

TITLE: STANDARDIZATION OF SPECIFIC PROBES FOR IDENTIFICATION OF *CANDIDA DUBLINIENSIS* IN A DNA MICROARRAY PLATFORM

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ABSTRACT

Opportunistic systemic fungal infections, such as candidemias, affect immunocompromised patients and have shown high mortality and morbidity rates. Rapid identification of the causative pathogen is essential for a proper treatment and a good prognosis. Conventional microbiological diagnosis may present low sensitivity and require a long time, due to the difficulty of fungal cultivation and morphological identification. In this context, molecular diagnostic techniques stand out for their high sensitivity and specificity, and for allowing a rapid identification. The DNA microarray technique has been studied for molecular detection of fungi and other organisms through genus / species-specific probes. During the standardization of specific probes that differentiate fungal species on a DNA microarray platform, we obtained non-specific results, with presence of cross reactivity between two species of *Candida*, since positive samples for *Candida albicans* reacted with the probes of *Candida dubliniensis*. Thus, the aim of this study was to standardize new specific probes designed for *C. dubliniensis*, in our DNA microarray platform. Twelve new probes for *C. dubliniensis* were design and tested on the DNA microarray platform using DNA from culture isolates of *C. albicans* and *C. dubliniensis*. A total of 65 isolates from clinical samples identified by the conventional technique as *C. albicans* (42) and *C. dubliniensis* (23) were obtained from the microbiology laboratory of the Clinical Hospital of University of Campinas. After DNA extraction, ITS region of fungal ribosomal DNA was amplified using biotinylated primers. Subsequently, the PCR product was applied to the DNA microarray platform and sequenced. Seven new specific probes for *C. dubliniensis* were obtained, which did not show cross reactivity with *C. albicans*. Of the 65 samples, 45 were identified as *C. albicans*, and 20 as *C. dubliniensis*, with 100% correspondence between the result in the DNA microarray platform and DNA sequencing. These results indicate that our DNA microarray platform has high specificity for *C. albicans* and *C. dubliniensis* species, and may help in the diagnosis of patients with candidemia.

Keywords: *Candida albicans*, *Candida dubliniensis*, molecular diagnosis, DNA microarray

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