#### **Abstract**

Conjunctival infections cause a worldwide problem and affects people of different ages. Proper treatment can reduce symptoms, recovery time, contagious spread, possible re-infection and risk of complications but prolonged use of antibiotics can cause resistance strains. Infections with MDROs can lead to inadequate or delayed antimicrobial therapy.

Staphylococcus spp. is a clinically relevant pathogen due to its antimicrobial resistance and evasion of the host immune system. Its virulence factors in avoiding host responses and influencing disease make them able to form biofilm. Emergence of resistants staphylococci from the conjunctiva is of great concern because its virulence is related to the clinical outcome of ocular infections like keratitis or endophthalmitis.

Biofilm forming multidrug resistant *Staphylococcus* spp. are major reservoirs for transmission of ophthalmic infections. Various extracellular substances enable bacteria to form biofilm. Production of these components is dependent on the presence of biofilm-essential genes such as the *ica* operon.

The aim of this study was to determine association of *ica A* with biofilm formation in staphylococci isolated from patients with conjunctivitis, determine association of *ica A* gene with antibiotic resistance profile in *Staphylococci* causing conjunctivitis.

Fifty subjects suffering from staphylococcal bacterial conjunctivitis were included in this study. Specimen collection was done followed by identification for bacterial identification, antibiotic sensitivity test was done for each isolate to detect proper antimicrobial treatment. Biofilm formation was detected by CRA and MTP (phenotypic detection). Conventional PCR technique was done to detect the presence of *ica A* gene and its relation to the biofilm.

Among the 50 patients; 30 S. aureus (60%) and 20 CoNS (40%) were isolated.

Detection of *S. aureus* biofilm by MTP showed 8 strong biofilm forming isolates (26.7%), 7 moderate (23.3%), 3 weak (10%) and 12 negative (40%). While detection of biofilm of *S. aureus* by CRA showed 7 Strong (23.3%), 8 moderate (26.7%), 3 Weak (10%) and 12 negative (40%). Detection of *ica A* gene in *S. aureus* was found in 18 biofilm producing isolates by phenotypic methods and was not found in 12 biofilm producing isolates.

**Keywords:** Association of Intercellular Adhesion Gene A (ica A) with Biofilm Formation in Staphylococci Isolated from Patients with Conjunctivitis

# Association of Intercellular Adhesion Gene A (ica A) with Biofilm Formation in Staphylococci Isolated from Patients with Conjunctivitis

#### Thesis

Submitted for partial fulfillment of MD degree in Basic Medical Sciences (Medical Microbiology & Immunology)

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## **List of Abbreviations**

Aap	Associated accumulation protein
Agr	Accessory gene regulatory
AHL	Acylated homoserine lactone
AI	Auto Inducer
AIP	Auto inducing cyclic thiolactone peptides
AK	Amikacin
AMP	Ampicillin
AMR	Antimicrobial resistance
AMX	Amoxicillin
Bap	Biofilm associated protein
ВНІ	Brain heart infusion agar
CDC	Centers for Disease Control and Prevention
clfB	Clumping factor B
CLSI	Clinical and laboratory standards institute
CN	Gentamycin
CoNS	Coagulase negative staphylococci
СР	Capsular polysaccharide
C/P	Ciprofloxacin
CRA	Congo Red Agar
CRO	Ceftraixone
DA	Clindamycin
DNase	Deoxyribonuclease

Dpnag	De poly-N-acetylated glucosamine
Edna	Extracellular Deoxyribonucleic acid
DspB	Dispersin B
${f E}$	Erythromycin
ECDC	European Centre for Disease Prevention and Control
EPS	Extracellular polymeric substances
FnBP	Fibronectin binding protein
FOX	Cefoxitin
HIV	Human immunodeficiency virus
HSV	Herpes simplex virus
Ica	Intracellular adhesion gene
IL	Interleukin
IOL	Intraocular lenses
LZD	Linezolid
LPS	Lipopolysaccharides
M.B.E.C	Minimum biofilm eradication concentration
МНА	Muller Hinton agar
MRSA	Methicillin resistant S. aureus
MSCRAMM	Microbial Surface Components Recognizing Adhesive Matrix Molecules
MSSA	Methicillin sensitive S. aureus
MTP	MicroTitre plate

N. gonorrhoeae	Neisseria gonorrhoeae
OD	Optical density
ODc	Cut-off value
OPD	Ophthalmology outpatient department
P	Penicillin
P. aeruginosa	Pseudomonas aeruginosa
PBP	Penicillin binding protein
PDR	Pan drug resistant
PIA	Polysaccharide intercellular adhesin
PNAG	Poly N- acetylglucosamine
PSM	Phenol soluble modulin
PVC	Polyvinyl chloride
QQ	Quorum quenching
QRDR	Quinolone resistance determining region
QS	Quorum sensing
RD	Rifampicin
S. aureus	Staphylococcus aureus
S. capitis	Staphylococcus capitis
S. caprae	Staphylococcus caprae
S. chromogenous	Staphylococcus chromogenes
S. epidermidis	Staphylococcus epidermidis
S. hominis	Staphylococcus hominis
S. hyicus	Staphylococcus hyicus

S. pneumoniae	Streptococcus pneumoniae
S. saprophyticus	Staphylococcus saprophyticus
SCCmec	Staphylococcal cassette chromosome mec
SCV	Small colony variant
SE	Staphylococcal Enterotoxin
TMP-SXT	Trimethoprim Sulphamethoprim
TSB	Tryptic soy broth
TSST	Toxic Shock Syndrome Toxin
VA	Vancomycin
VISA	Vancomycin intermediate resistant <i>S. aureus</i>
VKC	Vernal keratoconjunctivitis
VRSA	Vancomycin drug resistance
VZV	Varicella Zoster virus
XDR	Extensively drug resistant