



# METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS (MRSA);

PREVALENCE AND SUSCEPTIBILITY PATTERN OF (MRSA) ISOLATED FROM PUS IN TERTIARY CARE OF DISTRICT HOSPITAL OF RAHIM YAR KHAN.

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**ABSTRACT... Background:** Methicillin resistant Staphylococcus aureus (MRSA) is one of the notorious bugs to cause nosocomial human infections worldwide. However its prevalence and susceptibility pattern quite varies in hospitals among different countries or within same country. **Objectives:** To study the prevalence and antibiotic susceptibility pattern of MRSA isolated from pus sample. **Study Design:** Cross sectional study. **Period:** 6 months. **Setting:** Tertiary care hospital of district Rahim Yar Khan -Punjab, Pakistan at its microbiology laboratory. **Materials and Methods:** A total 100 clinical specimen of pus and wounds were collected from various indoor and outdoor department of this institute. The Staphylococcus aureus was identified on the basis of colony morphology, Gram's stain, and biochemical tests. Antibiotic susceptibility pattern was done according to CLSI guidelines. **Results:** Out of 100 clinical sample of pus and wounds, 21 coagulase positive staph. aureus were isolated, 14 (66.7%) strains were found to be methicillin resistant and 07(33.3%) were found to be methicillin sensitive. Higher percentage of MRSA was isolated from surgical unit (66.7%), indicating its nosocomial source of spread. Maximum resistance to MRSA isolate were developed to Amoxicillin/ clavulanic acid (80%) followed by ofloxacin (71%), cefoxitin (66.7%), tetracycline (58%), Ciprofloxacin (48%), Erthromycin (46%) and Amikacin (40%). However, all strains were sensitive to vancomycin. **Conclusion:** There is high frequency of resistance among coagulase positive Methicillin resistant staph. aureus. Regular surveillance of hospital-associated infection and frequent monitoring of antibiotic sensitivity pattern is required to reduce MRSA prevalence.

**Key words:** MRSA, Prevalence, Antibiotic Susceptibility, Drug Resistance.

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## INTRODUCTION

*Staphylococcus aureus* is one of notorious microorganism that causes a wide variety of human diseases, from superficial skin infection to life threatening pneumonia, sepsis and endocarditis. Being a human commensal, it is difficult to eradicate and has a strong tendency to cause severe infection in both community as well as in hospitalized acquired patients. This increases morbidity as well as mortality and pose an enormous social and clinical burden on public health system in terms of treatment, hospital stay, equipment and isolation.<sup>1</sup>

Initially staphylococcus aureus responded well to the beta lactum group of antibiotics. Unfortunately MRSA (methicillin resistance staphylococcus aureus) have been reported in a very short

period of time. Now a day's MRSA is resistant to most of the antibiotics used to treat ordinary staphylococcus infection. The global prevalence of hospital as well as community acquired MRSA infection continue to increase worldwide.<sup>2</sup> Alteration in penicillin binding protein (PBP) and production of autolytic enzymes are the two main mechanism of resistance seen in MRSA associated infections. Moreover wide spread and irrational uses of antibiotics are its adding factors. So MRSA has become a serious threat and therapeutic challenge not only for clinicians but also for laboratory personal.<sup>3</sup>

Hospitals are one of the major sources of MRSA stain dissemination that leads to MRSA outbreaks both epidemically and endemically. Prolonged hospital stay, wide spread use of antibiotics,

nursing home exposure, immune suppression, improper antibiotic dosage, indwelling catheterization, invasive medical device, drug abusers, unsterilized instrumentations are another predisposing factors to MRSA spread. MRSA is important health issue in both developing & developed countries because it is contagious in nature and difficult to eradicate and treat.<sup>4-6</sup>

Prevalence and susceptibility pattern of MRSA isolates quite varies in hospitals among different countries or within same country. There was increase in prevalence of MRSA in United States over a period of 10 years from 32.7% in 1998 to 53.8% in 2007 and MRSA related hospitalization became doubled.<sup>7</sup> European SENTRY Study which constitute 25 hospitals showed MRSA isolates prevalence of 25% which were more prevalent in South region.<sup>8</sup>

MRSA prevalence largely varies among 25 European countries hospital (5% to 20%) with highest variation found in Germany while Lowest in Slovenia.<sup>9</sup> There is quite variation in prevalence of MRSA (22.6-77.6%) in hospitals of many Asian countries too.<sup>10</sup> The present study was conducted to prevalence of MRSA in tertiary care hospital of Rahim Yar Khan, Punjab-Pakistan.

## MATERIALS AND METHODS

This cross sectional study of 6 month duration (January to June 2017) was conducted at tertiary care hospital of district Rahim Yar Khan -Punjab, Pakistan at its microbiology laboratory. A total 100 clinical specimen of pus and wounds were collected from various indoor as well as outdoor department of this institute. A careful history was taken and it was ensured that patients were not using any antibiotics.

