

“The plant lectin ArtinM activates neutrophils during in vitro infection with *Paracoccidioides brasiliensis*.”

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Abstract

Paracoccidioidomycosis is a systemic mycosis of Latin America, occurring especially in Brazil, Argentina, Colombia, Ecuador and Venezuela. The disease is caused by the dimorphic fungi *Paracoccidioides brasiliensis* and *Paracoccidioides lutzii* and is an important health problem among agricultural workers. Both innate and adaptive immune responses are fundamental for disease control, and neutrophils are described to have a role in restraining the infection. Currently, the antifungal chemotherapy for the disease is limited by toxic side effects, prolonged treatment and relapses. The lectin ArtinM, extracted from the jackfruit seeds (*Artocarpus heterophyllus*), shows immunomodulatory activity against several pathogens. The administration of ArtinM to mice leads to the development of a Th1 immune response, which reduced the fungal load and protected mice against *P. brasiliensis*. We aim to evaluate the role of ArtinM in human neutrophils, stimulated *in vitro* with *P. brasiliensis* yeasts. In our study, using human neutrophils, ArtinM was able to induce the production of the inflammatory cytokines TNF- α , IL-8, IL-1 β , both alone and combined with the fungus. The immunostimulatory effect of the lectin was balanced by the production of IL-10 by neutrophils, which would collaborate to restrain the immunopathology *in vivo*. Flow cytometry analysis has confirmed that ArtinM is a neutrophil activator during *P. brasiliensis* infection, since the lectin induced an increase in CD54 expression. These preliminary results might support the possibility of using ArtinM lectin as an adjuvant therapy for *P. brasiliensis* infection.

Keywords: *Paracoccidioides brasiliensis*, Neutrophil, ArtinM

Introduction

Paracoccidioidomycosis (PCM), the most prevalent systemic mycosis in Latin America^[1], is caused by the thermo-dimorphic fungi *Paracoccidioides brasiliensis* and *P. lutzii*. Resistance to PCM infection is associated with a strong cell-mediated immune response, including the activation of neutrophils.^{[2][3][4]}

Currently, the antifungal chemotherapy for PCM is limited by factors like toxic side effects, prolonged treatment and relapses^[5]. In response to these challenges, studies are now focusing on the modulation of the immune response to better deal with fungal infections, including PCM.

The plant lectin ArtinM is a potent activator of cells of the immune system, including neutrophils^[6]. Immunization of mice with ArtinM reduces the fungal burden and protects mice against PCM^[7]. In our study, we aim to evaluate the role of ArtinM in human neutrophils, stimulated *in vitro* with *P. brasiliensis* yeasts.

Results and Discussion

Polimorfonuclear neutrophils (PMN) were obtained from peripheral blood from healthy individuals and cultured with different concentrations of ArtinM, in the presence or not of *P. brasiliensis* yeasts. The supernatant was used to evaluate cytokine production through ELISA. ArtinM was able to activate human neutrophils, leading to production of IL-1 β , TNF- α , and IL-8. Notably, the production of pro-inflammatory

cytokines occurred both in the presence of the lectin alone or combined with the fungus. We also found that the effect of the lectin was balanced by the production of IL-10, which could collaborate to restrain the immunopathology. Flow cytometry analysis has confirmed that ArtinM is a neutrophil activator during *P. brasiliensis* infection, since the lectin induced an increase in CD54 expression, both alone or in the presence of fungal cells.

Conclusions

These preliminary findings might support the possibility of using ArtinM lectin as an adjuvant therapy for *P. brasiliensis* infection.

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