

Antibiotic sensitivity to methicillin and methicillin plus macrolide-lincosamide-streptogramins resistant Staphylococcus aureus (MRSA& MLS-MRSA) in Qazi Hussain Ahmed Medical Complex Nowshera – Hamzullah Khan - Nowshera Medical College

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Objectives: Culture and Sensitivity pattern of Methicillin and Methicillin plus Resistant Staphylococcus aureus (MRSA & MLS-B type MRSA) isolates (sputum and pus samples) in a tertiary care hospital of Nowshera. **Methods:** This cross sectional study was performed in the pathology laboratory of Qazi Hussain Ahmed Medical Complex Nowshera from 1st May 2018 to 30th Oct 2019. A total of 235 isolates were studied and 86 samples showed MRSA. Out of 86 MRSA, 19 were also resistant to Macrolide-lincosamidestreptogramin-B antibiotics (MLS-B). Relevant information were recorded on a pre-designed proforma prepared as per CLSI Recommendation for data collection. **Results:** Out of 235 patients, 86 MRSA Positive isolates (36.59%) were selected for antibiotic susceptibility. Out of total, 52(60.5%) were females and 34(39.5%) were males. The mean age with standard deviation was 28.48+6.85. The age range was from 20 years to 55 years of age. Mode of age was 25years. The sensitivity pattern to MRSA was; Vancomycin 100%, Linezolid 100%, Rifampicin 86.04%, Clindamycin, 73.25%, Fusidic acid 60.46%, Erythromycin 44.18%, Doxycycline 32.55%, levofloxacin 16.27%, Gentamycin 13.95% and ciprofloxacin 11.62%. 27% cases with MRSA were also resistant to (MLS-B) antibiotics and were labeled as MPRSA (Methicillin plus resistant Staph.aureus) present study. It is further to exhibit that MRSA are prevalent in younger age (20-30 years) 61(68%) as compared to the older age mean youth under threat of MRSA. Using logistic regression analysis it was noted that the probability of MRSA in female gender was 1.5 times more than male gender with a significant p-value (P-value=0.04, OR=1.54). The sensitivity to Vancomycin and Linezolid was 100 in MRSA and MLS-MRSA isolates.

Conclusion: sensitivity of Vancomycin and Linezolid was 100% in MRSA and should be kept reserved for MRSA cases to avoid misuse of antibiotics and to reduce resistance to these precious antibiotics. Methicillin resistant infections are now a challenge to the clinician and at the same time are extremely expensive to be afforded by a common person. MRSA is a public health threat that needs accumulative response through advocacy, communication and social mobilization **Keywords:** Antibiotic resistance, MRSA, Misuse of antibiotics. Introduction Antimicrobial resistance (AMR) is a serious global health concern, specially reported with Gram-positive bacteria, the important bug notorious for it is the MRSA that is emerging with serious threats. Infections associated with Methicillin-Resistant Staphylococcus aureus (MRSA) is a major global healthcare issue and challenging the clinicians to treat such type of infections. S. aureus bacteremia, exhibits a very high rate of morbidity and mortality worldwide and can lead to life threatening conditions like infective endocarditis or sepsis by metastasis². Resistance to methicillin is mediated by mec-A gene, which encodes the polypeptide PBP2a protein³. The timely advised antibacterial therapy has better outcomes in treatment of methicillin resistant Staph aureus infections. Bacteremia caused by methicillin sensitive Staph aureus i.e. MSSA, therapy of choice is β -lactam antibiotics with acceptable results but for infections caused by MRSA strands, the antibiotics of choice are vancomycin or daptomycin as per recommendations of the Infectious Diseases Society of America guidelines and many other meta analysis⁴⁻⁵. Clindamycin is an important drug duly approved by FDA and is used to treat MRSA infections. Unlike other antibiotics such as

