TLR4 Recognizes *Pseudallescheria boydii* Conidia and Purified Rhamnomannans

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*Pseudallescheria boydii* (*Scedosporium apiospermum*) is a saprophytic fungus widespread in the environment, and has recently emerged as an agent of localized as well as disseminated infections, particularly mycetoma, in immunocompromised and immunocompetent hosts. We have previously shown that highly purified \(\alpha\)-glucan from *P. boydii* activates macrophages through Toll-like receptor TLR2, however, the mechanism of *P. boydii* recognition by macrophage is largely unknown. In this work, we investigated the role of innate immune receptors in the recognition of *P. boydii*. Macrophages responded to *P. boydii* conidia and hyphae with secretion of proinflammatory cytokines. The activation of macrophages by *P. boydii* conidia required functional MyD88, TLR4, and CD14, whereas stimulation by hyphae was independent of TLR4 and TLR2 signaling. Removal of peptidorhamnomannans from *P. boydii* conidia abolished induction of cytokines by macrophages. A fraction highly enriched in rhamnomannans was obtained and characterized by NMR, high performance TLC, and GC-MS. Preparation of rhamnomannans derived from *P. boydii* triggered cytokine release by macrophages, as well as MAPKs phosphorylation and I\(\kappa\)B\(\alpha\) degradation. Cytokine release induced by *P. boydii*-derived rhamnomannans was dependent on TLR4 recognition and required the presence of non-reducing end units of rhamnose of the rhamnomannan, but not O-linked oligosaccharides from the peptidorhamnomannan. These results imply that TLR4 recognizes *P. boydii* conidia and this recognition is at least in part due to rhamnomannans expressed on the surface of *P. boydii*.

Word Keys: rhamnomannans, Toll-like receptor, *Pseudallescheria boydii*

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