RealHelix™ *Premier* qPCR Kit [Green, Low ROX]

Kit Contents

RealHelix™ *Premier* qPCR Kit [Green, Low ROX]		
Cat. No.	**PQL-S200 (200rxns)**	**PQL-S500 (500rxns)**
Premier 2x Premix [GL]	1ml x 2ea	1ml x 5ea
Instruction for Use	1ea	1ea

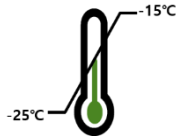
Description

The RealHelix™ *Premier* qPCR Kit [Green, Low ROX] is designed to perform a rapid, highly specific, and sensitive real-time quantification of target DNA. Specially designed buffer and antibody-mediated hot start enzyme considerably reduce the primer dimer formations and ensure producing reliable data. The convenient 2x concentrated premix contains *Ab+Taq* polymerase, dNTPs, buffers, Mg^{2+}, a green fluorescent dye, ROX passive dye (100nM concentration), and stabilizing agent. The ROX dye does not take part in the PCR reaction but allows to normalize for non-PCR related signal variation and provides a baseline in multiple reactions.

Application

Quantification of target DNA sample by real-time PCR

Storage



In the dark at -20±5℃

Shelf life

12 months

NanoHelix Co., Ltd.

A-dong and B-dong, 43-15, Techno 5-ro, Yuseong-Gu, Daejeon, 34014, South Korea. TEL : 82-42-867-9055, FAX : 82-42-867-9057

E-mail : info@nanohelix.net <**www.nanohelix.net** **www.nanohelix.co.kr/KOR**>

Quality Control

In accordance with NanoHelix's ISO 13485-certified Quality Management System, each lot of **RealHelix™ *Premier* qPCR Kit [Green, Low ROX]** was tested against predetermined specifications to ensure consistent product quality.

Protocol

1. **Program a real-time PCR instrument according to the recommendations below. The excitation and emission maxima of the included green fluorescence dye are at 494 nm and 521 nm, respectively.**

<2-step cycling method >

Step	Condition		Cycle(s)
Enzyme Activation		95℃ for 5 min	1
PCR Amplification	Denaturation	95℃ for 20 sec	40
	Annealing & Extension	60℃ for 40 sec **Collect the fluorescence data**	

<3-step cycling method >

Step	Condition		Cycle(s)
Enzyme Activation		95℃ for 5 min	1
PCR Amplification	Denaturation	95℃ for 20 sec	40
	Annealing	[1)]**AT**℃ for 30 sec	
	Extension	72℃ for 30 sec **Collect the fluorescence data**	

[1)]**AT,** annealing temperature of primers used

Annealing Temperature = T_m – (4 ~ 6℃)

Where, T_m (Melting Temp.) = [4℃ x (G + C)] + [2℃ x (A + T)]

☞ For melting curve analysis of products, refer the instrument documentation.

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2. Add following components for a 20μl volume reaction.

Components	Volumes
DNA Template	X μl
Premier 2x Premix [GL]	10μl
Forward primers (10μM)	0.5μl (final 0.25μM)[1)]
Reverse primers (10μM)	0.5μl (final 0.25μM)[1)]
RNase-free Water	Adjust to final 20μl

[1)] The amount of each primer should be adjusted according to the efficiencies of target amplification.

3. Gently mix and briefly centrifuge the reaction mix.

4. Perform the Real-time PCR.

※ Instrument compatibility

- 7500 Real-Time PCR System (Applied Biosystems)
- CFX96 Real-Time PCR Detection System (Bio-Rad)
- Rotor-Gene Q (QIAGEN)
- LightCycler (Roche)
- Mx3000P (Stratagene)

Products

Cat. No.	Products	Size
PQL-S200	RealHelix™ *Premier* qPCR Kit [Green, Low ROX]	200rxns
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