A sample of pus was obtained after incision and drainage of abscess. A sterile cotton swab was used to collect pus from wounds if it was not discharging. The inner side of the infected wound was swabbed gently and it was transferred to microbiology laboratory for further processing. Special precautions were taken during collection of sample to avoid any contamination. All collected sample immerse in Amies transport medium was

labeled and transported to the sterile laboratory profusion tube.

Inoculation of the samples was done on blood agar aerobically at 37°C for 48 hours. The identification of the staphylococcus stain was done on the basis of morphology, gram stain and other biochemical test following standard guidelines. A yellow to creamy 1-2 mm beta hemolytic colonies after overnight incubation on blood agar was used for preliminary identification.

## Antimicrobial Susceptibility Testing

A standard disk diffusion technique on Muller Hinton agar (Oxoid) was used for the antimicrobial susceptibility testing of all isolates following Clinical and Laboratory Standards Institute (CLSI) guidelines. Antibiotic impregnated disks (Oxoid) of amoxicillin (AM) 20 ug:10 ug, ciprofloxacin (CIP) 5 ug, amikacin (AK) 30 ug, erythromycin (E) 15 ug, vancomycin (VA) 30 ug, tetracycline (TE), ofloxacin and ceftiofloxacin (FOX) 30ug were applied to the plates inoculated at 15 mm distance from the edge of the petri dish. Plates were inverted and incubated at 37°C in an incubator for 24 hours.

## Interpretation of Susceptibility Testing

After an overnight incubation of 24 hours plates were examined to read the zones of inhibition by Vernier caliper and interpretation of results were done following were interpreted according to CLSI protocols. The strains of Staphylococcus aureus which were found to be resistant to ceftiofloxacin were screened as MRSA.

## RESULT

Clinical specimens were collected from various indoor as well as outdoor department of tertiary care hospital (Table-I). Out of 100 clinical specimens, 59% include pus and 41% wounds respectively. Out of 100, 65% sample showed growth while 35% sample showed no growth on blood agar plates. Out of 65 growth sample, there were (27) gram positive cocci and (38) gram negative rods. From (27) gram positive cocci (21) was staphylococcus aureus and others were S.epidermidis, and Streptococcus species. Out of 21 S.aureus there were (14) MRSA and (07)

MSSA (Table-II). Susceptibility pattern of MRSA and MSSA strains shown in Figure-1 and Figure-2 respectively.

Ward	Frequency	Percent
surgical ward	39	39.0
OPD	29	29.0
Ortho ward	13	13.0
Labor room	19	19.0
Total	100	100.0

Table-I. Sample distribution in different ward

Ward	Frequency	S. aureus	MRSA No. (%)	MSSA No. (%)
Surgical ward	38	09	06 (66.7)	03 (33.3)
OPD	30	08	05 (62.5)	03 (37.5)
Ortho ward	13	01	01 (100)	0 (0)
Labour room	19	03	02 (66.7)	01 (33.3)
Total	100	21	14 (66.7)	07 (33.3)

Table-II. Number and percentage of isolated MRSA and MSSA from different clinical samples

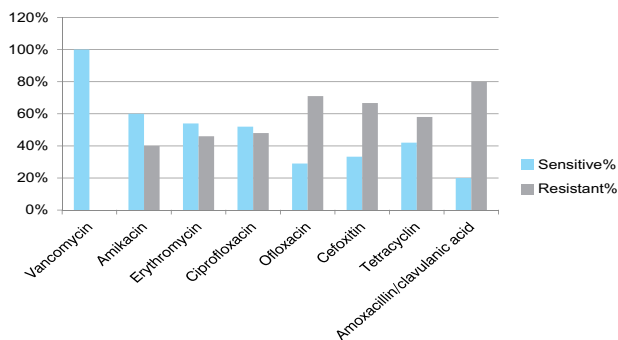


Figure-1. Susceptibility pattern of MRSA strain

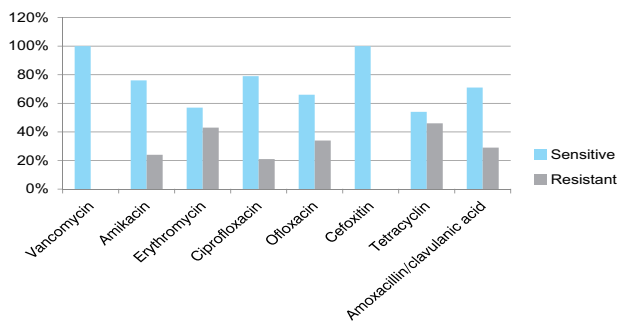


Figure-2. Susceptibility pattern of MSSA strain

## DISCUSSION

Nosocomial MRSA infections become a challenge for scientists by emerging new antibiotics day by day against this notorious microorganism. MRSA infection not only leads to significant morbidity and mortality, but it also adds significant social and economic burden especially to hospitalized patients. Vancomycin is now only the drug of choice against this multi drug resistant MRSA, however a low level resistance is being reported to some extent from different region of world.<sup>11</sup>

