PhileKorea

1st PhileKorea,Inc. (주)필코리아테크놀로지

Product Insert

QuantiSpeed SYBR One-Step No-Rox Kit

Cat. No.

QS145-01 100 x 20ul reaction: 1 x 1ml
QS145-02 200 x 20ul reaction: 2 x 1ml
QS145-05 500 x 20ul reaction: 5 x 1ml

Shipping: On Dry/Blue Ice
Store at -20℃

Product description:

SYBR Green One-Step Kit uses the latest developments in reverse transcriptase technology and buffer chemistry for efficient cDNA synthesis and PCR in a single tube.

Our modified MMLV reverse transcriptase (RTase) is both thermostable and extremely active. The enzyme is blended with RNase inhibitor preventing degradation of RNA by contaminating RNase. The RTase is not inhibited by ribosomal and transfer RNAs, total RNA is an ideal substrate.

SYBR Green mixes use an intercalating dye which does not inhibit PCR, unlike other popular dyes.

SYBR Green One-Step mix uses proprietary small molecular inhibitor technology that prevents formation of primer-dimers to improve reaction sensitivity and specificity.

High-throughput screening has resulted in a buffer system that allows efficient amplification from GC-rich and AT-rich templates, under fast and standard cycling conditions.

Components:

Reagents	100rxns	200rxns	500rxns
2x SYBR Green One-Step Mix	1 x 1ml	2 x 1ml	5 x 1ml
20x RTase (with RNase inhibitor)	1 x 200ul	2 x 200ul	5 x 200ul

Shipping and Storage:

On arrival the kit should be stored at -20℃.avoid prolonged exposure to light. If stored correctly the kit will retain full activity for 12months. The kit can be stored at 4℃ for 1 month. The kit can go through 30freeze/ thaw cycles with no loss of activity.

Limitations of product use:
The product may be used only for in vitro research purposes.

Instrument compatibility

Manufacturer	Model
Illumina®	Eco™
Bio-Rad	iCycler®, MyiQ®, iQ™5, Opticon™, Opticon2™, MiniOpticon,Chromo4™, CFX96, CFX384
Roche	LightCycler® 480, LightCycler®Nano
Qiagen	Rotor-Gene™ 3000 & 6000 & Q
Takara	Thermal Cycler Dice® (TP800)
Eppendorf	Mastercycler®, ep realplex, Mastercycler®realplex25
Cepheid	SmartCycler™
Techne	Quantica®
Analytica Jena	qTower
Applied Biosystems	7500, 7500FAST, Viia7™
Stratagene(Agilent)	MX4000P®, MX3000P®, MX3005P®

Primer design: For efficient amplification under fast cycling conditions we recommend amplicon lengths between 80bp and 200bp. With all manufacturers master mixes the shorter the amplicon length the faster the reaction can be cycled. Amplicon lengths should not exceed 400bp. Primers should have a predicted melting temperature of around 60℃, using default Primer 3 settings (http://frodo.wi.mit.edu/primer3/).

Reaction setup

1) Before starting, briefly vortex 2x SYBR Green One-Step Mix

2) Prepare a master mix based on following table, we recommend also setting up a no-RTase control:

PCR Reaction Conditions (for 20ul volume)		
Reagent	**20uL reaction**	**Final concentration**
2x SYBR Green One-Step Mix	10ul	1x
Forward primer (10uM)	0.8ul	400nM
Reverse primer (10uM)	0.8ul	400mM
20xRTase	1.0-2.0ul	1x or 2x
Template RNA	1pg to 1ug total RNA >0.01pg mRNA	variable
PCR grade dH_2O	Up to 20ul final volume	

3) Program the instrument using following conditions, acquiring data on the appropriate channel:

Cycle	Temperature	Duration	Comments
1	45°C to 55℃	10 min	Reverse transcription, 45℃ is recommended for most applications, 55℃ should be used only when amplicon contains regions of high secondary structure
1	95°C	2min	Polymerase activation, 2minutes
40	95°C 60°C to 65°C	5sec 20-30sec	Denaturation Anneal/Extension, do not exceed 30sec, do not use temperatures below 60℃
Melt analysis	Refer to instrument instructions	Optional melt profile analysis	

Technical Support

If the troubleshooting guide does not solve the difficulty you are experiencing, please contact Technical Support with details of reaction setup, cycling conditions and relevant date.

Address : Woolim Lions Valley A-606B, 168, Gasan digital 1-ro, Geumcheon-gu, Seoul, Korea 08507
E.mail : info@philekorea.co.kr
Website : www.philekorea.co.kr
Tel. +82 42 862 9636 +82 2 2105 7020
Fax. +82 42 862 9638 +82 2 2105 7025