STUDY OF β LACTAM RESISTANCES IN MRSA FOR DEVELOPMENT OF VACCINE

*Nagesh N. Patil¹, Abhishek Salunkhe², Abhijeet Nimbalkar³, Aditya Patil², Hirachand Khade²

Biocon Academy, Biocon, Bangalore India ¹
Department of Biotechnology, TKIET, Kolhapur. India²
Department of Biotechnology, KIT, Kolhapur. India³

ABSTRACT

The Staphylococcus aureus is evolutionary develops resistance to any antibiotics. The widespread use develops resistance to β lactams in Staphylococcus aureus. It reduces susceptibility towards penicillin, methicillin and vancomycin antibiotics. MRSA is a major cause of soft tissue and skin infection. The high growth rate, rapidly spread with in community are characteristics of community-MRSA. The mecA gene of SSCmec cassette is developing resistance against antibiotics. The mecA gene encodes the penicillin binding protein (PBP2a) with low affinity for β-lactam. The type IV SCCmec chromosome cassette produces Panton Valentine leukocidin (PVL) toxin, which is responsible for more virulence. The control of MRSA is very difficult due to changes in study data from publications to publications, change in screening, populations, protocols, healthcare practices and infection control efforts. Now to reduce the drug resistance this review suggests that use non β lactam antibiotics to cure MRSA. In 2010, FDA approved linezolid (Zyvox – Pfizer) for treatment of MRSA. The several new approaches are under investigation to replace antimicrobial therapy. Antimicrobial peptides, vaccines are most areas of interest to develop new therapy. Researcher use glycolipids, mutated protein A as an antigen for vaccine development but all cases are unsuccessful. Target vaccine should be against the Panton Valentine leukocidin (PVL). Interleukin 17 secreted by the T\textsubscript{H} 17 cell are the key factor for development of vaccine for MRSA.

Key words: β lactam, CA-MRSA, SSCmec, PVL, linezolid, Interleukin 17.
INTRODUCTION

*Staphylococcus aureus* is opportunistic it inter in to the body through cut or other wound. But in case of some people who have hospitalized for a long time, then ordinary staphylococcus infections can cause serious illness. *Staphylococcus aureus* is naturally developing resistance to any antibiotics. The antibiotics prescribed for common cold, cough, flu and viral infection are mostly developed the anti-antibiotics response in the various bacteria, result in the development of super bugs. In the US the antibiotics are found in animals feed as well. The routine uses of antibiotic for the animals like pig, chicken are runoff into contaminant livestock \[1-3\]. This resistance develops by the horizontal gene from outside side. The uncontrolled uses offβ-lactam antibiotics are responsible for the development of MRSA. MRSA is a main source of blood, skin, soft tissue infections and surgical site infections. This *Staphylococcus aureus* reduced antibiotic susceptibility for daptomycin, mupirocin, vancomycin, clindamycin. Wider use of antibiotics treatments, longer hospital stay and high treatment cost are main factor responsible for the development of MSRA \[2-5\]. The acquisition of mecA gene is developing methillicin resistance in the *Staphylococcus aureus*. The mecA gene encodes the PBP2a protein with low affinity for β-lactam. PBP2a gene produces resistance to all β-lactam antibiotics and cephalosporin.

Development of MRSA

In early 1940, the wide use of penicillin develops penicillin resistant in the *Staphylococcus aureus*. This strain contains penicillinase which hydrolyze β-lactam ring. In 1960 broad spectrum antibiotic methicillin is used against the penicillin resistance *Staphylococcus aureus*. Methicillin resistance is introduced in 1961. One study in the UK were reported that *Staphylococcus aureus* isolate develop anti methicillin effect. The staphylococcus cassette contains chromosome SSCmec, which carry methillicin resistance mecA determinant. In 1970, MRSA is spread in hospitals worldwide. The increasing infection of MRSA, lead to uncontrolled use of vancomycin. *Staphylococcus aureus* are then susceptible to vancomycin and then develop Vancomycin resistance *Staphylococcus aureus*. In 1990, Western Australia researcher fined some strains of MRSA are develop resistant to the non β-lactam antibiotics \[6-9\]. The genes responsible for antibiotic resistance are due to mutation and deletions of determinants. mecA, ermC, blaZ, tetK, fusC, fusA, dfrA, thyA, ileS-2, aacA-aphD, grlA, gyrA, rplC, ermC \[10-12\].
Community Acquired MRSA (CA-MRSA)