There is wide variation in the prevalence of hospital acquired MRSA infection. Even in United States hospitals, where health care facilities are quite up to date, there was drastically increase in the number of MRSA isolates from 22% to 57% over a period of six years.<sup>12</sup> Similar increase was noted in ICU setting, where MRSA isolate jumped from 35.9% to 64.4% within a decade.<sup>13</sup> There were reports of lower trend of MRSA hospitalization (2010-2014) too in United Sates, which reflect better control of skin and soft tissue infections. However hospitalization due to invasive MRSA remains unchanged.<sup>14</sup> Europe too showed a quite variation in MRSA infection ranging from 1-50%. MRSA infection prevalence in Europe is less than 5% in Denmark, Iceland, Netherland, Sweden and Norway. Some European countries showed above 25% MRSA prevalence such as Italy, United Kingdom, Romania, Spain, Turkey and Greece while Malta and Portugal showed above 50% MRSA prevalence.<sup>15</sup>

On the contrary MRSA was highly prevalent in Asian countries with 25.5% as community acquired and 67.4% as hospital acquired. The highest prevalence of MRSA was seen in hospitals of China, Japan, Taiwan and Korea (70%–80%).<sup>10</sup> Similarly India showed about 43% MRSA infection from 15 tertiary care centres over a period of two year.<sup>16</sup>

There were limited studies conducted on MRSA prevalence in Pakistan. A study conducted in Islamabad showed 39% total prevalence of MRSA infection, with 35 % in surgical ward and 65 % surgical intensive care unit (ICU). Our study also showed 66.07% MRSA isolates from surgical

ward including ICU indicating a source of its nosocomial spread.<sup>17</sup>

Most of the studies conducted in Pakistan revealed that overtime there was increase in the prevalence of MRSA infection i.e. Sargodha (23%) in 1999,<sup>18</sup> Lahore (38.5%) in 2001,<sup>19</sup> Karachi (43%) in 2005,<sup>20</sup> and, Rawalpindi (50%) in 2014<sup>21</sup> and Karachi (52%) in 2015-2017.<sup>22</sup> Our study showed 66.03 % MRSA prevalence which was quite high to above mentioned studies. A study conducted by Parveen et al<sup>23</sup> showed high prevalence of MRSA (60.40%) among five major government hospitals of Rawalpindi. A study conducted by Khan et al<sup>24</sup> showed 53.3% MRSA prevalence in two major hospitals of Rawalpindi- Islamabad. Up till now high MRSA prevalence was found to be 71.1% in 2015 at National Institute of Health (NIH), Islamabad.<sup>25</sup> However, two studies which were conducted in Peshawar during 2012-2013 revealed very low MRSA prevalence (1-5.2%).<sup>26,27</sup>

On the contrary a multicentre study conducted in Pakistan showed a geographical variation of MRSA infection being lowest MRSA resistance seen in Sukkur (2%) followed by Quetta (26%), Azad Kashmir (32%), Rawalpindi -Islamabad (46%) and highest resistance was seen in Lahore (61%) & Karachi (57%) respectively.<sup>28</sup> In above mentioned international and national studies, variations in MRSA prevalence caused by a number of factors in hospitals such as age, immune status, self medication, rationale use of antibiotics, prior hospitalization, hygienic condition of hospitals, population dynamics, health care facilities, implementation of infection control program and severity of infection.

Vancomycin showed 100% susceptibility to all MRSA strains in our study and no MRSA strain showed resistance to this antibiotic, which is comparable to other studies. Maximum resistance to MRSA isolates from pus samples in our study was developed to Amoxicillin/ clavulanic acid (80%) followed by ofloxacin (71%), cefoxitin (66.7%), tetracycline (58%), Ciprofloxacin (48%), Erthromycin (46%) and Amikacin (40%). This was lower than previous other studies which may be due to better in hospital antibiotic policy.<sup>29,30</sup> From

our study, we can assume that glycopeptides is the drug of choice for the treatment of multidrug resistance MRSA stain. However it should be reserved for life threatening infection caused by MRSA.

Antibiotic resistance emerging at an alarming rate and becomes a health challenge in Pakistan. In order to prevent antibiotic resistance there is strong need to develop antibiotic stewardship which should be done by the mutual cooperation of clinicians, pharmacist and clinical microbiologist. Moreover strict policies should be made and implemented so that irrational use of antibiotics should be discouraged. Good hygienic conditions and local surveillance of antibiotics resistant profile on continuous basis is the best way to prevent MRSA infection.<sup>31</sup>

## CONCLUSION

There is high frequency of resistance among coagulase positive Methicillin resistant staph. Aureus. Early detection of this nosocomial pathogen is also very necessary in asymptomatic colonized health care workers in hospital in order to prevent its dissemination and life threatening complications.

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NO MAN IS RICH ENOUGH  
TO BUY BACK HIS PAST.



“Oscar Wilde”

### AUTHORSHIP AND CONTRIBUTION DECLARATION

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