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RELATION BETWEEN BIOFILM FORMATION AND MTL GENES IN CANDIDA TROPICALIS ISOLATED FROM HOSPITALIZAD PATIENTS.

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INTRODUCTION: Sexual reproduction in *Candida tropicalis* is regulated by genes encoded in the mating type-like (MTL) loci. Homocygotic MTLa cells are able to mate with homocygotic MTLalpha cells forming MTLa/alpha cells, conjugation being possible during infection in humans. The *Candida albicans* biofilm is an advantage for conjugation, with high levels of sexual pheromones, facilitating mating of homocygotic opaque cells. **The aim of this study** is to determine whether a correlation between mating type and biofilm-forming ability exists in *C.tropicalis*.

METHODS: Biofilm-forming ability of 28 *C. tropicalis* clinical isolates (urinary tract 15, respiratory tract 7, others 6) in RPMI-1640 medium at 37 ºC for 24 and 96 h using a 96-well microtiter polystyrene plate were determined by OD_{490nm} and by Slime Index (SI) (Biofilm OD_{492nm}/Growing culture OD_{492nm} %). Biofilm microscopic observations to determine filamentation level were made. All experiments were performed at least 3 times. MTL genes were studied by PCR: strains were incubated in Sabouraud broth for 24 h at 37 ºC, cells were removed from the medium, and RNA was isolated; 500 ng of RNA was used for subsequent cDNA generation. PCR was performed by using gene-specific primers. Statistic studies: SPSS11.5 for windows and Mann-Whitney test for statistic significance were used.

RESULTS: (i) Most strains (72.4%) showed heterozygosis for MTL (MTLa/alpha) and 8 strains (27.6%) strains were homozygotic for MTL (7 strain MTLa/-; 1 strain MTL-/alpha). **(ii)** Biofilm-OD_{490nm} in the group of homozygotic cells was lower than in the heterozygotic ones for the periods of time studied. However, no statistical significance was found (0.40 vs 0.49 for 24 h, p=0.53; 0.39 vs 0.48 for 96 h, p=0.26). **(iii)** The SI of biofilm in the group of homozygotic cells was lower than in the heterozygotic ones for the periods of time studied. No statistical significance was found (44.34 vs 51.88 for 24 h, p=0.43; 38.49 vs 44.85 for 37 ºC and 96 h, p=0.33). **(iv)** The highest level of filamentation was reached mostly for 96 h. The homozygotic cells presented a lower level of filamentation than the heterozygotic ones.

CONCLUSIONS: Our study shows high presence of MTL homozygous *C. tropicalis* in clinical isolates. The MTL heterozygous strains of *C.tropicalis* had higher filamentation and biofilm-forming ability than the homozygotic ones. Once biofilm is formed by heterozygous non-mating competent cells, a small existing homozygotic population can switch to opaque form, mating being possible with the subsequent formation of recombinant cells. Future studies with more strains must be done to investigate whether statistical significance can be achieved and to assess clinical significance relating *Candida* genetic interchange with increased pathogenicity and resistance.

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