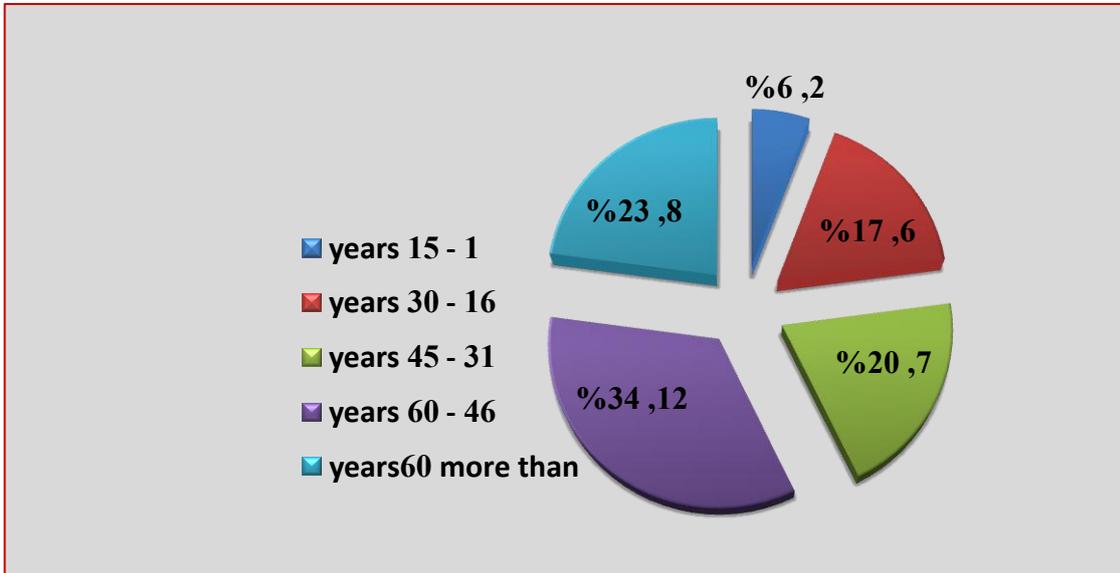
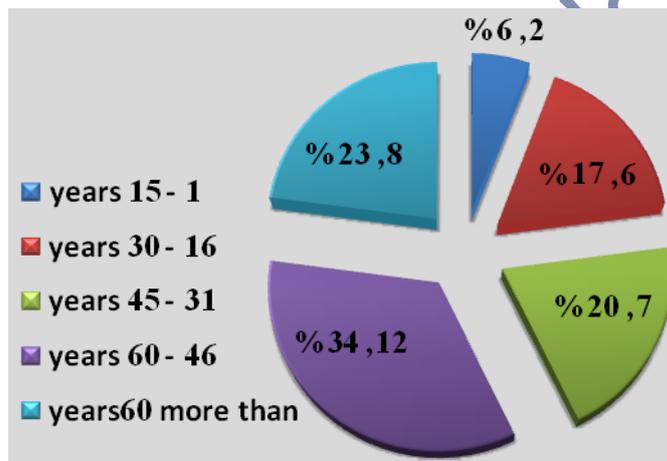
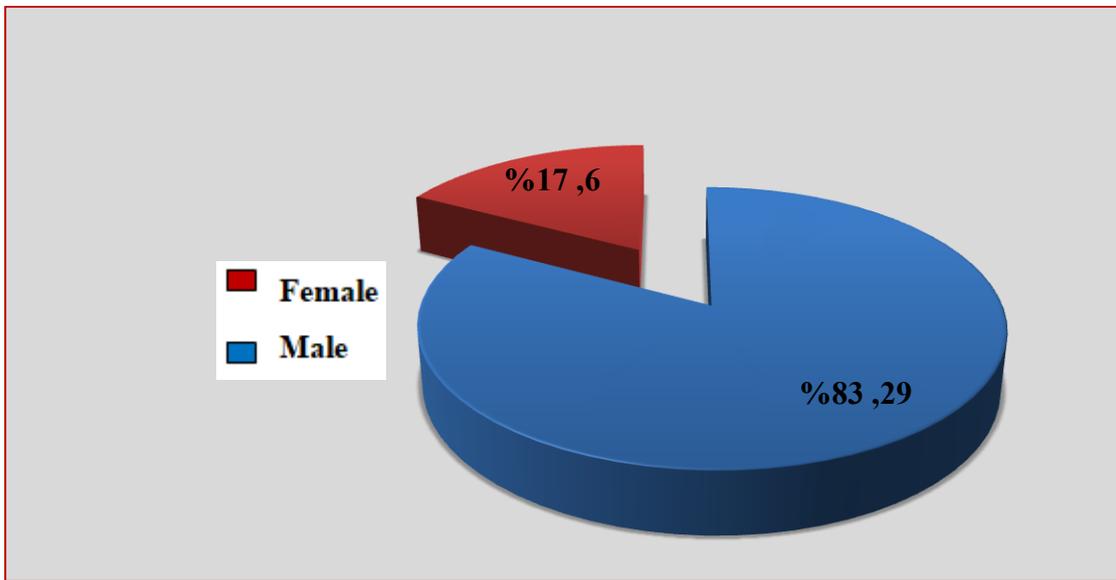


