Emilie-Jeanne Pink, 1 Christine Blais, 2 Mathilde Laurin, 1 Claire Rosset, 1 Olga St-Onge, 1 Laurence Labelle, 1 Adam Sabin, 2 Patrick Tremblay, 1 Marc-André Lévesque, 1 Joseph Carrier, 1 Jean-François Tremblay, 1,3* 

1Centre de recherche sur le métabolisme, Université de Montréal, Montréal, Canada
2Département de pharmacie, Université de Montréal, Montréal, Canada
3Département des sciences de la santé, Université de Montréal, Montréal, Canada

*Corresponding author: jeanfrentremblay@umontreal.ca

Received: 6 June 2021 Revised: 6 June 2021 Accepted: 6 June 2021

Abstract: Metabolomics is a powerful tool for investigating changes in metabolism that occur in response to environmental stressors. Here, we used this approach to investigate the effects of a novel dietary intervention on the gut microbiome and metabolic phenotype of rats. Our results showed that the intervention resulted in significant changes in the abundance of certain microbial taxa and in the production of specific metabolites, indicating that this intervention could have beneficial effects on metabolic health. This study highlights the potential of metabolomics as a tool for understanding the impact of dietary interventions on the gut microbiome and metabolism and underscores the importance of further investigations to validate these findings.
Objective: To detect by chitinase PCR and Species-Specific PCR (SS PCR) the DNA of *Sporothrix* spp in fresh tissue and Formalin Fixed Paraffin-Embedded (FFPE) tissue from patients with sporotrichosis and to evaluate the susceptibility to antifungal of *Sporothrix* spp isolates obtained from patients diagnosed with sporotrichosis in the Medical Mycology group of the Faculty of Medicine of the University of Antioquia.

Materials & Methods: At the Medical Mycology group service, 24 patients with suspected sporotrichosis were evaluated between 2018 and 2022. Samples of the skin lesions (fresh tissue) were taken for molecular and microbiological culture. Regarding the FFPE tissue, 67 samples stored by different histopathological diagnosis came from 1971 to 2022 were chosen: 45 samples had a histopathological diagnosis of sporotrichosis, and 42 samples of other diseases with involvement in subcutaneous tissue, were used as controls.

DNA extracts previously extracted from fresh tissue and FFPE tissue were used to perform chitinase nested PCR and SS nested PCR. The chitinase-nested PCR amplifies a 219 bp fragment for the gene *Sporothrix*. For species identification, species-specific primers were used (SS PCR), which amplify a 311 bp sequence for *S. schenckii* s. str and 245 bp for *S. globosa* of the calmodulin gene. For the SS PCR, a nested PCR was implemented using the primers CAL1 and CAL2 for the external sequence, and the species-specific primers for the internal sequences.

The antifungal susceptibility tests were performed according to the Clinical and Laboratory Standards Institute (CLSI) M27-A3 protocol for years. Six antifungals were used: itraconazole, terbinafine, voriconazole, posaconazole, amphotericin B, and fluconazole.

Results: The culture was positive for *Sporothrix* spp in 11 (46%) patients and the chitinase nested PCR was positive in 14 of these, with a sensitivity of 93% and a specificity of 91%. In 11 patients both the culture and the chitinase nested PCR were negative.

SS nested PCR was applied to the 14 DNA extracts with positive chitinase PCR, of which seven were positive for *S. schenckii* s. str. and one for *S. globosa*. For the other six samples, the results of this PCR were negative. The results of these PCR were confirmed by the identification of species from the isolates recovered in culture: 13 were identified as *S. schenckii* s. str. and 1 as *S. globosa*.

Of the 45 FFPE tissue samples with histopathological diagnosis of sporotrichosis, the chitinase PCR was positive for 25 (55%) of these and all FFPE tissue samples were negative for SS nested PCR.

Conclusions: Chitinase nested PCR had a sensitivity of 93% and a specificity of 91% with respect to culture, in samples from fresh tissue. This PCR also has a good performance when it is applied to a good-quality DNA obtained from FFPE tissue, hence, PCR positivity decreased in samples stored for >15 years. The results of the nested SS PCR are encouraging, however, it