Importance of Sphingolipids Biosynthetic Pathway for *Pseudallescheria boydii* Growth

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Introduction. *Pseudallescheria boydii* is an emerging pathogen able to cause a wide-spectrum infection ranging from mycetoma to invasive infections in immunocompromised patients (1). Sphingolipids are abundant components of membranes in fungal cells, playing a variety of roles such as heat stress response, signal transduction, endocytosis and apoptosis (2). Particularly, glucosylceramides (GlcCer) have been studying by our group during the last decades in *Pseudallescheria/Scedosporium* complex, being associated to fungal growth and pathogenesis (3). **Objectives.** This study aims to describe *P. boydii* GlcCer synthase gene and evaluate the enzyme activity during fungal germination and growth. Besides, the importance of sphingolipid biosynthesis was evaluated by inhibiting different steps of the pathway. **Material and Methods.** The present study used molecular biology approaches and cellular biology and biochemistry assays in order to amplify and express *P. boydii* GlcCer synthase gene to check its activity, as well as to evaluate the importance of sphingolipids for *P. boydii* growth by using inhibitors of different steps of biosynthetic pathway. **Results and Discussion.** For the first time, *P. boydii* GlcCer synthase gene was amplified by polymerase chain reaction and its nucleotide composition was sequenced and identified. Using *in vivo* radiolabeling assay, it was observed that GlcCer synthase is expressed during the whole germination process at 37°C while it is only expressed at the beginning of the process at 30°C. Preliminary studies showed that inhibiting different steps of *P. boydii* sphingolipid biosynthesis impairs fungal growth. Treatment with Myriocin, for example, an inhibitor of the first step of sphingolipid biosynthesis, resulted in a decrease of *P. boydii* germination at 37°C, but not at 30°C. **Conclusions.** These data suggest that sphingolipids play an important role for fungal growth at host condition and indicate a difference in sphingolipids biosynthesis in different conditions.

References:
1. Cortez et al. Clin. Microbiol. Rev. 21 (1): 157-197. 2008.
2. Heung et al. Infect Immun 74(1), 28-39. 2006.
3. Rollin-Pinheiro et al. PLoS ONE. 9(5):e98149. 2014.

Key words: Sphingolipids; *Pseudallescheria boydii*; Glucosylceramide.