Figure 2. Distribution of MRSA isolates according to age group.



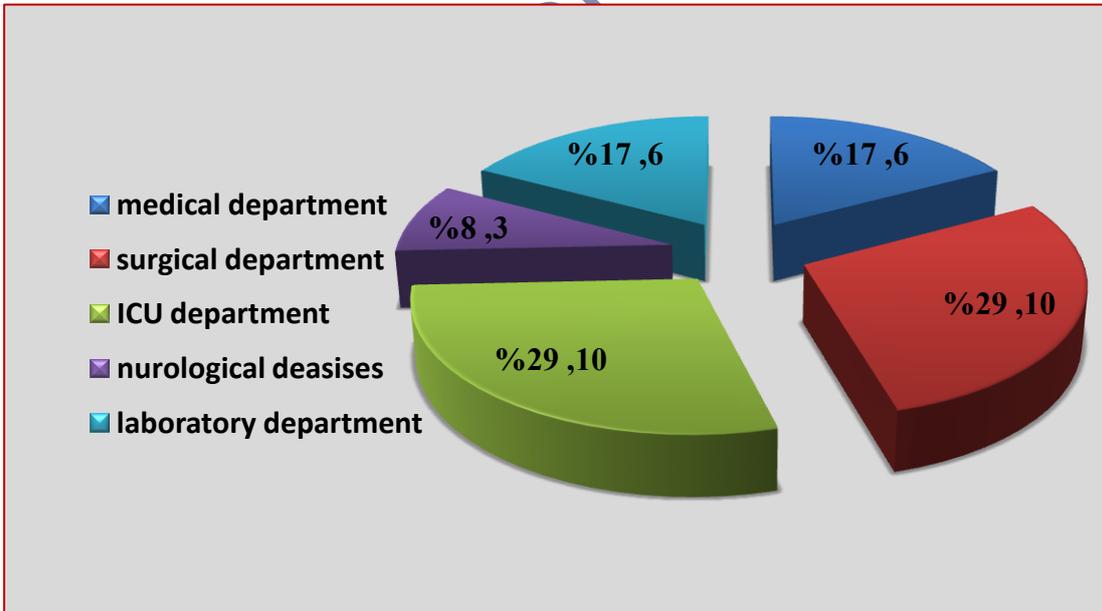
The figure 3 showed that the most of the MRSA isolates were in male about 29(83%), whereas the female participants only about 6(17%).



