

TITLE: EVALUATION OF BIOFILM FORMATION AND RELATED GENES IN DIFFERENT LINEAGES OF *Staphylococcus aureus* ISOLATES FROM RIO DE JANEIRO HOSPITALS

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ABSTRACT:

Biofilm formation by *Staphylococcus aureus* is associated with chronic infections and persistence in the hospital settings. However, there are few studies that evaluated the biofilm produced by *S. aureus* isolates presenting different genetic backgrounds and different methicillin resistance profiles. Therefore, this study investigated the biofilm formation and its related genes in different lineages of *S. aureus* isolated from Rio de Janeiro hospitals. A total of 62 isolates with well-characterized clonal profiles were selected, such as: USA100, USA200, USA300, USA400, USA600, USA800, USA1100, BEC, Hungarian, and EMRSA-15. Among them, methicillin-resistant *S. aureus* (MRSA) and methicillin-susceptible *S. aureus* (MSSA) representatives were included. The ability to form biofilm was investigated through a microtiter plate assay. All of the isolates were submitted to PCR analysis for detection of biofilm-related genes: *icaA*, *sasG*, *fnbA*, *fnbB*, *clfA*, and *clfB*. Presence and type of *agr* operon were determined by multiplex-PCR analysis, and its activity was assessed by the expression of δ -hemolysin through a phenotypic test. Selected isolates (seven) were tested for adhesion to fibrinogen, fibronectin, and collagen. Most of the isolates were moderate biofilm producers (47%) while strong, weak, and non-biofilm producers accounted for 18%, 27%, and 8%, respectively. All of the isolates investigated were positive for *icaA*, *clfA*, *clfB*, and *agr* genes, and there was a correlation between the *agr* type and the clonal complex of the isolates. The *sasG* gene was found in 82% of the isolates, but it was not detected in isolates of the USA200 and USA600 lineages. The *fnbB* gene was only detected in USA300, BEC, and Hungarian isolates. According to the phenotypic test, most of the isolates (87%) had an active *agr* operon. However, there was not a correlation between inactive *agr* operon and higher biofilm accumulation or clonal lineage. In general, MSSA isolates accumulated more biofilm than their MRSA counterparts. BEC isolates had the highest biofilm accumulation among the studied clones and presented all the genes searched. On the other hand, USA1100 isolates produced less biofilm than the other lineages. BEC and USA300 MRSA isolates were also able to bind higher concentrations of proteins than the other MRSA isolates evaluated. In conclusion, presence of biofilm-related genes was associated with the genetic background of the isolates and higher adhesion to proteins.

Keywords: *Staphylococcus aureus*, biofilm, biofilm-related genes, clonal lineages, methicillin resistance

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