Effect of antifungal agents on Non-*Candida albicans Candida* species enzymes secretion

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The infective ability of *Candida* species depends on specific virulence mechanisms that confer the ability to colonize host surfaces, to invade deeper host tissue or to evade host defences. During the pathogenic process many virulence attributes may be involved including, production of extracellular proteases and haemolytic activity. Nevertheless, *in vitro* studies have indicated that antifungal agents could be able to influence the enzymatic activity of *Candida* species. Therefore, the purpose of this work was to investigate the action of antifungals on proteinase and haemolytic activity of Candida species.

This study was conducted with *C. albicans* (1), *C. glabrata* (4), *C. parapsilosis* (5) and *C. tropicalis* (6) recovered from different body sites (blood, oral, vaginal and urinary tract). Four reference strains of *C. albicans* ATCC 90028, *C. glabrata* ATCC 2001, *C. parapsilosis* ATCC 22019 and *C. tropicalis* ATCC 750 were also examined. The susceptibility to fluconazole and amphotericin B was determined by the microdilution test in order to allow the determination of the minimal inhibitory concentrations (MIC) and the maximum antifungal concentration (MAC). Then, the proteinase and hemolytic activity was determined for yeasts grown at MIC and MAC.

It was observed that all *Candida* species assayed were sensible to both antifungal agents. Concerning the antifungal effect on enzymatic activity of Candida species, *C. parapsilosis* from candiduria presented a decreased proteinase and haemolysin activity for both MIC and MAC of both antifungal agents. Moreover, the other species presented differences in terms of production of proteinase and haemolysin at MIC and MAC. *Candida albicans* reference strain presented lower proteinase activity at MIC of fluconazole (46.7%) but presented higher activity for MAC (61.9%) in comparison to the control (60%). Furthermore, regarding haemolysin activity there were isolates that expressed high levels of enzymes in the presence of both antifungals such as: *C. glabrata* from urine and from vaginal tract; and *C. tropicalis* from urine. Conversely, some clinical isolates, presented low levels of enzymatic activity after contact with the antifungal agents, such as: *C. albicans* (oral isolate); *C. glabrata* (oral isolate and vaginal isolate); *C. parapsilosis* (from urine) and also *all C. tropicalis* except one urinary isolate.

It was possible to conclude that the proteinase and haemolysin activities were strain and species dependent and no correlation was found among activity profile and the site of isolation. Moreover, fluconazole and amphotericin B were able to influence the tested *Candida* species enzymatic activity.

Keywords: Candida; Fluconazole; Amphotericin B; Proteinase; Haemolytic activity