Adaptation of the capacity to form biofilm in *Staphylococcus aureus* isolates during the course of human chronic bone and joint infections

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Introduction

- Prosthetic joint infection (PJI) is associated with high rates of **chronicity** and relapse (10-20% of cases).
- One of the major bacterial mechanisms is **biofilm** formation, within which bacteria are protected from antimicrobials and host immune response.
- The present study aimed to determine and compare the biofilm formation ability of 3 pairs of isolates collected in 3 different patients during the initial and recurrent BJI episodes.

Materials and Methods

- Three couples (SM, GM and MD) of methicillin-susceptible *S. aureus* (MSSA) strains collected from patients with persisting or relapse of BJI were tested. The biofilm formation capacity of the initial and recurrent isolates were compared using:
  - **Biofilm Ring Test™ assay** (EARLY KINETICS ADHESION)
    - using Brain Heart Infusion media (BHI) – incubation time: 0, 2, 4, 6 and 24h
  - **Crystal Violet assay** (MATURE BIOFILM CAPACITY)
    - using BHI + 1% glucose (BHlg) – incubation time: 24h
    - using a pool of human serum + 1% glucose (SERg) incubation time: 7, 14, 21 and 28 days
  - **Microfermentors assay** on glass spatula (DYNAMIC BIOFILM CAPACITY)
    - using BHlg - evaluated by plate count – incubation time: 24h
  - **Antibiofilmogram® assay** (BIOFILM MINIMAL INHIBITORY CONCENTRATION)
    - using BHI – incubation time: 4h

Results

Early biofilm formed of clinical *S. aureus* using the BioFilm Ring Test® technologie

- **Early step** of biofilm formation was similar between initial and recurrent strains

Crystal violet absorbance of clinical *S. aureus* in BHlg 1% glucose media

- **GM recurrent isolate revealed a higher capacity to form mature biofilm in BHlg**

Crystal violet absorbance of clinical *S. aureus* in SERg 1% glucose media

- **SM recurrent isolate revealed a higher capacity to form mature biofilm in SERg**

Biofilm formed on a glass spatula after 24h incubation in a microfermentor in BHlg 1%

- **GM and MD recurrent isolate revealed a higher capacity to form biofilm in dynamic model**

Conclusion

- Our results suggest that *S. aureus* PJI chronicization is **associated** with an in vivo bacterial adaptation/selection regarding **biofilm formation**.
- Biofilm formation differed from one couple to another, depending of the experimental conditions, suggesting different adaptation processes.
- In any case, the enhanced capacity of biofilm formation affect the recurrent strains compared to initial strain in each patient.