

Detection of dermatophytes in the environment of a podiatry clinic

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Abstract

The detection of dermatophytes in podiatric clinics is a matter of concern since the environmental presence of these fungi can contribute to spread the infection to podiatry workers and to other patients treated in the podiatry clinics. The aim of the present study was to characterize the dermatophyte burden during podiatric activities by the use of culture and molecular methodologies for fungal DNA detection directly from the collected samples.

Over the period of 4 weeks, environmental samples from a podiatric clinic were collected for both conventional and molecular methodologies. In the first week, 1 out of 17 samples had a positive result for *Trichophyton rubrum* (detected in an air sample), whereas in the remaining weeks, no dermatophytes were identified in the remaining 129 samples (weeks 2, 3 and 4). The molecular detection of dermatophytes was performed in the 14 air samples using two different DNA volumes: 2 and 5 µl. Globally, 5 out of the 14 (36%) samples analyzed showed positive results for the detection of *T. rubrum* DNA.

Giving the ratio of positive/inhibited PCR reactions, we cannot rule out the hypothesis of more positive samples. Nevertheless, the obtained results emphasize the importance of the application of new methodologies for an air quality assessment approach and reinforce the complementarity of both cultural and conventional methodologies.

To our knowledge, this study presents for the first time, the application of the Dermatophyte PCR kit for dermatophyte DNA detection directly from environmental samples (air). The promising results indicate the need of optimization of this procedure specifically in this type of samples in order to use this methodology in a routine basis, for occupational and indoor air quality exposure assessments.

Introduction

Podiatry is a healthcare profession that specializes in the management of disease and disorders of the lower limb and foot. Podiatric treatments potentially generate substantial concentrations of organic dusts (up to 3g of toenail dust may be generated per day in a podiatry practice). The detection of dermatophytes in podiatric clinics is a matter of concern since the environmental presence of these fungi can contribute to spread the infection to podiatry workers and to other patients consulted in the podiatry clinics.

Methods

- The study was conducted at a hospital based Podiatry out patient clinic located in Galway, Ireland, during 11 days.
- During the study podiatrists worked in one of two treatment rooms and treatments included nail reduction, corn and callous debridement and wound debridement following usual mechanical debridement technique with a scalpel
- A 300 liter air sample was collected using impaction air sampler operating at a flow rate of 100 l min⁻¹. Surface samples were collected by swabbing the podiatrists work bench located parallel to the patient
- 44 air samples and 39 swabs from surfaces were inoculated in Mycosel agar.
- Fourteen air samples from the same sampling sites were collected for direct detection of fungal DNA. Air samples ranging from 88 to 300L were collected using a calibrated impinger device, at 2.2L/min airflow rate.
- Five milliliters of the collection liquid was used for DNA extraction using the ZR Fungal/Bacterial DNA MiniPrep Kit. Detection of dermatophytes species (in general) and *Trichophyton rubrum* (in particular) were both achieved by using the Dermatophyte PCR kit, (SSI Diagnostica, Herredsvæjen Hillerød, Denmark). PCR amplifications were performed using 2 µL and 5 µL of the extracted DNA.

Objectives

The aim of the present study was to characterize the dermatophyte burden during podiatric activities by the use of cultural methods but also molecular methodologies for fungal DNA detection directly from the collected samples.

Results

- ❖ With the exception of one air sample collected on day 2, all air samples analysed using culture based methods were negative for dermatophytes. Furthermore, all surfaces samples assessed using culture based methods were negative for dermatophytes.
- ❖ The molecular detection of dermatophytes (culture independent) was performed on the fourteen air samples collected via impingement. DNA volumes of 2 µl and 5 µl were used to perform the PCR analysis for dermatophytes.
- ❖ Using 2 µl of template DNA, one out of fourteen samples tested was positive for *T. rubrum* and none of the PCR reactions were inhibited. Using a 5 µl volume of DNA, of fourteen samples tested, five were positive for *T. rubrum* but the PCR reaction was inhibited in the remaining six (Table 1). Dermatophytes were detected in DNA extract collected on day 6 – 9 only, the PCR reaction was inhibited on samples collected on day 2, 3, 4, and Day 5, and negative in samples collected on Day 1 and 10. Globally, five out of the fourteen (36%) environmental samples analyzed were positive in the detection of *T. rubrum* DNA. Fungal nail treatments were performed on day 6, 7 and 8.

