TITLE: DEVELOPMENT OF A MOUSE MODEL REPRODUCING THE PATHOPHYSIOLOGY OF SPOROTRICHOSIS TO EXPLORE VIRULENCE AND IMMUNOGENICITY IN *SPOROTHRIX GLOBOSA.*

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ABSTRACT:
Sporotrichosis is a chronic cutaneous or subcutaneous fungal infection that occurs in humans and animals after a traumatic inoculation of *Sporothrix* propagules. Molecular phylogenetic analysis revealed that the classical agent *Sporothrix schenckii* now comprises several molecular siblings clustered in a clinical clade with *S. brasiliensis*, *S. globosa*, and *S. luriei*, whereas non-pathogenic *Sporothrix* (e.g. *S. chilensis*) occurs at a large genetic distance in an environmental clade. *Sporothrix brasiliensis* is by far the most virulent agent followed by *S. schenckii s. str.* and *S. globosa*. Despite displaying differential virulence levels, *S. brasiliensis* and *S. globosa* emerge as important threats to human health in Brazil and China, respectively. Therefore, the aim of this study was to evaluate the virulence profiles of *S. globosa* (n=7) globally distributed (South America, Europe, and Asia), compared with *S. brasiliensis* (n=1) and *S. schenckii s. str.* (n=1). We describe here two mice models in which *Sporothrix* spp. caused clinical pictures varying from a localized lesion to disseminated sporotrichosis affecting several organs and systems. For virulence characterization, BALB/c mice were divided into 20 groups (one group for each *Sporothrix* isolate and two negative control group). Mice were infected with $5x10^6$ yeast cells/animal following a subcutaneous or intravenous route. After the challenge, *S. globosa* isolates were classified as non-virulent or presenting low, medium or high virulence, based on induction of death of infected animals, the fungal burden in the organs evaluated, and weight loss. We demonstrated that the route of infection is important for the manifestation of sporotrichosis. The CFU showed dissimilar results from each route of infection (subcutaneous and intravenous) and histopathological findings revealed tissue damage in nearly all isolates, including in *S. globosa*, a species previously assumed to be non-virulent. For immunogenic characterization, whole cell extracts of *Sporothrix* yeasts (Ss226 *S. brasiliensis* and Ss126 *S. schenckii*) were used in western blot analysis. A glycoprotein of 70 kDa was the main antigen recognized by IgG circulating in the sera of infected mice. Our results revealed that the route of infection is decisive to the virulence. Understanding how *Sporothrix* infect mammals and how mammals respond to infection could ultimately lead to new therapeutic strategies to help control the emergence of *Sporothrix* species.

Keywords: *Sporothrix*, *Sporothrix globosa*, sporotrichosis, virulence, murine model.

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