Diagnosis of Dermatophytes using Molecular Technology – a one year experience

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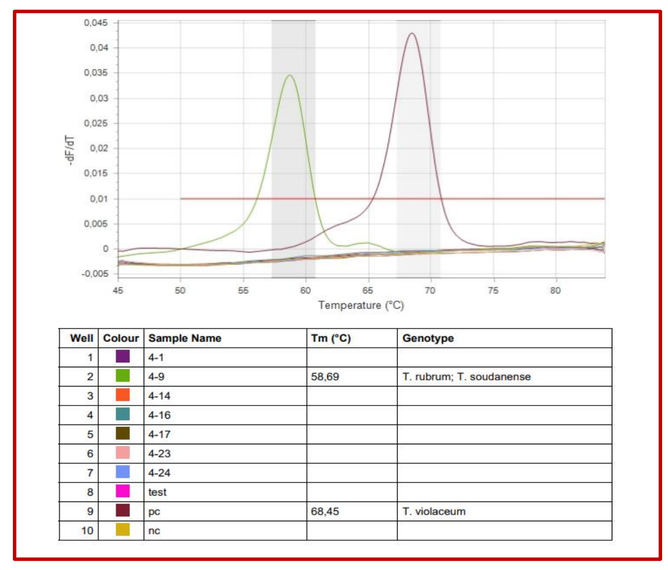
Objectives and aim: Although fungal culture is a well-established method for diagnosing fungi, the PCR is a highly sensitive molecular technology and provides results in short time. The study's objective was to compare multiplex qPCR and culture using samples from various cutaneous regions and nail scrapings.

Patients and Methods: A total of 971 samples from patients referred to the Outpatients Centre (87%) or mailed (13%) were simultaneously analysed by cultivation in fungal culture media (Sabouraud agar at 28°C for 21 days), KOH direct tests and by multiplex qPCR (DermaGenius, Pathonostic). The samples (41% men, 59% women) from the skin (26%) and nails (74%) were examined.

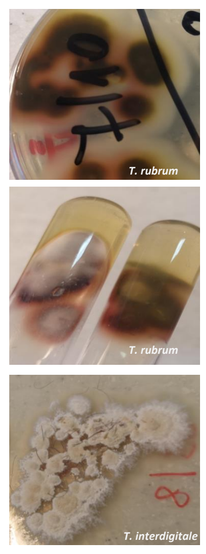
DermaGenius® 3.0 Complete RT-PCR

Dermatophytes

Antropophil:	Zoophil:	Others:
T. interdigitale	*M. canis*	*E. fluccosum*
T. rubrum	*T. benhamiae*	*M. audouinii*
T. soudanense	*T. mentagrophytes*	*N. gypsea*
T. tonsurans	*T. verrucosum*	
T. violaceum		



Well	Colour	Sample Name	Tm (°C)	Genotype
1		4-1		
2		4-9	58,69	T. rubrum; T. soudanense
3		4-14		
4		4-16		
5		4-17		
6		4-23		
7		4-24		
8		test		
9		pc	68,45	T. violaceum
10		nc		



Results:

Dermatophyte infection was detected in 359 (37%) patients. Both technologies yielded positive results in 176 (18%) of all samples tested, while 183 infections (51%) were missed by culture (Table 1). Of the qPCR-positive samples, growth of the dermatophyte was observed in 66% of the samples from skin (Fig.1) and 39% from nail scrapings (Fig. 2).

There was not a single case of a positive culture with a negative PCR result.

The most common species was *T. rubrum*, detected by cultivation in 11% and by qPCR in 26% of all 971 samples (Table 2).

Table 1: Comparison of the sampes tested by PCR und culture

Swabs		Skin	Nails
PCR pos /	176	65	111
Cult pos		25%	16%
PCR pos /	**183**	**33**	**150**
Cult neg		**13%**	**21%**
PCR neg /	612	157	455
Cult neg		62%	63%
Total	971	255	716

Fig.1 Skin n = 255

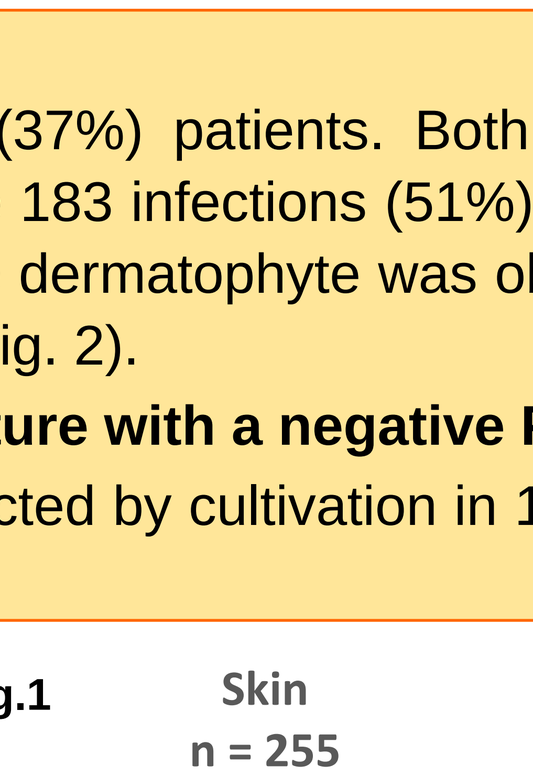


Fig.2 Nails n = 716

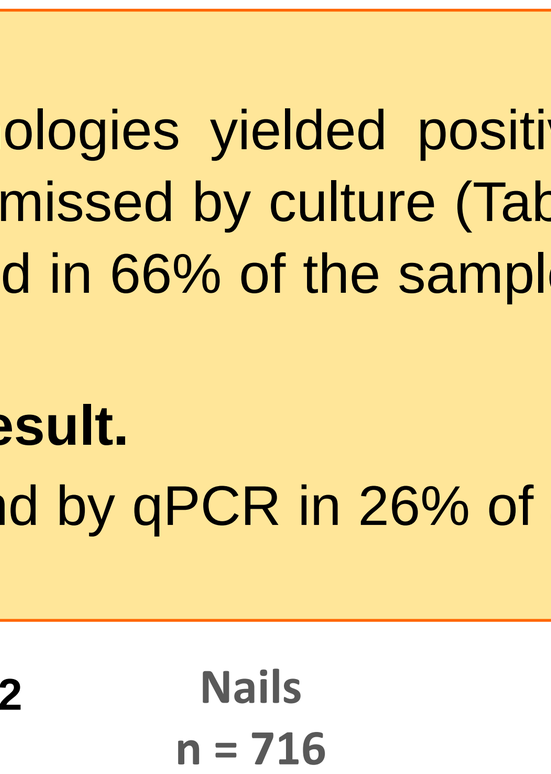


Table 2: Frequency of detection of the most prevalent dermatophyte species in PCR and culture

Species	PCR pos	Cult pos
Microsporum canis	8	8
T. benhamiae	5	3
T. interdigitale	37 (4%)	18 (2%)
T. mentagro/interdig komplex	8	5
T. rubrum	**251 (26%)**	**107 (11%)**
T. tonsurans	9	9

Conclusion:

Our results demonstrate the high sensitivity of qPCR. It overcomes cumbersome culture methods. Importantly, one half of dermatophyte infections were detected only by qPCR. The rapid and accurate performance of qPCR allows timely and appropriate treatment allocation. Cultivation methods remain important for antifungal resistance testing and non-dermatophyte diagnosis.