ABSTRACT

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Title of diploma thesis: Detection of Sap2 in the secretome of *Candida albicans* cell wall and secretory mutants

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Study program: Pharmacy

Backgound: The aim of this diploma thesis was to search for *C. albicans* proteins involved in the secretion of the secreted aspartyl proteinase 2 enzyme (Sap2) evaluating the ability to degrade BSA (bovine serum albumin) as a source of nitrogen in several cell wall and secretory mutants of *C. albicans.* The work was carried out at the Department of Microbiology II, Faculty of Pharmacy, Complutense University of Madrid.

Methods: The supernatant samples of several *Candida albicans* mutants were tested by SDS-PAGE electrophoresis and stained. Bands corresponding to BSA were observed and compared to controls. The other method was counted with 96-well plate.

Results: The correlation between optical density and degradation of BSA was observed. Some mutants with disability to degrade BSA were found in a pilot screening of the ability to degrade BSA using 96-well plate method. That fact was confirmed by SDS-PAGE electrophoresis. *C. albicans* mutants showing this defect, that was proved by both methods, were *ecm33*Δ, *kex2*Δ, *ypt72*Δ, *orf*.19.1567Δ and *pbs*2Δ.

Conclusions: The mutants with disability to degrade BSA as a sole source of nutrients gradually died in liquid YCB-BSA medium and their OD were considerably lower than in the other cases. It was possible to confirm this absence of degradation by SDS-PAGE electrophoresis. Assumption was that mutants with disability to degrade BSA had problems with Sap2 secretion. This fact was confirmed by western blot using Sap2 antibody.

KEYWORDS: *C. albicans*, mutant, optical density, bands, electrophoresis, supernatant, BSA, degradation, Sap2, cytoplasmic extract.