## **Application Manual**

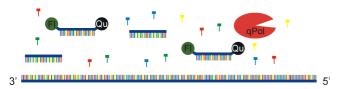


### **Dual Labeled Fluorescent Probes**

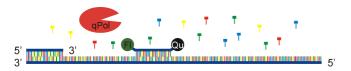
#### **Technical Information**

**Real-Time PCR** 

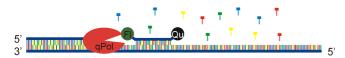
## Principle of real-time sample DNA quantification using dual labeled fluorescent probes



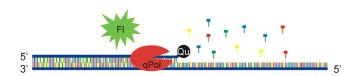
Quantitative real-time PCR assay requires qPol, dual labeled fluorescent probe, primers and template. The proximity of fluorophore and quencher prevents the reporter from fluorescing.



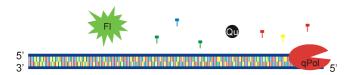
Both the dual labeled fluorescent probe and the PCR primers anneal to the target sequence during the annealing step.



During the PCR extension step, the qPol Taq polymerase extends the primer.

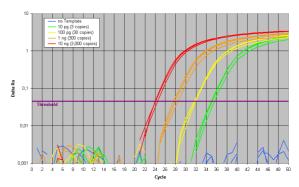


When the enzyme reaches the probe, its  $5' \rightarrow 3'$  exonuclease activity cleaves the fluorophore from the probe. The fluorophore is released and its signal can be measured.



After complete extension of the amplicon the following PCR amplification cycle is run.

### Jena Bioscience qPCR reagents provide exceptional sensitivity and accuracy



qPCR Master: Amplification of a beta-actin gene fragment from human genomic DNA

Dual Labeled Fluorescent Probes are shipped as lyophilisates and can be stored at -80°C for at least one year. Before use, make a stock solution by re-suspending the dry probe in PCR grade water or TE-buffer and aliquot the solution into microvials. Store the aliquots at -20°C and avoid frequent thawing and freezing.

# **Application Manual**



## **Dual Labeled Fluorescent Probes**

#### **Technical Information**

Real-Time PCR

Fluorescence Dyes		
Dye	Excitation max [nm]	Emission max [nm]
LC°Cyan500	450 nm	500 nm
5-FAM	495 nm	520 nm
6-FAM	495 nm	520 nm
FITC	490 nm	525 nm
TET	521 nm	536 nm
JOE	522 nm	548 nm
Yakima Yellow	530 nm	549 nm
HEX	535 nm	556 nm
СуЗ	546 nm	563 nm
TAMRA	564 nm	579 nm
ROX	576 nm	601 nm
Texas Red	586 nm	610 nm
LC®Red610	590 nm	610 nm
LC®Red640	625 nm	640 nm
Cy5	646 nm	662 nm
Cy5.5	683 nm	705 nm
IRD700	685 nm	705 nm

Dark Quencher		
Quencher	Quenching max [nm]	Quenching Range [nm]
TAMRA*	544	520-570
DABCYL	453	380-530
ECLIPSE	522	390-625
BHQ-1®	535	480-580
BHQ-2®	579	550-650
BHQ-3®	672	620-730
BBQ®-650	650	550-750

<sup>\*</sup> TAMRA is widely used as quencher, especially in combination with the reporter FAM. Please note that TAMRA is no dark quencher and contributes to an increase in background signal because of its own fluorescence emission.

Black Hole dark quencher (BHQ-1) probes are an advanced alternative to TAMRA and ensure a higher signal-to-noise ratio.

For custom-made Fluorescent DNA Probes or further information, please contact us at: pcr@jenabioscience.com