

Quantification Strategies in real time qRT-PCR

M. W. Pfaffl, *BioSpektrum* 2004 (Sonderausgabe PCR)

absolute quantification

external calibration curve
one color detection system
SYBR Green I

external calibration curve
two color detection system
e.g. Probes, ROX

ROX

external calibration curve

- RT-PCR product
- plasmid DNA
- *in vitro* transcribed RNA
- synthetic DNA Oligos
- synthetic RNA Oligos

relative quantification

normalisation

via one reference gene

via reference gene index
>3 HKG

external calibration curve **without** any reference gene

ROX

without real-time PCR efficiency correction

$2^{-\Delta\Delta CP}$

with real-time PCR efficiency correction

REST, qGene
LC software

Gene Quantification

Sabine Zange



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