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Product Information ExcelTaq™series

2X Q-PCR Master Mix (TaqMan, ROX)

TQ2110	**200 RXN**	
	2X Q-PCR Master Mix (TaqMan, ROX)	1 ml x 2

Storage

Aliquot to avoid multiple freeze-thaw cycles
Protect from light
-20°C for 12 months

Features

- High sensitivity and specificity
- With ROX reference dye

Description

The ExcelTaq™2X Q-PCR Master Mix (TaqMan, ROX) is a ready-to-use reagent with all the essential components for quantitative real-time PCR (qPCR) except primers, TaqMan probes and templates. The master mix includes a 5’ to 3’ exonuclease activity to cleave TaqMan probes that hybridize to target sequence, releasing the fluorophore during probe displacement. With TaqMan probes, the master mix features high specificity and high sensitivity (Fig. 1).

The ExcelTaq™ 2X Q-PCR Master Mix (TaqMan, ROX) contains a hot-start *Taq* polymerase in an optimized buffer that allows for sensitive and precise amplification, real-time tracking of the amplification process, and simultaneous quantification for targeted DNA molecules. The master mix includes ROX reference dye for normalization of each qPCR assay.

The ExcelTaq™ 2X Q-PCR Master Mix (TaqMan, ROX) is ready-to-use and greatly reduces pipetting errors, while largely improving the reproducibility for the process.

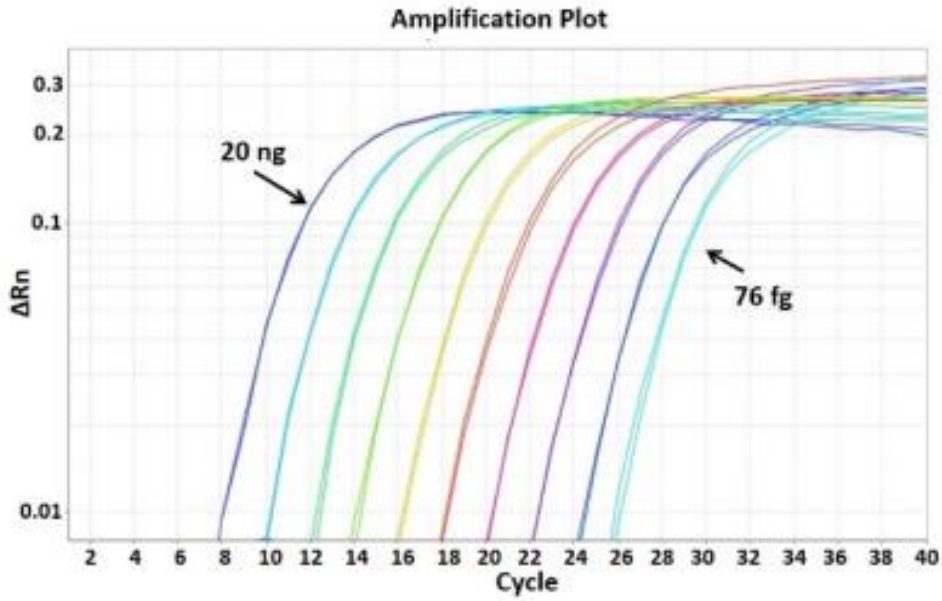

Fig. 1. The amplification plot of real-time PCR with cDNA templates ranging from 76 fg to 20 ng in quantity, analyzed by using TQ2110 ExcelTaq™ 2X Q-PCR Master Mix (TaqMan, ROX) for qPCR amplification.

Application

- Quantitative real-time PCR
- Quantitative two-step real-time PCR

Instrument compatibility

- Applied Biosystems system:
  - 5700, 7300, 7000, 7700, and 7900HT system
  - StepOne™ / StepOne Plus™
  - QuantStudio™ 3 / 5/ 6 / 7
- BioRad system:
  - CFX96 / CFX384
  - Chromo 4™ Real-Time Detector
  - DNA Engine Opticon™ / Opticon™ 2

Instrument compatibility, continued

- Roche system:
  - Roche LightCycler® 480 / Nano
- Cepheid system:
  - Smart Cycler®
- Eppendorf system:
  - Mastercycler® ep realplex
- QIAGEN system:
  - Rotor-Gene™ Q

Note: Selection of fluorescent reporter dye of TaqMan probe should refer to optical detection system of instruction. ExcelTaq™ 2X Q-PCR Master Mix (TaqMan, ROX) is compatible with a variety of real-time instruments, including but not limited to the list above.