**Figure 3. Distribution of MRSA isolates according to gender.**

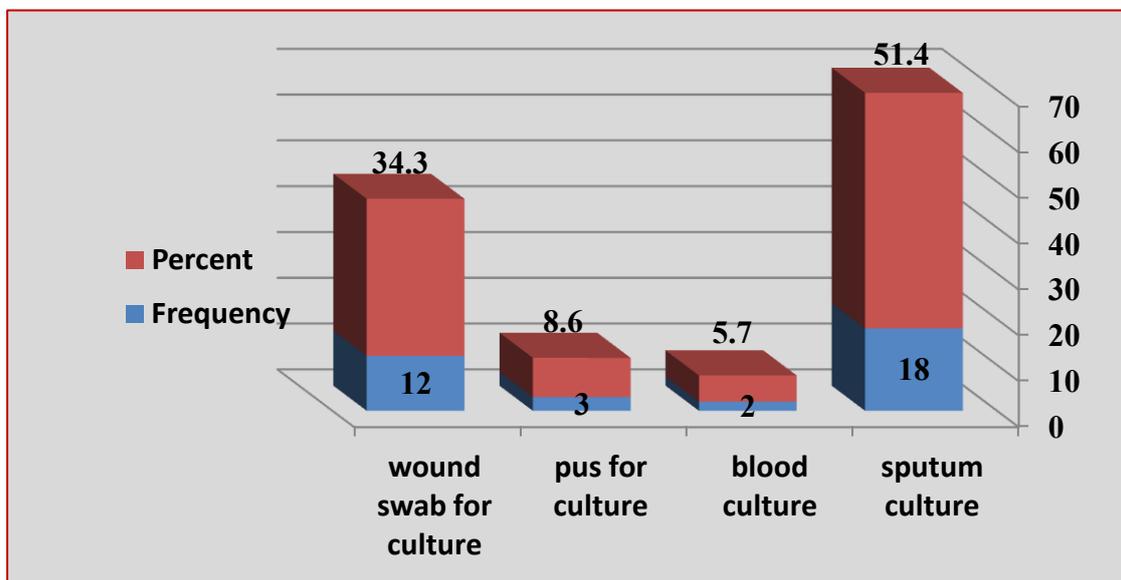
- Female
- Male

According to the study findings, ICU and surgical departments had the same and most prevalence of MRSA about 10(29%), followed by the medical and laboratory departments with rank about 6 (17%), and finally the neurological department only about 3(8 %).



**Figure 4. Distribution of MRSA isolates according to hospital departments.**

The figure 5 showed that more than half of MRSA isolates from sputum culture about 18(51.4%), followed by the wound swab culture about 12 (34.3%), and finally blood culture only about 2(5.7%).



**Figure 5. Distribution of MRSA isolates according to sample test.**

In this study, the most of medication that had resistance to MRSA test were about 12 drugs (80%), whereas the medication that sensitive to MRSA test about 3 drugs (20%). Also the study results showed that MRSA isolates were resistant to levofloxacin 100 %, amoxicillin/clavulanic acid 100%, moxifloxacin 76 %, gentamicin 94 %, clindamycin 70.5%, ciprofloxacin 97%, cfuroxime 91 % erythromycin 71.5%, and ampicillin/ sulbactam 94%. The highest frequency of sensitivity (100 %) was observed with trimethoprim/sulfamethoxazole, linezolid, and vancomycin (table 1).

**Table 1. The antibiotic susceptibility pattern for MRSA isolates**

Antibiotic	Expected options	Response		Antibiotic	Expected options	Response	
		F	%			F	%
Imipenem	S	10	30%	Moxifloxacin	S	0	00%
	R	18	55%		R	25	76%
	I	5	15%		I	8	24%
Amoxicillin / Clavulanic Acid	S	0	00%	Gentamicin	S	2	6%
	R	33	100%		R	32	94%
	I	0	00%		I	0	00%
Ampicillin/ Sulbactam	S	1	6%	Clindamycin	S	10	29.5 %
	R	16	94%		R	24	70.5 %
	I	0	00%		I	0	00%
Cefuroxime	S	3	9%	Levofloxacin	S	0	00%
	R	31	91%		R	35	100 %
	I	0	00%		I	0	00%
Erythromycin	S	10	28.5%	Trimethoprim/	S	35	100

				<b>sulfamethoxazole</b>			%
	R	25	71.5%	<b>ole</b>	R	0	00%
	I	0	00%		I	0	0.0%
<b>Ciprofloxacin</b>	S	1	3%	<b>Linezolid</b>	S	31	100%
	R	33	97%		R	0	00%
	I	0	00%		I	0	00%
<b>Doxycycline</b>	S	2	5.8%	<b>Vancomycin</b>	S	35	100%
	R	31	91.2%		R	0	00%
	I	1	3%		I	0	00%

The table (2) showed there were no statistical significant relationship between the usage of vancomycin and linezolid age group (P value = 0.74, 0.44).

**Table 2. The sensitivity pattern of vancomycin and Linezolid within age group.**

Age group	Vancomycin			The level of (P value)
	S	R	I	
1 - 15 years	2	0	0	<b>0.74</b>
16 - 30 years	6	0	0	
31 - 45 years	7	0	0	
46 - 60 years	11	0	0	
more than 60 years	8	0	0	
	linezolid			
1 - 15 years	2	0	0	<b>0.44</b>
16 - 30 years	6	0	0	
31 - 45 years	7	0	0	
46 - 60 years	10	0	0	
more than 60 years	6	0	0	

The table (3) showed there were no statistical significant relationship between the sensitivity pattern of vancomycin and linezolid hospital department (P-value = 0.63, 0.35).