CA-MRSA infection transmitted within family, person to person associated with pediatric fatalities. This is a major cause of soft tissue and skin infection with fulminant and lethal infection. High growth rate, rapidly spread with in community and resistance to antibiotics are characteristics of CA-MRSA. The acquisitions of mecA gene are circulated within the community. This infection contains type IV SCCmec chromosome cassette. Some of Staphylococcus aureus are have ability to develop Panton Valentine leukocidin (PVL) toxin, which is responsible for more virulence. The methillicin susceptible Staphylococcus aureus are leading strains for the CA-MRSA [13]. The first case of CA-MRSA is reported in the US. The strain Staphylococcus aureus MW2 causes the community infection related with PVL negative protein clone WA-1. US300 strain is more virulent than other strains. The virulence of this strain is more lethal and cause extensive diseases in animals. The genome of US300 stain contains plasmids, phasmids, pathogenecity genes. The toxin PVL contain two subunit LukS-PV and LukF-PV are causes cell lyses, release mediators of inflammations [14-15]. There are various different SCCmec genes, which have different in insertion sequence and antimicrobial resistance gene. The gene mecA encodes the PBP 2a protein while mecR1 and mecI are regulatory genes of mecA. The increase in antibiotic resistance are corresponding to changes in SCCmec gene. The SCCmec IV gene is responsible for CA-MRSA, which is evolved from SCCmecI gene [16, 17].

Limitations in current research

The resistances of β-lactam are evolutionary developed in Staphylococcus aureus. The effective surveillance and appropriate treatment are essential to control MRSA infection. The various authors reported, the proportion of MRSA increasing worldwide, but it’s difficult for comparison of results between studies. The baseline rate of MRSA changes publications to publications due to change in screening, populations, protocols, healthcare practices, infection control efforts. This change is due to the number of total admission and virulence of MRSA to stay in hospitals. The most challenge is preventing bias arising in collecting bacteriological samples and interpretation of results. The current research not contain any guideline for when we should collect culture, storage of sample, automation in reporting for minimizing variations in decision making process [18-20].

The pooling of data from studies, surveillance programs are helps in identification and potential of antimicrobial resistance. The proportion of hospitalization and death in MRSA
are important to study the mortality. The attendants on mortality data obtained from various studies are important for investigation of limitations in infection control and transmission of bacteria. The methillicin resistance between strains are varies from countries, hospitals, populations. The rate of MSRA in USA is high while in Europe 0.5 % strains are only methillicinresistance. It is most important to the evolutionary study of Staphylococcus aureus from various regions to eliminate potential differences of infections, virulence and role of pathogen [21-23].

DISCUSSION
In India Mangosteen (Garciniamangostana) fruit is used as traditional medicine to cure skin diseases and wounds [24]. The bioactive compound Xanthone derivatives presents in mangosteen shows antifungal, antibacterial, anti-inflammatory action. αmangostin is major derivative used as antimicrobial compound. Cytoplasmic enzymes are the main target of αmangostin for bactericidal action. [25-26]

In Japan, Lactobacillus probiotics are used to cure MRSA infection. Basically Lactobacilli shows beneficial effect against gram positive and gram negative bacteria. Where the same study reported that lactobacillus acidophilus have special antagonistic effect against the Staphylococcus aureus. These lactic acid producing bacteria are generating bactericidal bioactive peptide and enzymes prevent biofilm formation of MRSA. Lactacin B& F, nicin, casein 80 are the bacterisin produced by the lactobacillus [27-28]. The LAB (lactic acid bacterium)lactobacillus acidophilus CL1285 reduce the MRSA cells up to 5 CFU within 24 hours [29].

This review suggest that use non β lactam antibiotics to cure MRSA e.g. linezolid. The vancomycin still used to treat MRSA but in case of high prevalence of non-susceptible strains use linezolid or damptomycin. Now days broad spectrum antibiotics like ceftobiprole, dalbavancin is shows high affinity to kill Staphylococcus aureus. Several approaches are under investigation to replace antimicrobial therapy. Vancomycin is the glycopeptides antibiotics used to treat MRSA in late 2000. Due to increase in use of vancomycin, Staphylococcus increases the inhibition to vancomycin up to MIC 4. In some cases, treatment failure, when the concentration of vancomycin is within susceptible range. In 2010 FDA approved linezolid (Zyvox – Pfizer) for treatment of MRSA. Linezolid is the oxazolidinone antibiotics used in antimicrobial therapy against the Gram positive bacteria. It inhibits the protein synthesis by binding to mRNA. FDA approved for Vancomycin resistance bacteria,
MSSA [30-34].