Recommended primer design

- Amplicon size: 80-150 bp
- Tm value: around 60°C (calculated with Primer3 software)
- Primer length: 17-25 mer
- Sequence:
  - 45-55% of GC content is recommended.
  - Avoid regional high GC or AT content
  - Avoid palindrome sequence
  - Sequence with G or C at the 3' end is recommended.
- Specificity of primers should be confirmed through a BLAST search.

Recommended probe design

- Tm value: 6-10°C higher than primers
- Probe length: 20-30 mer
- Sequence:
  - 35-65% of GC content is recommended.
  - Avoid regional high GC or AT content
  - Select the strand contains more C's than G's
  - Avoid palindrome sequence
  - Avoid a G at the 5' end to prevent quenching of the 5' fluorophore.
- Specificity of probe should be confirmed through a BLAST search.

Recommended reaction mixture set up for qPCR

Template	2 µl*
Forward primer	125 – 900 nM**
Reverse primer	125 – 900 nM**
TaqMan Probe	100 – 200 nM**
2X Q-PCR Master Mix (TaqMan, ROX)	10 µl
H_2O	to 20 µl
Total volume	20 µl

*Template amount varies depending on the copy number of target present in the template solution. The recommended amount of template is: **1 pg -100 ng of cDNA, 4 ng -50 ng of gDNA or 10^3-10^8 molecules of plasmid**.

** The PCR primer and probe concentration for an optimal qPCR reaction may vary according to primers' and probe's properties.

Recommended qPCR program

Try Two-step cycle protocol first, and optimize the reaction conditions if necessary. If the two-step protocol still does not give optimal results (e.g., if Tm values for the primers are low), try the Three-step cycle protocol.

Two-step cycle for qPCR

Steps	Temp.	Time	Cycles
Template denature and enzyme activation	95°C	10 min[#]	1
Denature	95°C	15 sec	40
Annealing/Extension	60°C	60 sec	

Three-step cycle for qPCR

Steps	Temp.	Time	Cycles
Template denature and enzyme activation	95°C	10 min[#]	1
Denature	95°C	15 sec	40
Annealing	55-60°C	30 sec	
Extension	72°C	30 sec	

[#]We suggest 10 minutes for the first step to thoroughly denature DNA and activate enzymes.

Other Information

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Caution: Not intended for human or animal diagnostic or therapeutic uses.

Related Products

RP1000	ExcelRT Reverse Transcriptase, 20,000 U
RP1100	ExcelRT One-step RT-PCR Kit, 50 RXN
RP1400	ExcelRT Reverse Transcription Kit II, 100 RXN
RI1000	RNAok RNase Inhibitor, 2000 U
TQ1100	ExcelTaq 2X Q-PCR Master Mix (SYBR, no ROX), 200 RXN
TQ1110	ExcelTaq 2X Q-PCR Master Mix (SYBR, ROX), 200 RXN
DM2300	ExcelBand 100 bp+3K DNA Ladder, 500 µl
DM3100	ExcelBand 1 KB (0.25-10 kb) DNA Ladder, 500 µl
DL5000	FluoroDye DNA Fluorescent Loading Dye (Green, 6×), 1 ml
NS1000	FluoroVue Nucleic Acid Gel Stain (10,000X), 500 µl
PM2510	ExcelBand Enhanced 3-color Regular Range Protein Marker, 250 µl × 2
TF1000	SMO-HiFi DNA Polymerase, 100 U × 1
TP1000	ExcelTaq Taq DNA Polymerase, 500 U × 1
TP1200	ExcelTaq 5X PCR Master Dye Mix, 200 RXN
CK1000	Champion E. coli Transformation Kit
WM1000	YesBlot Western Marker I, 250 µl