TITLE: RITONAVIR AND LOPINAVIR ACTION AGAINST *Phialophora verrucosa* CONIDIA AFTER INTERACTION WITH HUMAN MACROPHAGES

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ABSTRACT:
Phialophora verrucosa is a filamentous dematiaceous fungus and an etiologic agent of chromoblastomycosis (CBM), phaeohyphomycosis and mycetoma. These diseases are difficult to treat since the available antifungal drugs have several drawbacks, such as side effects and resistance. Thus, new active substances are crucial to enhance fungal infection treatments. Human immunodeficiency virus aspartic peptidase inhibitors (HIV-PIs) raise as potential drugs for their effects. In recent years, our group has demonstrated that HIV-PIs were able to inhibit other CBM fungi growth, morphogenesis and their interaction with the host cells. The aim of this study was to evaluate the effects of ritonavir and lopinavir (HIV-PIs), apart or in combination, on *P. verrucosa* interaction with human macrophage. First, a colorimetric assay was performed using MTT for cytotoxicity assessment after treatment of macrophages with these HIV-PIs. Then, *P. verrucosa* conidia (1×10^6) were incubated for 1 h with a human macrophage cell line THP-1 (2×10^5) and treated for 20 h with non-cytotoxic concentrations of ritonavir and lopinavir, apart or in combination. In order to investigate their killing properties, the macrophages were lysed and the number of ingested conidia was determined using the colony-forming unit (CFU) assay. Conidia treated with ritonavir (25 μM) and lopinavir (100 μM) were more susceptible to macrophages, since only around 30% and 25% of viable conidia were detected, respectively. In combination, ritonavir (12.5 μM) and lopinavir (50 μM) were more effective and inhibited around 90% of *P. verrucosa* conidia viability. This study corroborated that *in vitro* HIV-PIs are able to reduce fungal burden, an important step to considering antifungal drug efficacy. Additional cellular interaction experiments are in progress in our laboratory to determine the cytokine profile produced by THP-1 macrophage, as well as the effect of these HIV-PIs *in vivo*.

Keywords: *Phialophora verrucosa*, HIV aspartic peptidase inhibitors, cellular interaction, antifungal drugs

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