

A novel multiplex PCR assay for rapid detection and typing of *Candida* using melting curve analysis

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BACKGROUND

Candida spp. is the most common opportunistic pathogens, and the five common clinical species are *Candida albicans*, *Candida parapsilosis*, *Candida tropicalis*, *Candida glabrata*, and *Candida krusei*. *Candida* infection ranks fourth in hospital blood-borne infections, with a mortality rate of up to 40%-50%, and is also the most clinically prevalent deep fungal disease, accounting for 95% of human pathogenic fungal infections. Traditional laboratory methods for the clinical identification of fungi are time-consuming and lack sensitivity and specificity, and the results are unreliable and subjective. Fluorescent PCR is a rapid, reliable and sensitive clinical diagnostic technique and aids in the diagnosis of clinically pathogenic fungal infections, with a relatively high rate of pathogen detection for invasive or disseminated fungal infections. Based on the multiplex fluorescence PCR technique combined with melting temperature analysis, the detection of *Candida* spp. and the clinical classification of five common pathogenic *Candida* species will be critical for the early diagnosis of Candidiasis and improved outcomes.

METHOD

The diagnostic efficacy of the kit was evaluated by testing 143 clinical samples, including 60 serum samples, 43 plasma samples, and 40 whole blood samples, using the *Candida* test developed by Dynamiker Biotechnology (Tianjin) Co., Ltd. The reliability of the test for *Candida* typing was also evaluated using the sequencing results as the standard.

RESULT

The sensitivity specificity of the *Candida* test kit developed by Dynamiker Biotechnology was above 90% for all three different sample types, with the best sensitivity specificity for serum (Table 1). *Candida* typing results obtained by multiplex fluorescence PCR combined with melting curve analysis were 100% consistent with sequencing results (Figure 1).

CONCLUSION

The *Candida* reagent developed by Dynamiker Biotechnology is a rapid and flexible PCR product for the detection of *Candida*.

Table 1 Results of diagnostic efficacy assessment of the kit

Sample Type	Positive samples (n) Proven/probable	Negative samples (n)	Sensitivity%	Specificity%
1 Serum	29	31	93.1(27/29)	96.8(30/31)
2 Plasma	21	22	90.5(19/21)	90.9 (20/22)
4 Whole blood	20	20	90.0(18/20)	90.0(18/20)

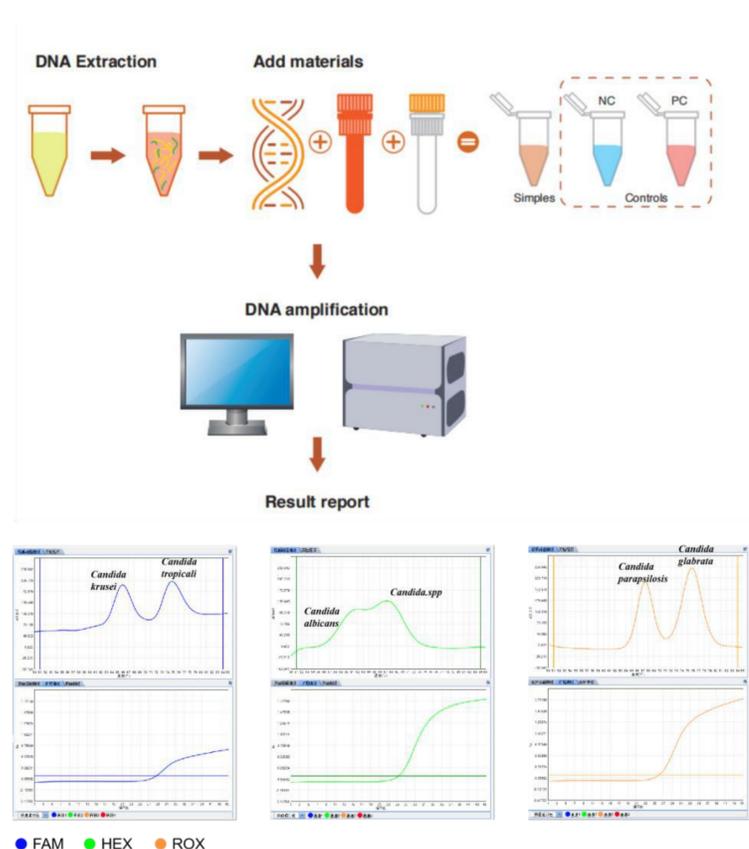


Figure 1 Results of fluorescent PCR for five species of *Candida*