

Mediator Probe PCR:

Improved real-time PCR detection using mediator probes and universal reporters

Mediator Probe PCR enables highly sensitive nucleic acid detection and precise quantification, using label free mediator probes in combination with optimised fluorogenic universal reporter molecules.

In Mediator Probe PCR hydrolysis probes are replaced by label-free target-specific mediator probes. During amplification these probes are cleaved and the mediator sequences are released, which subsequently activate the fluorogenic universal reporters. Since universal reporters are target sequence independent, once optimised, they will improve the fluorescence signal generation of all assays they are used in.

Features

- Target-specific label-free mediator probes
- No performance limitations due to target sequence characteristics
- Precise guidelines for mediator probe design
- Standardised fluorogenic universal reporters
- Optimised fluorescence signal generation in each assay, right from the start

Application

- Quantitative multiplex real-time PCR
- Reverse transcription real-time PCR
- Digital PCR

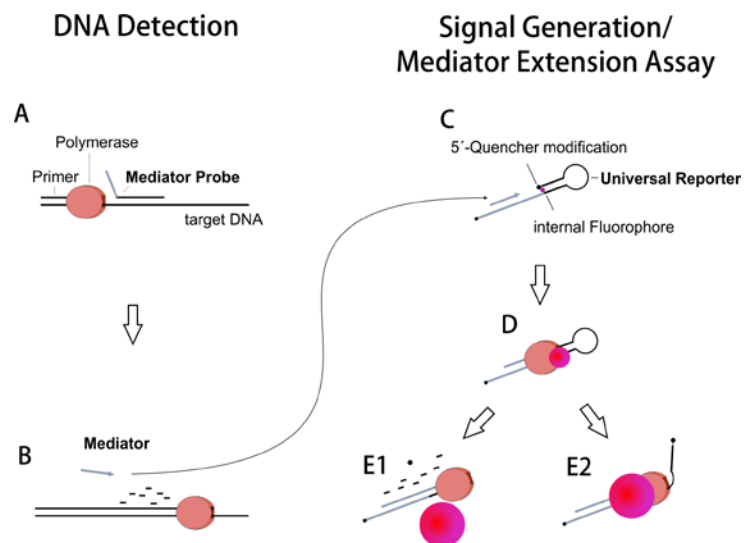


Fig. 1: Reaction mechanism of Mediator Probe PCR. As can be seen, target sequence detection (left) and fluorescence signal generation (right) are two independent processes.¹

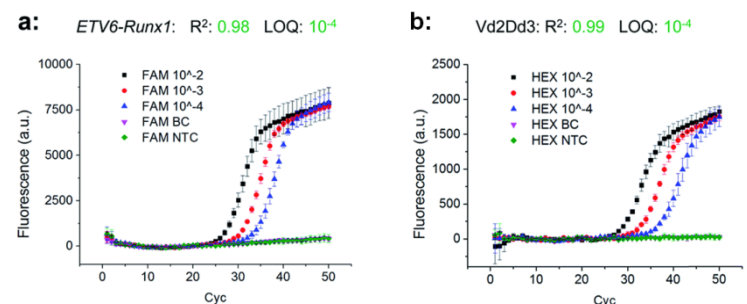


Fig. 2: Biplex Mediator Probe PCR, simultaneously quantifying the gene-fusion *ETV6-Runx1* (a) and the gene-rearrangement *Vd2Dd3* (b). In each reaction 1 000 – 10 copies of target DNA were detected in a background of 100 000 copies of buffy coat (BC) background DNA.¹

References

- ¹ Lehnert et al.: Fluorescence signal-to-noise optimisation for real-time PCR using universal reporter oligonucleotides. *Anal. Methods*, (2018), DOI: 10.1039/c8ay00812d. –open access-
- ² Wadle et al.: Simplified development of multiplex real-time PCR through master mix augmented by universal fluorogenic reporters. *BioTechniques*, (2016), DOI: 10.2144/000114443 –open access-

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