ANTIBIOGRAM OF BIOFILM PRODUCING AND NON-PRODUCING COMMUNITY ACQUIRED-METHICILLIN RESISTANT *Staphylococcus aureus* ISOLATED FROM POTENTIAL RISK POPULATION OF DHARAN, NEPAL

A
Dissertation
Submitted to the **Department of Microbiology,**
Central Campus of Technology, Tribhuvan University, Dharan, Nepal, in Partial Fulfillment of the Requirements for the Award of Degree of Masters of Science in Microbiology
(Medical)

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RECOMMENDATION

This is to certify that **Mr. Jenish Shakya** has completed this dissertation work entitled **"Antibiogram of Biofilm Producing and Non-Producing Community Acquired–Methicillin Resistant S*taphylococcus aureus* isolated from Potential Risk Population of Dharan, Nepal"** as a partial fulfillment of the requirement of M.Sc. degree in Microbiology (Medical) under my supervision. To my knowledge, this work has not been submitted for any other degree/s.

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CERTIFICATE OF APPROVAL

On the recommendation of **Asst. Professor Mr. Hemanta Khanal** this dissertation work of **Mr. Jenish Shakya** entitled **"Antibiogram of Biofilm Producing and Non-Producing Community Acquired–Methicillin Resistant S*taphylococcus aureus* isolated from Potential Risk Population of Dharan, Nepal"** has been approved for the examination and is submitted for the Tribhuvan University in Partial fulfillment of the requirements for M.Sc. degree in Microbiology (**Medical**).

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ABSTRACT

Staphylococcus aureus is one of the common cause of hospital acquired infection and community acquired infections. Nowadays these organisms gas become resistant towards variety of drugs. MRSA is the emerging antibiotic resistant bacteria that are resistant to methicillin antibiotic and known to be the infectious pathogen causing severe infection and a cause of fatal mortality. Aim: Altogether 200 nasal swabs and 200 hand swabs were taken from and transported to microbiology lab in cold chain. The samples were swabbed in mannitol salt agar containing oxacillin powder of 6mg/L and incubated at 37°C for 24 hrs. *Staphylococcus aureus* colonies were identified based on growth characteristics on MSA plates (golden yellow colonies), Gram stain and positive results for coagulase and catalase test. The pure isolated MRSA were subjected to antibiotic susceptibility tests, biofilm formation assays, and MIC. From our study the overall prevalence of CA-MRSA was 61.5%. Higher frequency of multi-drug resistant MRSA was isolated. The biofilm producing CA-MRSA were 51.2% and rest (48.7%) were non-producers. There was significant association in biofilm production with multi-drug resistance ($p<0.05$). The prevalence of CA-MRSA was found more in barbers followed by beauticians and municipal waste workers in comparison to healthy controls. The 51.2% isolates' were biofilm producing CA-MRSA were which showed significant drug resistance. Ciprofloxacin was most sensitive drug against the isolates which was statistically significant ($p<0.05$). The resistant pattern of biofilm producers reported high ability of multi-drug resistance compared to non-biofilm producers ($p<0.05$). Microtitre plate method was found to be gold standard over tube and congo red agar method for screening biofilm formation. The prevalence of VISA and VRSA among CA-MRSA was found to be 49.5 % and 40.6% respectively among the isolates. Improvement in personal hygiene and formulation of appropriate health policy helps to prevent CA-MRSA infection. This study concludes that CA-MRSA is still emerging with multi-drug resistance. The emergence of VISA and VRSA strains has increased concern in vancomycin treatment failure.

Keywords: CA-MRSA, VISA, VRSA, biofilm, antibiotic susceptibility test, multi-drug resistance

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LIST OF ABBREVIATIONS

AMR	:	Antimicrobial Resistance
ATCC	:	American Type Culture Collection
CA-MRSA	:	Community Acquired Methicillin Resistant *S. aureus*
CDC	:	Centre for Disease Control
CLSI	:	Clinical and Laboratory Standards Institute
CRA	:	Congo Red Agar
DNA	:	Deoxyribonucleic Acid
ELISA	:	Enzyme Linked Immunosorbent Assay
FDA	:	Food and Drug Administration
HA-MRSA	:	Hospital Acquired Methicillin Resistant *S. aureus*
KDa	:	Kilo Dalton
NB	:	Nutrient Broth
MDR	:	Multidrug Resistance
MIC	:	Minimum Inhibitory Concentration
MSA	:	Mannitol Salt Agar
NCCLS	:	National Committee for Clinical Laboratory Standards
OD	:	Optical Density
PBP	:	Penicillin Binding Protein
PBS		Phosphate Buffer Saline
PVL	:	Panton-Valentine leucocidin
TSB	:	Trypticase Soya Broth
TM	:	Tube Method
VISA	:	Vancomycin-intermediate *S. aureus*
VRSA	:	Vancomycin-resistant *S. aureus*
VSSA	:	Vancomycin-susceptible *S. aureus*
WHO	:	World Health Organization

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