Titulo: Quantitative and structural analysis of biofilm produced by of multi-resistant *Corynebacterium striatum* isolated from nosocomial outbreak in Rio de Janeiro, Brazil.

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Abstract

Introduction: Corynebacterium striatum is a potentially pathogenic microorganism with the ability to produce nosocomial outbreaks. Additionally, C. striatum has been associated with increasing number of invasive infections. However, there are few studies focused on virulence factors that may contribute to elucidate the mechanisms concerned about health-care associated infections (HAIs) by C. striatum. The ability to produce biofilm is an important microorganism's defence ploy to fend off external agents. Recently our group reported a C. striatum nosocomial outbreak in an University Hospital at Rio de Janeiro, were most of strains were multidrug resistant. Starting from these results we initiated several studies to identify possible factors that may contribute to the persistence of C. striatum in the hospital environment. Aim: To investigate the production of biofilm by C. striatum on several abiotic substrates using quantification methods and electron scanning microscopy (ESM). Methods: Four different C. striatum clones isolated from nosocomial outbreak were investigated. The biofilm formation analysis was performed by quantification of CFU associated with different glass, thermanox slides and catheter fragments which were incubated with 10⁶ substrates: CFU/mL in Trypticase Soy Broth for 37°C/48h. After, the biofilm was extracted by abrasion and the number of CFU determined as previously described. Biofilm structure was analyzed by ESM. Briefly sections of the different substrates were fixed with 2.5% glutaraldehyde, post-fixed with 1% osmium tetroxide solution and dehydrated with ethanol. Subsequently, catheter segments were submitted to critical point drying with carbon dioxide, covered with 10nm gold layer and examined by ESM. Discussion of Results: C. striatum was able to adhere to all abiotic surfaces analyzed at different intensities. Additionally, C. striatum strains produced biofilm on different substrates including the polyurethane catheter at 48h post-incubation. The biofilm maturation was observed by the generation of a complex architecture with channels and pores that formed their three-dimensional structure, and also the presence of extracellular matrix. Conclusion: C. striatum ability to adhere to different substrates and also to produce biofilm may contribute to its persistence at the Hospital environment.

Key words: Corynebacterium striatum, Multi-resistant, biofilm, outbreak e analysis.

Agências de Fomento: FAPERJ, CAPES, SR2-UERJ.