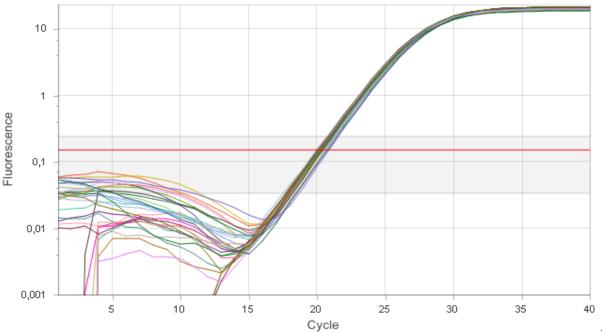
Rob™ OneDip™ QPCR set-up reproducibility for 2µl template.



single stranded DNA fragment containing the beta-actin amplicon sequence. Primers are specific for mouse beta-actin.

The template is a

Results:

Based on single template

Cq_{Average}: 21.25

Standard deviation (Cq): 0.19

Cq_{Max} – Cq_{Min}: 0.82

Based on average Cq value of triplicate

Cq_{Average}: 21.25

Standard deviation (Cq): 0.075

Cq_{Max} - Cq_{Min}: 0.23

Run conditions:

QPCR: HotStart activation, 2 min at 95°C followed by 40 cycles with 10 s at 95°C and 10 s at 60°C.

Run time: 44 min.

Set-up: 18 μl master mix was dispensed using multi dispense followed by 2 μl template using OneDip™ technology

(multi dispense) from one tube. Run time: 4 min 57s.

Reagents:

qPCRBIO SyGreen Mix from PCRBiosystems was selected because it is the fastest and most sensitive kit available on the market. It combines latest polymerase and buffer technology with a proprietary intercalating dye that doesn't inhibit PCR.

QPCR instrument:

Mic QPCR-instrument from Bio Molecular Systems is the most precise QPCR instrument available today. Virtually no temperature differences between samples during the run combined with the magnetic induction heating ensures highest possible quality on data.

Reproducibility of Mic is within Cq 0.20.

Conclusion:

OneDip^m technology is useable from 2 μ l template volume for detection and concentration calculations. The variance between average triplicate values is less than for individual templates. Larger template volumes will substantially improve precision. Lower volumes are not recommended.

We also recommend to use single dispense for accurate expression analysis and concentration determinations for low template volumes.