



**Karolinska
Institutet**

Department of Microbiology, Tumour and Cellbiology, MTC

Department of Molecular Medicine and Surgery

**Characterization of extracellular and surface bound
adherence proteins of *Staphylococcus aureus***

AKADEMISK AVHANDLING

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ABSTRACT

Staphylococcus aureus (*S.aureus*) is a pathogen causing infections. In recent years *S.aureus* formed resistance to most of known antibiotics on the market. It is today a global interest to find alternative treatment against multi drug resistant bacteria.

The course of an infection is a multifactorial process. The first step after invasion is bacterial adhesion. Specific binding proteins are expressed on the cell surface of *S. aureus* with ability to bind different matrix proteins e.g. fibronectin, fibrinogen, collagen, laminin and vitronectin. Immunization with *S.aureus* adhesion proteins, recombinant developed in *E.coli* (*Escherichia coli*) gave rise to specific antibodies, targeting binding proteins on the cell wall of *S.aureus* and could block bacterial binding *in vitro*. Trials *in vivo*, performed in experimental endocarditis model in rats, further proved a significant protection against infection after immunization.

To survive the environment after adherence and colonization, the invading microorganism need to express proteins interfering with the host immune system. Several extra cellular proteins expressed from *S.aureus* have a plurality of biological effects on the host. Examples of these proteins, investigated in this survey, are Efb (extra cellular fibrinogen binding protein) and Eap (extra cellular adherence protein). Efb bind fibrinogen thus interfering with primary fibrin formation. Efb also bind and inactivate crucial factors in the complement system. These benefits of *S.aureus* are believed to cause delayed wound healing, prolonged bleeding and accessed scar formation found in *S.aureus* infected wounds.

Eap has the ability to bind most of all known matrix proteins. It also serves several functions that interfere with the host immune system. This could be one explanation to the poorly functioning immunologic memory found after an *S. aureus* infection.

The hypothesis in this thesis is mainly based on two theories: Disturbed adhesion primarily affect the virulence of an invading microorganism. Immunization with recombinantly produced adherence proteins stimulate opsonization and make hidden virulence factors visible to the immune system and thus facilitate the ability to clear out infections. The proteins expressed are representative for most common species of *S. aureus*, irrespective of antibiotic resistance.

To mimic the clinical situation in experimental wound infection models it's important to use small inoculate. It is also important to obtain a strong adaptive immune response with circulating memory cells that consequently would protect against recurrent infections. In the modern way of thinking it is also essential to attack several steps in the course of infection. An experimental wound infection model was thus performed, infected with a minimal infectious inoculate. Multi component immunization was performed with four recombinant proteins, targeted at different steps in the infectious process. The results indicated a significant reduction of bacterial load in the immunized group and a more rapid clearing out ratio compared to the control group after infection challenge with *S.aureus*.

Conclusion: The present studies support the theory that it is conceivable to use immunizations with recombinant extra cellular and cell bound proteins as an alternative to prevent and further treat infections caused by multi resistant microorganisms.

Key words: *Staphylococcus aureus*, vaccination, surface bound adhesion proteins, extra cellular binding proteins, immunization.

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