**Table 3. The sensitivity pattern of vancomycin and linezolid within hospital department.**

Hospital Department	vancomycin			The level of (P value)
	S	R	I	
Medical department	6	0	0	<b>0.63</b>
Surgical department	10	0	0	
ICU department	9	0	0	
Neurological diseases	3	0	0	
Laboratory department	6	0	0	

Hospital Department	linezolid			0.35
Medical department	5	0	0	
Surgical department	10	0	0	
ICU department	8	0	0	
Neurological diseases	2	0	0	
Laboratory department	6	0	0	

Also the study findings showed that there was no statistical significant relationship between the sensitivity pattern of vancomycin and linezolid and sample type (P value= 0.80, 0.23).

**Table 3. The sensitivity pattern of vancomycin and linezolid within sample type.**

Sample Type	Vancomycin			P-value
	S	R	I	
Sputum culture	17	0	0	0.80
Blood culture	2	0	0	
Pus for culture	3	0	0	
wound swab for culture	12	0	0	
	linezolid			
Sputum culture	14	0	0	0.23
Blood culture	2	0	0	
Pus for culture	3	0	0	
Wound swab for culture	12	0	0	

#### 4. Discussion:

Recently, the prevalence of MRSA has steadily increased worldwide <sup>(4)</sup>. In addition, the antimicrobial susceptibility pattern of MRSA strains should be described. In our study, most *S. aureus* strains (n=81/199; 40.7%) were isolated from pus specimen. This is consistent with a previous study done in Nairobi<sup>(10)</sup>. In a study done to determine the antimicrobial susceptibility pattern of *S. aureus* strains isolated from hospitalized patients in Iran, most of the isolates were from blood specimens (29%)<sup>(11)</sup>. Another study done on prevalence and antibiotic susceptibility pattern of *S. aureus* from clinical isolates in Nigeria showed a majority of the isolates were from urine specimens (76%)<sup>(12)</sup>. The high number of *S. aureus* isolated in pus may be attributed to exposure of wounds which makes them more prone to infections and poor hygiene.

In this study, overall MRSA prevalence was 17.6 % (n=35/199; 17.6). This prevalence was lower than in previous studies that reported 31.5%<sup>(13)</sup> and 46.3%<sup>(14)</sup>. This difference could be due to various interventions during the study period such as infection control and appropriate antibiotic usage<sup>(14,15)</sup>.

Also MRSA prevalence was higher in studies done in two private hospitals in Nairobi, Kenya, which showed a 3.8% prevalence<sup>(16)</sup>. In addition, a study in Eritrea reported 0.03% of prevalence in Dutch hospitals<sup>(17)</sup>. This approved there is high variance of MRSA prevalence from different countries. The low prevalence of MRSA in private hospitals could be attributed to better infection controls<sup>(14)</sup>.

According to current study results, majority of MRSA was isolated from sputum specimens, 18/35 (51.4%). In contrast to study findings in Nigeria<sup>(6)</sup>, and Iran<sup>(11)</sup> showed different specimens were predominant. This variation in prevalence may be because of several factors like healthcare facilities available in the particular hospital, implementation and monitoring of infection control committee, rationale antibiotic usage which varies from hospital to hospital<sup>(14,15)</sup>.

In this study, 100% of MRSA isolates were sensitive to vancomycin. This finding is similar to a study done on antimicrobial susceptibility of MRSA in hospitalized patients in Iran, two hospitals in India and two private hospitals in Kenya showed 100% susceptibility to vancomycin<sup>(17, 18, 10)</sup>. On other hand, studies done in Iran<sup>(19)</sup> which showed 5% of the MRSA isolates were resistant to vancomycin and in a tertiary care hospital in India<sup>(20)</sup> and pediatrics and neonatal intensive care patients at Nairobi<sup>(6)</sup> which respectively showed 3.5 and 1% resistance to vancomycin among MRSA. Recently, many antibiotics with anti-MRSA activity were introduced to the market, some of which are available in Yemen. Linezolid is among these agents which is not widely available and it is not in use in many Yemeni hospitals including the setting of this study. In the present study, 100% of MRSA isolates were sensitive to linezolid. Garcia et al. have reported the first clinical outbreak of linezolid resistant *S. aureus* (LRSA) from a tertiary teaching university hospital in Madrid, Spain<sup>(21)</sup>. In their article they reported 12 patients with LRSA from which 6 patients died (with one death ascribed to LRSA infection). In the last two decades prevalence of MRSA has steadily increased worldwide. Therefore, they stated that this increase was associated with nosocomial transmission and extensive use of this antibiotic<sup>(22)</sup>.

