

Title: VALIDATION OF METHODOLOGY FOR SEMI - QUANTITATIVE ANALYSIS BY RT-PCR GENE ERG11 IN *CANDIDA ALBICANS*

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Candida albicans is an important opportunistic infection agent that can cause serious diseases in immunocompromised patients including cancer patients, transplant recipients and patients who received any immunosuppressive therapy in general. The infection spectrum is quite wide and the treatment line and fluconazole based on a triazole derivative with potent antifungal activity against *C. albicans*, and amphotericin B, an antifungal polyene. However, resistance to these drugs can lead to treatment failures and thus become a clinical problem for such infections. The test was validated in a standard strain of *C. albicans* (ATCC 10231). For this, the strain was grown in the presence and absence of subinhibitory concentrations of amphotericin B and fluconazole as determined by the microdilution broth method. Total RNA was extracted by Trizol® method and performed cDNA synthesis by reverse transcriptase samples of standard strain of *C. albicans* grown in the above conditions. From the synthesized cDNA, the expression of the *ERG11* gene was evaluated by PCR reaction in real time using the fluorophore Sybr Green®. RT-PCR primers were designed to detect the level of mRNA expression of the *ERG11* gene. Primer efficiency curve was made to determine the best amount to be used thereof. The determination C_T possible to detect the presence of the target mRNA and validate the amplification. Results with undefined C_T were considered valid and reported as negative for the presence of target DNA. Results with $C_T \leq 35$ were reported as positive for the presence of the target mRNA. The higher the C_T value, the lower the number of copies of the amplified gene. More tests are being performed to verify the influence of azoles and polyenes in the expression of the gene *ERG11*.

Keywords: *Candida albicans*, *ERG 11*, resistance, RT –PCR

Promotion agency: CAPES, FAPEMIG