doxycyclin, trimethoprim, sulfamethoxazole, rifampin, and linezolid, its use has been less restrained by safety considerations and precautions. Vancomycin and linezolid is used are gold standard drug for MRSA associated bactremia. Concern associated with Vancomycin is its bactericidal activity is relatively slow and it poorly penetrates some tissues. Present study was conducted to determine the antimicrobial efficacy of different antibiotics for the treatment of bacteremia caused by MRSA. This work is partly presented at 7th World Congress and Exhibition on Antibiotics and Antibiotic Resistance March on 16-17, 2020 held at London, UK Vol.8 No.1 Extended Abstract International Research Journal of Pharmacy and Pharmacology 2020 Materials and Methods: This Cross sectional study was performed in the Pathology department of Qazi Hussain Ahmed Medical Complex Nowshera from 1st May 2018 to 30th Oct 2019. A total of 235 isolates were studied and 86 samples showed MRSA pattern (36.59%). The inclusion criteria were all cases irrespective of age and gender received in the laboratory. Exclusion criteria were samples received in the laboratory 24hour after collection, patient already on the antibiotic therapy and improperly collected sputum and pus samples. The samples were received in the pathology section from the respective unit under observance of strict aseptic technique after education of patients on pus and sputum sample collection. Media were prepared as per CLSI (Clinical and laboratory standard institutes). Sample size was calculated on the following assumption. The anticipated proportion of the MRSA in surgical units, 42% Absolute precision: 6% Confidence level: 95% Sample size was 265. Drop Outs.(lost to follow up)-acceptable upto 10%.to be added to the sampling side. $265+25=285$ Sample size was $335+35$ (lost to follow up) $=285$ All samples were inoculated on selective medium MSA (Mannitol Salt Agar). Then the specimens were incubated under ambient air 35 ± 2 C for 18-20 hours. In case growth is obtained on MSA then further inoculated on Mueller Hinton agar for

sensitivity to antibiotics as per CLSI recommendations. The antibiotic disks used were; VA-Vancomycin, LZD-Linezolid, RD-Rifampicin, D-clindamycin, E-Erythromycin, Fd-Fusidic acid, Dox-Doxycyclin, Lev- Levofloxacin, CNG-Gentamycin, Cip-ciprofloxacin, SXZ (Cotrimaxazole) and Foxcefoxetin. The accepted zones of sensitivity taken in consideration as per CLSI (Clinical Laboratory Standard Institute) Guideline 20158 for different antibiotic disks were: Linezolid >21 mm, Vancomycin >20 mm (Now MIC Test is recommended in fresh CLSI), Rifampicin >20 mm Clindamycin >21 mm, tetracycline >19 mm, ciprofloxacin >21 mm, levofloxacin >19 mm, Gentamycin >15 mm and CXZ (Trimethoprim Sulfamethoxazole) >16 mm. For recognition of MLS-MRSA, or MRSA with further resistance to MLS-B antibiotics, the disk of Erythromycin and Clindamycin were placed at a distance of 20mm center to center. Phenotypically MSL resistance was confirmed as inhibition of zone of clindamycin towards erythromycin as a straight line, resembling the alphabet "D" and was considered to be positive for D-Test phenomenon (Figure 1). Any haziness in the zone of inhibition of clindamycin is also phenotypically representative of resistance. Finally the data obtained from the culture and sensitivity was entered in a SPSS version 25 for descriptive and correlation analysis of different parameters. Results: Out of 235 patients, 86 MRSA Positive isolates (36.59%) were selected for antibiotic susceptibility. Out of total, 52(60.5%) were females and 34(39.5%) were males, with male to female ratio of 1.4:1 (Table 1). The mean age with standard deviation was 28.48 ± 6.85 . The age range was from 20 years to 55 years of age. Mode of age was 25 years (Table 2). The sensitivity to Vancomycin and Linezolid was 100 in MRSA isolates. Sensitivity to The next antibiotic showed higher sensitivity was: Rifampicin 86%, Clindamycin, 73.25%, Erythromycin 44.18% and Doxycyclin was 32.55%. Sensitivity to quinolones and fluoroquinolones was not remarkable. Similarly no

sensitivity was recorded for MRSA with cotrimaxazole. Using logistic regression analysis it was noted that the probability of MRSA in female gender was 1.5 times more than male gender with a significant p-value (P-value=0.04, OR=1.54) Table 4a&b. The sensitivity to clindamycine was 73% due to expression of erm gene encoding Methylase by erythromycin confirmed phenotypically by D-test Phenomenon in-vitro. It is further to exhibit that MRSA are prevalent in younger age (20-30 years) 61(68%) as compared to the older age mean youth under threat of MRSA. Discussion: Continuously Emerging resistance to available antibiotics by MRSA is a global threat and challenge to the clinicians in both clinical facilities and community settings⁹. We observed the prevalence of MRSA in teaching hospital set up as (36.59%) among the total Staph aureus isolates. MRSA is reported worldwide nearly from all regions of the world with variable frequencies. The frequency of MRSA in Pakistan and other neighboring countries like India has been shown to be high if compared with developed world like USA and Europe. Many factors can contribute to this difference¹⁰. A study reported from Africa showed the prevalence of methicillin resistance in S. aureus from 42 to 51 %¹¹ that quit matching our findings.