

STRUCTURAL ANALYSIS OF THE EXTRACELLULAR MATRIX MATRIX OF *Candida albicans* BIOFILMS

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Candida albicans form biofilms in central venous catheter (CVC) and such microbial construction are frequently related to invasive infections. An important feature of the fungal biofilm, and required to be defined as such, is the presence of a polymeric extracellular matrix (ECM) embedding the cells. It is technically challenging to observe the ECM. By electron microscopy methods it is imperative to have an optimal sample preservation. Other techniques, such as light and fluorescence microscopy, do not have the resolving power to reveal its details. Our purpose was to unravel the structure and composition of *C. albicans* biofilm and its ECM after *in vitro* growth in several media and over cvc sections. To achieve our goal, we allowed *C. albicans* biofilms to grown in different carbon sources, analyzed the ECM composition by mass spectroscopy, and observed the fine detail of *C.albicans* biofilms by scanning electron microscopy after sample processing by plunge freezing and freeze substitution. After 24h, fungal growth and total biomass was higher when glucose was the sole carbon source. Independent of growth condition, Mass spectrometry revealed that a similar ratio of glucose, mannose, arabinose, glucosamine and galactose compose the ECM. All conditions tested showed germ tube formation and pseudohypha. Interestingly, glucose alone induced a higher hyphae formation. Scanning electron microscopy after cryoprocessing of samples showed the details of EMC and surrouind cells. This EMC on lactate grown *C. albicans* appeared as a heavily cohesive strucutre, whereas EMC in glucose cultures had a fibrillar arragement. On a biofilm, its construction and structuration depends on the EMC. Our results shed light on EMC formation and provide evidence that is a consequence of the carbon source available to *C. albicans*. Since body sites of infection have variable levels of glucose and lactate, a bias toward one or another might dictates the level of organization of biofilm and also fungal morphology. This is important since a structured biofilm and hyphae are phenotypes associated with cells resistant to antifungal and invasive infections.

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