Proteomic analysis of Rat Lungs infected by Cryptococcus deuterogatti (R265)

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Cryptococcosis is the infection caused by yeasts of the *Cryptococcus neoformans* and *Cryptococcus gattii complexes*, with the *C. gattii complex being* the most virulent. In general, cryptococcosis is characterized by infection in specific sites, presenting tropism mainly in the lungs and brain. Epidemiologically, it is considered one of the most frequent invasive fungal diseases (IDD) in humans, making the molecular understanding of the infectious process essential. Although host immunity helps fight pathogens, dysregulation of related responses can have a deleterious effect on the host. The induction of inflammatory responses by the invasion of pathogens can lead to the promotion of hemostasis and coagulation processes. Regarding those potential effects, in this work adult male Wistar rats were experimentally infected by *C. deuterogattii* R265 and were monitored during infection. The proteomic data were obtained by collecting the infected lungs and the proteins present there were extracted, digested and analyzed by mass spectrometry using the *Multidimensional Protein Identification Technology* (MudPit) technique. The mass spectrometry results, were analyzed using PatternLab software (Carvalho et al. 2012) to identify the modules of differentially identified proteins (TFold) and AAPV (Venn diagram approximately proportional to the area). Comparisons between the treated group (*C. deuterogattii* R265 3 days after inoculum) and control (no infection) were performed using spectral count data in a *TFold module* with the *p* -value ?0.05. Proteins that were not detected in at least three of the six runs per condition were not considered. The Database for Annotation, Visualization and Integrated Discovery (DAVID) was used to characterize the differential proteome, and metabolic pathway analyses were performed according to KEGG and Reactome. The monitoring of the rats demonstrated a procoagulatory profile during infection, since over the days the platelet aggregation induced by ADP increased significantly, on the other hand, the amounts of blood platelets were significantly decreased. The proteomic analysis identified 1,508 proteins, 627 of which were exclusive to group I and 476 were exclusive to the control, and of the 405 proteins present in both conditions, 10 proteins were up-regulated and 63 down-regulated. Through gene set enrichment analysis (GSEA) we observed an enrichment of hemostasis pathways and platelet degranulation suggesting a procoagulant state in the lungs of infected rats. Thus, the results suggested that rats experimentally infected by *C. deuterogatti* R265 present a molecular and biochemical procoagulation profile, which may be deleterious and a new identified feature of pathogenesis.

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