Table 1. Culture and molecular detection of dermatophytes in air samples of a podiatry clinics

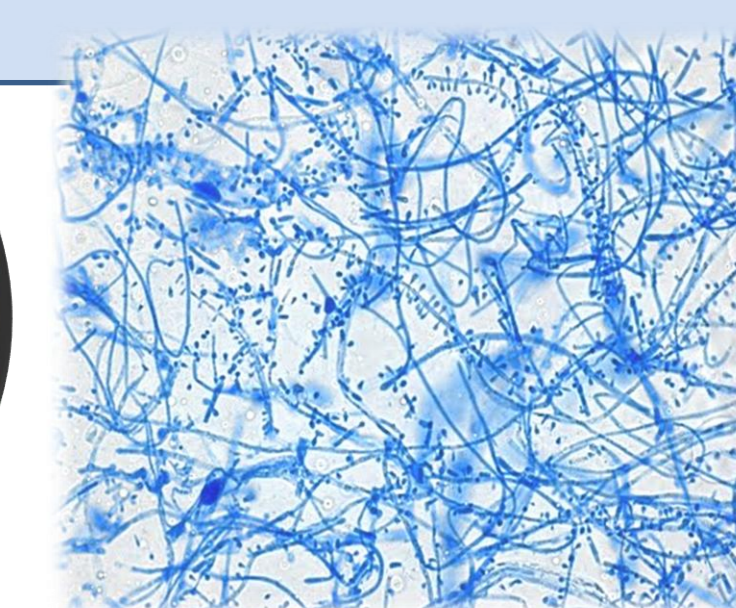
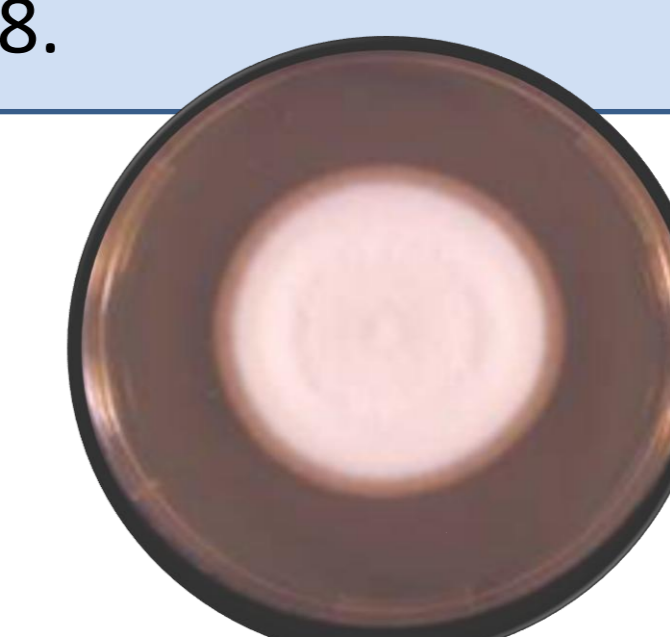
Air Sampling Code	Culture for dermatophytes	Dermatophyte PCR result (using 2 µl of the extracted DNA)	Dermatophyte PCR result (using 5 µl of the extracted DNA)
Indoor day 1	Negative (air and surfaces)	Negative	Negative
Indoor day 2	Positive for <i>T. rubrum</i> (air)	Negative	PCR Reaction inhibited
Indoor day 3	Negative (air and surfaces)	Negative	PCR Reaction inhibited
Indoor day 4	Negative (air and surfaces)	Negative	PCR Reaction inhibited
Indoor day 5 - 1	Negative (air and surfaces)	Negative	PCR Reaction inhibited
Indoor day 5 - 2	Negative (air and surfaces)	Negative	Negative
Indoor day 6	Negative (air and surfaces)	Positive for <i>Trichophyton rubrum</i>	Positive for <i>Trichophyton rubrum</i>
Indoor day 7 - 1	Negative (air and surfaces)	Negative	Positive for <i>Trichophyton rubrum</i>
Indoor day 7 - 2	Negative (air and surfaces)	Negative	Positive for <i>Trichophyton rubrum</i>
Indoor day 8	Negative (air and surfaces)	Negative	Positive for <i>Trichophyton rubrum</i>
Indoor day 9	Negative (air and surfaces)	Negative	Positive for <i>Trichophyton rubrum</i>
Indoor day 10	Negative (air and surfaces)	Negative	Negative
Outdoor day 10	Negative (air and surfaces)	Negative	PCR Reaction inhibited
Indoor day 11	Negative (air and surfaces)	Negative	PCR Reaction inhibited

Conclusions

✓ To our knowledge, this study presents for the first time, the application of the Dermatophyte PCR kit for dermatophyte DNA detection directly from environmental samples (air).

✓ Giving the ratio of positive/inhibited PCR reactions, we cannot rule out the hypothesis of more positive samples.

✓ The promising results indicate the need of optimization of this procedure specifically in this type of samples in order to use this methodology in a routine basis, for occupational and indoor air quality exposure assessments.



Trichophyton rubrum