The Physicians should obtain the reservoir sample from patients to treat the MRSA. After the investigation of strain and infection prescribe the antibiotic therapy. For soft tissue and skin infection we should recommend oral antimicrobial therapy. We should differentiate between methillicin resistance and methillicin susceptible *Staphylococcus aureus* and patient receive optional treatment. In order to rapid technique to cure MRSA, researcher should develop reliable ways for identification and characterization of MRSA. To develop economical health care services, researcher should efforts for infection control, efficient clinical treatment; reduce the methillicin resistance in bacteria. The limitations of antibiotic therapy are minimized by using new effective therapy. Researcher should develop a new diagnosis for MRSA. Further research is necessary for increase antimicrobial methods and non-antimicrobial methods use to cure MRSA.

**Vaccine development for MRSA**

Every year about 100,000 blood infections are causes by the MRSA. The pharmaceutical manufacturers are developing new antibiotics. Again after some time *Staphylococcus aureus* develop resistance against antibiotics. MRSA develop resistance against β lactams up to vancomycin. Few researchers are interested to replace this antimicrobial technology with Antimicrobial peptides or vaccines. The basic principle of the vaccine is introduce weak antigen of heat killed or attenuated disease organism in to the body. Immune cells create the response against the antigen. B cells secret antibody to neutralize antigen. B cells stores antigen–antibody iteration in the form of memory cells and host will be immune to letter infection.

StaphVax vaccine is the first vaccine developed in 1990 by using *Streptococcus pneumonia*. The antigen consist glycolipid present on the capsule of bacterium. This was unsuccessful. Merck develop a formula by using cell wall proteins but it withdraw in clinical trial phase III because of negative results. *Staphylococcus aureus* invading the Panton Valentine leukocidin (PVL) toxin in to blood stream to cause local skin abscesses. Target vaccine should be against the Panton Valentine leukocidin (PVL).

There are some antibodies are in market for bacterium. In case of MRSA, there is no any information about immune response characters. Some vaccine manufacturers like Novartis, GlaxoSmithKline are developed some antibodies, some are trying to develop passive
immunization but no one approved by FDA. The researcher Olaf Schneewind was study the role of Staphylococcus aureus protein A and develops one mutated protein A. This mutated protein is fails to bind the antigen and this trial also unsuccessful [35].

In 2009 a group of researcher found a girl with an orphan disease Job’s syndrome, which produces special type of thymus cell, $T_H$ 17 cell. $T_H$ 17 cell secrets interleukin 17. This interleukin 17 are the key for vaccine development against the staphylococcus aureus. They develop vaccine which stimulates the Interleukin 17 production [36]. This vaccine gives good result in animal models. Now researchers are working on vaccine development for cure CA-MRSA pneumonia and skin infection. Finally the vaccine developed against the CA-MRSA is the universal vaccine.

**CONCLUSION**

In developed countries antibiotics are widely used in animal feed. The same antibiotics runoff into contaminate stream. The excessive and unnecessary use of antibiotics are develop the resistance in staphylococcus aureus. The staphylococcus aureus contains SSCmec, which carry mecA gene. Since 1940 it develop resistance to penicillin, methicillin and now vancomycin. The long stay in hospital and treatment with fluoroquinolones are increase the risk of MRSA.

The current research have some limitations like, there is no any guideline for collection of culture, storage of sample, automation in reporting for minimizing variations in decision making process. To control the MRSA infection the physicians should obtain the reservoir samples, after the investigation of strain and infection prescribe the antibiotic therapy. In many cases we should recommend non antimicrobial therapy. Economical health care services is important for control any outbreak. The researcher should efforts for efficient and effective clinical treatment. The Interleukin 17 are increase the T cell activity against MRSA. The limitations of antibiotics therapy are minimized by using new effective therapy, use non-antimicrobial method and new diagnosis for MRSA.

**REFERENCES**