

DERMATOPHYTES MULTIPLEX PCR



STATENS
SERUM
INSTITUT

*prevents and controls
infectious diseases,
biological threats and
congenital disorders*

- *Identification of fungi causing nail infections*
- *Higher sensitivity than conventional methods*
- *Results within 5 hours*

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Description

The Dermatophyte PCR Kit is to be used for in vitro diagnostic detection of dermatophytes in general (pan-dermatophytes) and specifically *Trichophyton rubrum*.

The kit includes two buffers for DNA preparation, PCR ReadyMix (including loading buffer), primer mix and two control DNA samples. The primer mix contains two primer pairs directed towards genes encoding chitin synthase 1 for detection of dermatophytes in general and ITS2 (internal transcribed spacer) for detection of *T. rubrum*. Control 1 consists of dermatophyte genomic DNA and control 2 consists of *T. rubrum* genomic DNA. The package contains reagents enough to perform 100 multiplex PCR reactions.

Background

Nail infections are mainly caused by *T. rubrum*, followed by *T. mentagrophytes*. Traditionally the time required for species identification by culture may vary from 10 to 15 days up to 3 to 4 weeks¹. This PCR based diagnostic method can detect dermatophytes in general and specifically *T. rubrum* within 5 hours.

Specificity

A total of 118 nail specimens were tested for pan-dermatophyte and *T. rubrum* infection by both the multiplex PCR method and the conventional methods (microscopy and/or culture). Overall, 42.4% of the specimens were pan-dermatophyte positive by PCR whereas 38.1% were positive using the conventional methods. The sensitivity of pan-dermatophyte identification in nail specimens was therefore increased by 4.3% using the multiplex PCR method. Furthermore, the test showed that the sensitivity for identification of *T. rubrum* was increased by 18.6% using the PCR based diagnostic (see table)¹.

	Pan-dermatophytes	<i>T. rubrum</i>
Conventional methods	38.1%	22.9%
PCR	42.4%	41.5%
Increased detection	4.3%	18.6%

Procedure

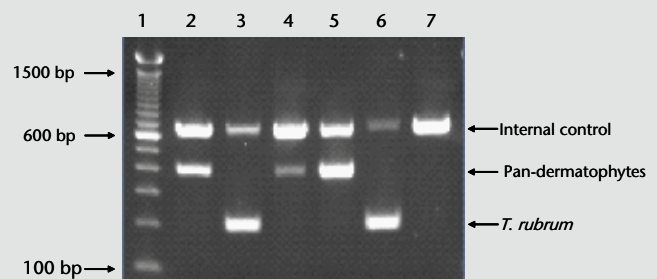
Identification of nail infections using the Dermatophyte PCR Kit requires the following steps:

- 1) Preparation of template from nail specimens
- 2) Mixing reagents for the PCR reactions
- 3) PCR amplification in a thermocycler
- 4) PCR product analysis using agarose gel electrophoresis

It is possible to prepare and run up to 30 samples within 5 hours. The total process time will only increase marginally when running more samples.

Figure: *T. rubrum* specific and pan-dermatophyte multiplex PCR product analysis.

Lane 1: Molecular size marker (100 bp DNA ladder);
Lane 2: Control 1 (pan-dermatophyte genomic DNA);
Lane 3: Control 2 (*T. rubrum* genomic DNA);
Lane 4: Pan-dermatophyte positive nail specimen;
Lane 5: Pan-dermatophyte positive nail specimen;
Lane 6: *T. rubrum* positive nail specimen;
Lane 7: Negative control (A/B buffer).



Storage and shelf life

The PCR Kit should be stored at -20°C. Dispense the PCR ReadyMix, the primer mix and the DNA controls into several aliquots and store them at -20°C. Primer mix and control aliquots in use can be stored at 4-8°C for up to two weeks.

The expiry date of the kit is printed on the label.

Available product

Article no. 73411 Dermatophyte PCR-kit, 100 multiplex PCR reactions.

Ordering and Information

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References

1. Brillowska-Dabrowska, A., Saunte, D. M. and Arendrup, M. C. 2007. Five-Hour Diagnosis of Dermatophyte Nail Infection with Specific Detection of *Trichophyton rubrum*. *Jour. Clin. Microbiol.* 1200-1204.