**TITLE:** MACROPHAGE ACTIVATION AND CELL DEATH DURING INTERACTIONS WITH *Paracoccidioides brasiliensis* AND *P. lutzii* YEASTS.

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**ABSTRACT:**
Paracoccidioidomycosis is a systemic granulomatous infection caused by thermodimorphic fungi of *Paracoccidioides* genus. Phylogenetic analysis have supported the occurrence of at least two species: *P. brasiliensis* and *P. lutzii*. Recently, it was shown that isolates from both species induce different patterns of disease, suggesting lower pathogenic potential of *P. lutzii*. Since macrophages have crucial role in *Paracoccidioides* infection control and host immune response, this study aimed to investigate macrophages activation and cell death upon interaction with yeasts of *P. brasiliensis* and *P. lutzii*. Macrophage-like J774.16 cells were infected with non-opsonized *P. brasiliensis* (Pb18 isolate) or *P. lutzii* (Pb01 isolate) yeasts in 10:1, 5:1 and 1:1 (yeast:macrophage) ratios for 24 hours. Analysis of phagocytosis and cell adhesion was performed using flow cytometry or giemsa staining. Fungicidal activity and production of nitric oxide in culture supernatants were also evaluated after 18h of IFN-gamma stimulation. Macrophage cell death upon interaction with yeasts was determined by flow cytometry. Yeast plasma membrane and cell wall crude protein extracts were analyzed by SDS-gel electrophoresis. Levels of *P. lutzii* phagocytosis and cell adhesion were higher than those of *P. brasiliensis*. Increased fungicidal activity against *P. lutzii* isolates was detected. Both species induced macrophage cell death, but lower levels were identified with low proportion (1:1) of *P. lutzii* infection. Production of nitric oxide was not detected in any conditions. Electrophoretic profile of cytosol and cell wall proteins did not show significant variations among species. Our data suggests that macrophages have higher capacity of recognition and killing of *P. lutzii*. Besides, *Paracoccidioides* infection induces macrophage cell death, which is higher during *P. brasiliensis* interaction and can explain its increased pathogenicity.

**Keywords:** *Paracoccidioides*, macrophages, cell death.

**Development Agencies:** FAPESP, CAPES, CNPq.