In this study, the results showed that MRSA isolates were resistant to levofloxacin 100%, amoxicillin/clavulanic acid 100%, moxifloxacin 76%, gentamicin 94%, clindamycin 70.5%, ciprofloxacin 97%, cefuroxime 91% erythromycin 71.5%, ampicillin/ sulbactam 94%, and imipenem 55%. The highest frequency of sensitivity (100%) was observed with trimethoprim/sulfamethoxazole, linezolid, and vancomycin. Multidrug-resistance made the treatment of MRSA more difficult. This may be due to dispensing antibiotics without prescription and inappropriate selection of antibiotics for infections as empirical treatment worldwide. Also the use of broad-spectrum antibiotics for treating infections such as imipenem may increase the rate of MRSA and other resistant pathogens, so a more careful monitoring of antibiotics should be instituted. Vancomycin is the universally accepted drug of choice. We found

all MRSA isolates to be susceptible to vancomycin. Similar results have been reported by other authors<sup>(23,24)</sup>.

Resistant to quinolones (Levofloxacin, ciprofloxacin, and moxifloxacin) was high (100, 97%, and 76%; respectively) in the present study. In a previous study<sup>(25)</sup>, the resistant rate was also high (87.5%), but another study<sup>(26)</sup> conducted in 2003, reported the resistant rate of ciprofloxacin to be only 32.6%. The rapid emergence of quinolones was probably due to the indiscriminate empirical use of these drugs. An important finding of the present study was that the MRSA cases from ICU accounted for 17.6 % of all cases. Our finding was in agreement with the findings of Dominique et al.<sup>(27)</sup> from Switzerland. Also slightly higher than study conducted in 2003 from Nebal, reported the MRSA cases from ICU was only 10% from all MRSA cases<sup>(37)</sup>. A European study on prevalence of MRSA infection on samples from intensive care, estimated close to 65%<sup>(28)</sup>. In the present study, the low prevalence of MRSA, which included clinical samples from all hospital wards, illustrated the efficacy of relatively good infection control practice in the study setting. Also maximum resistance was seen with ampicillin-sulbactam, amoxicillin-clavulanic acid and gentamicin (table 3). Similar results were obtained in a previous study<sup>(29)</sup>. In addition, resistance was also seen with cefuroxime (91%), doxycyclin (91.2%), and erythromycin (71.5%). Gentamicin is a most commonly used drug, because of its low cost and synergistic activity with beta-lactam antibiotics. In the present study, 94% of gentamicin resistance was seen, which was slightly higher than a previous study<sup>(30)</sup>. Most common reason for multidrug resistant MRSA is indiscriminate use of antibiotics without drug sensitivity testing which may be due to lack of advanced laboratory facilities or negligence on the part of medical practitioners or patients poor economic status. Also this is due to the fact that MRSA is often multidrug resistant<sup>(6)</sup>.

Although vancomycin seems to be the only antimicrobial agent which showed 100% sensitivity and may be used as the drug of choice for treating multidrug-resistant MRSA infections, vancomycin is not a commonly prescribed drug, which is almost due to the higher nephrotoxic antibiotic and its unavailability in many parts of the country. The control of MRSA transmission seems to be the only hope to complete eradication of MRSA. The most effective way to control MRSA is good hand hygiene along with environmental cleaning of hospital rooms to reduce nosocomial rates of infection,<sup>(7)</sup>. The results of current study showed that there were highly susceptible to newer drugs such as linezolid. This finding differs from a study done by Arianpoor et al. in Iran which showed 5.5% of MRSA isolates were resistant to linezolid<sup>(19)</sup>.

## **5. Conclusion and recommendations:**

The present study showed a low level prevalence of MRSA. Also MRSA isolates were highly susceptible to newer drugs such as linezolid and to vancomycin which is not a commonly prescribed drug due to the higher nephrotoxic antibiotic. Also this study

showed that ICU and surgicalwards had the highest proportion of MRSA isolates of hospital departments. Information from this study may be used in future as abaseline for follow-up to the susceptibility trend of variousdrugs to be used for the treatment of *S. aureus* infections.Routine screening of MRSA and regular studiesshould be conducted to predict the trend of MRSA.

### **Conflict of interest**

The authors declare that they have no competing interests.

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