Title: Amino acid substitutions in ERG11 gene in *Candida* sp species of clinical isolates with azoles susceptibility

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ABSTRACT

In the last decades, there have been many cases of resistance to antifungal agents used in the prophylaxis and treatment of infections caused by *Candida* spp. The molecular mechanisms responsible for the resistance development in *Candida* species on the main class of antifungal used, the azoles, can be attributed to mutations and increased expression of genes encoding enzymes responsible for the biosynthesis of ergosterol. The search for mutations in the ERG11 gene which encodes a key enzyme in the pathway of ergosterol biosynthesis (lanosterol 14α-demethylase), can provide better understanding of the molecular mechanisms involved in the resistance to antifungal agents and epidemiological research of *Candida* species. Based on this, the objective was to identify mutations in the coding region of ERG11 gene in clinical isolates of *Candida* with reduced susceptibility to azoles, reference strains the American Type Culture Collection (ATCC) were also used in the analyzes. To do so, the coding regions of the gene ERG11 from clinical isolates of *Candida* with resistance profile established for fluconazole, itraconazole and voriconazole were amplified and sequenced. The data revealed 20 silent mutations in the gene ERG11, three of which had not been reported in *C. glabrata* (C108G, C423T and A1581G); and 2 missense mutations not reported in the literature for *C. krusei* A497C (S166Y) and G1470A (G524R). The G524R mutation was identified in both the *C. krusei* ATCC 6258 strain as in isolates (*C. krusei*) dose-dependently sensitive, demonstrating that the presence or absence of this mutation may not be enough to predict the susceptibility in this yeast species. The S166Y mutation was found in an isolate dose dependent sensitive for voriconazole, however further investigations are needed to refine the results, so that resistance to azoles in this species can be explained. This study may be useful in epidemiological research and also in the exploration of new bioactive molecules with antifungal activity based on genetic and molecular characterization of isolates, particularly for resistant *Candida* species.

Keywords: yeast, *Candida krusei*, voriconazole, 14α-demethylase, S166Y

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