

Sporothrix brasiliensis infection of domestic cats in the Brazilian sporotrichosis hyperendemic region

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ABSTRACT

Microbial interactions have the potential to influence a patient's diagnosis, prognosis, and therapy. Sporotrichosis, caused by *Sporothrix brasiliensis*, is a hyperendemic neglected zoonosis in Brazil. In a sporotrichosis hyperendemic area of Brazil, four pairs of clinical isolates of *Sporothrix* were obtained from four ill cats (CIM01-CIM04, two isolates per animal), highlighting the potential of coinfection. In mycological cultivation, each of the pair's isolates had different pigmentation, and they were labelled "Light" or "Dark" for low and high pigmentation, respectively. By ELISA and FACS quantification, dark isolates responded strongly with monoclonal antibodies to melanin ($p < 0.05$), and immunofluorescence revealed a ring pattern with certain sections having stronger punctuated staining at the cell wall. Light isolates, on the other hand, responded less strongly, with fewer and more distinct punctuated labelling at the cell wall. *S. brasiliensis*, MAT1-2 idiomorph, was found in all isolates by PCR. Phylogenetic analysis of the β -tubulin and

calmodulin genes revealed all eight isolates in the same cluster as those from the Brazilian hyperendemic region. These strains were also tested for their capacity to trigger cytokine production in human PBMCs (Peripheral blood mononuclear cells). The cytokine profiles of the CIM01 and CIM03 Light and Dark isolates were similar to the control strain, while CIM02 and CIM04 reacted differently ($p < 0.001$), showing that variations in the isolates' surfaces can alter host-fungus interaction. Amphotericin B, terbinafine, caspofungin, micafungin, itraconazole, fluconazole, and voriconazole MICs (CLSI M38-A2/M27-A3) were determined. To at least one of the antifungals tested, pairwise comparisons revealed different MICs between *Sporothrix* Light and Dark isolates, higher than at least two-fold dilutions. Isolates from the same pair showed differences in fungistatic or fungicidal pharmacological action, particularly following exposure to itraconazole. Because the phenotypic features of *S. brasiliensis* Light and Dark isolates differ, it's probable that coinfection is a regular occurrence in the hyperendemic region, with clinical consequences for feline sporotrichosis dynamics. Future research will look at whether this species, like other fungus, has a wide range of phenotypic plasticity.

Key Words: *Phylogenetic; Mycoses; Host-fungus interaction*

INTRODUCTION

Sporotrichosis is a subcutaneous mycosis caused by *Sporothrix* fungus that are thermally dimorphic. These fungi, despite their global distribution, have diverse infection pathways and environmental ranges across the world. Much new information on this genus has been gathered in the recent decade, along with reports on the rising prevalence of sporotrichosis across the world, primarily linked to the detection of many cryptic species and its distinctive ecology and epidemiology. Infection is produced by cutaneous damage caused by plant thorns and wood splinters, generating so-called Gardner's disease, or by cat scratching/biting. Since the outbreak in the mid-1990s, this latter route of infection has been increasingly prominent in Brazil, and it now serves as the primary source of contamination, resulting to the now hyperendemic neglected zoonosis in the state of Rio de Janeiro. Recent epidemiological studies predict that around 4700 domestic felines and over 4000 people are afflicted in this area. *Sporothrix* has also spread to at least six additional states in Brazil. The Brazilian outbreak is caused by *S. brasiliensis*; nevertheless, in China, *S. globosa* developed an epidemic among farmers after being transmitted by the saprozoic pathway. *S. schenckii* zoonotic transmission by domestic felines has also been reported in the United States, Mexico, Panama, Argentina, India, and Malaysia. Recent epidemiological studies predict that around 4700 domestic felines and over 4000 people are afflicted in this area.

S. brasiliensis is a highly pathogenic emergent species limited to Brazil that has been implicated as the etiological agent of nearly all human and animal sporotrichosis cases documented in the nation. Cats have a special relationship with this fungus, presumably owing to their high sensitivity, which results in severe disease forms that need long-term antifungal treatment. Domestic cats are frequently resistant to itraconazole, the major azole used in sporotrichosis therapy, resulting in recurrence and treatment abandonment. In reality, treatment failure is the primary factor inhibiting

the breakup of the disease transmission cycle and, as a result, disease control. The current investigation looks into two different clinical *Sporothrix* isolates that were previously isolated from about 4% of ill domestic cats. These creatures were found in three different localities of the Brazilian state of Rio de Janeiro. This indicates an estimate of around 200 animals in the current sporotrichosis hyperendemia, based on only notified instances. Given the disease's well-known underreporting, the genuine number of afflicted felines might be tripled or quadrupled, i.e. 600–800 or possibly more. Furthermore, "differently coloured strains" have been isolated from both feline and human samples on rare occasions, one of which was a human meningitis case caused by *S. brasiliensis*.

Parallel to this, morphological and genotypic studies of these isolates are reported, as well as cytokine production profiles after human monocyte contact. Antifungal susceptibility profiles for yeast-like cells and conidia against amphotericin B, terbinafine, caspofungin, micafungin, fluconazole, itraconazole, and voriconazole are also presented. To our knowledge, this is the first time a domestic kitty has been described as being infected by several *Sporothrix* isolates. Because co-isolated *S. brasiliensis* differential host interaction and responsiveness to antifungal medicines would undoubtedly effect clinical course of sporotrichosis, the most common subcutaneous mycosis in Latin America, the findings revealed in this research should encourage further relevant studies.

CONCLUSION

Finally, co-infections of domestic felines with many *S. brasiliensis* isolates, each with possibly varied host interaction capacities and antifungal responses, may contribute to clinical and epidemiological serious issues. Future research will be able to identify whether co-infection and/or phenotypic flexibility play major roles in sporotrichosis dynamics using a greater number of animals and fungal isolates, as well as other phenotypic and genetic targets and *in vivo* interactions.

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