Effect of sanitizers in *Candida parapsilosis* polymicrobial biofilms with *Staphylococcus aureus* or *Acinetobacter* sp.

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Summary

The *Candida parapsilosis sensu stricto* is one of non-*albicans Candida* species with the highest incidences in clinical infections, primarily related with the ability to growth in biofilms, especially in implanted medical devices. This study evaluated the ability of this yeast to interact with *Staphylococcus aureus* and *Acinetobacter* sp. in two-species biofilms, and their tolerance to different sanitizers. The mature 48 h monomicrobial and polymicrobial biofilms were developed and treated with 70% ethanol, 1% sodium hypochlorite, and with the commercial formulations of 1% polyhexamethylene biguanide and 1% glucoprotamin (Fácil 41 and Incidin Extra N, respectively), for 5 and 10 min, independently. After Dey-Engley broth addition and sonication, biofilm cells were serially diluted and plated on Sabouraud Dextrose Agar with chloramphenicol (0.05 mg/mL) and/or Brain Heart Infusion Agar with amphotericin B (0.025 mg/mL). The number of viable cells was estimated by CFU/mL counts. The results indicated that *C. parapsilosis sensu stricto* was able to synergistically associate with both bacteria, since polymicrobial biofilms presented a significantly higher number of bacterial cells compared to their own monomicrobial condition. Conversely, bacterial cells did not interfere on yeast biofilm formation. All chemicals significantly decreased the number of viable cells in the biofilms. Ethanol was the least efficient, with an elevated number of remaining cells (10^2 to 10^4 CFU/mL) after 10 min exposure. Hypochlorite was significantly more effective than ethanol, but viable cells (10^2 to 10^3 CFU/mL) also remained after the same exposure time. The biguanide was more effective compared to ethanol and hypochlorite, but 10^1 to 10^2 CFU/mL surviving cells were still detected in most biofilms after 10 min. Nevertheless, glucoprotamin was able to eliminate all yeast cells in mono- and polymicrobial, as well as all bacterial cells in monomicrobial biofilms. However, in the polymicrobial ones, viable bacteria (10^1 CFU/mL) were able to survive after 10 min exposure. These results revealed the ability of *C. parapsilosis sensu stricto* to synergistically interact with bacterial species of clinical importance, and the effectiveness of different sanitizers over these biofilms, suggesting 1% glucoprotamin as the most suitable for hospital cleaning. Importantly, they also indicate that none of these chemicals was able to completely eliminate biofilm cells, especially in 5 min treatments.

Keywords: *Acinetobacter* sp., *Candida parapsilosis*, Ethanol, Sodium hypochlorite, *Staphylococcus aureus*.

Funding agencies: Fundação de Amparo à Pesquisa no Estado do Rio Grande do Sul (FAPERGS) and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES).