**Biofilm activity of clinical oral isolates of Non-Candida albicans Candida species**

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**Aims:** Oral candidiasis is one of the most common opportunistic infections, strongly associated with Human Immunodeficiency Virus (HIV) and patients undergoing treatment for cancer or organ transplantation. Furthermore, many Non-Candida albicans Candida (NCAC) species, such as Candida glabrata, Candida parapsilosis and Candida tropicalis have emerged as significant oral pathogens and are often associated with the formation of biofilms. These biofilms have medical significance once they exhibit high resistance to host defences and antimicrobial agents. Previous studies have focused NCAC species biofilms in terms of total biomass, however little is known regarding the biofilm activity of NCAC species. Thus, considering that the biofilm activity is a requirement in Candida infections, the main goal of this work was to study biofilms formed by different clinical isolates of C. parapsilosis, C. tropicalis and C. glabrata in terms of their metabolic activity.

**Methods:** A total of 6 oral isolates of C. parapsilosis, C. glabrata and C. tropicalis were studied as well as references strains of each species. Biofilms were formed in 96-well microtiter plates using inoculums of 1x10^7 cells/ml. Metabolic activity was determined measuring the XTT reduction after 48 hours. Final absorbance values were standardized per 1x10^8 biofilm viable cells (measured by CFU counting of biofilm cells).

**Results:** The results obtained showed that, biofilm metabolic activity is strain and species dependent. Nevertheless, C. glabrata biofilms presented, in average, lower metabolic activity compared with C. tropicalis and C. parapsilosis. However, it was noticed that C. glabrata biofilms presented, in average, higher number of viable cells compared with C. tropicalis and C. parapsilosis. Comparing the three species it was possible to infer that C. tropicalis presented a biofilm relative activity approximately 10 times higher than C. parapsilosis and 100 times higher than C. glabrata. Moreover, intrinsic differences in metabolic cellular activity among strains either in planktonic or biofilm form were also found. Specially, it was noticed that C. glabrata cells presented,
lower values of cellular metabolic activity compared with \textit{C. tropicalis} and \textit{C. parapsilosis} cells.

\textbf{Conclusions:} In conclusion it can be highlighted that biofilm metabolic activity is strain and species dependent. Furthermore, \textit{C. glabrata} biofilms presented lower metabolic activity than \textit{C. tropicalis} and \textit{C. parapsilosis} biofilms, despite the higher number of viable cells. Moreover, the results also demonstrate that there are intrinsic differences in cellular metabolic activity of planktonic and biofilm cells of the different species and strains.