AccuPower® *RocketPlex*™ RT-PCR PreMix

[Cat. No.] Please refer to the **Ordering Information**

Introduction

AccuPower® *RocketPlex*™ RT-PCR PreMix utilizes one-step RT-PCR performing cDNA synthesis and PCR of two or more target genes (up to 10 targets) in a single step, in a single tube. It can effectively synthesize cDNA with complex secondary RNA structures by using *RocketScript*™ Reverse Transcriptase (*RocketScript*™ RTase) developed by BIONEER. *RocketScript*™ RTase originated and engineered from *M-MLV* reverse transcriptase can provide increased thermal stability resulting in effective full length first strand cDNA synthesis. It can be used at various temperatures (42-70°C) allowing increased specificity, higher yields of cDNA, and more full-length product than other reverse transcriptase. This product contains vacuum-dried components including *RocketScript*™ RTase, reaction buffer, DTT, dNTPs, RNase inhibitor, and HotStart *Taq* DNA Polymerase. It simplifies preparation of reverse transcription reaction mixture and PCR mixture by adding template RNA and primers without any extra process. After the reaction, samples can be applied directly on agarose gel for analysis due to included tracking dye.

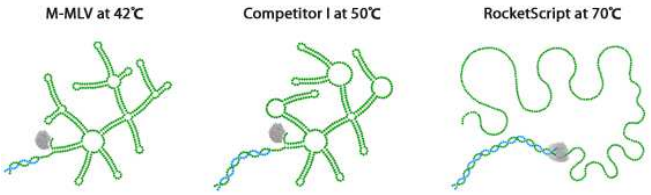


Figure 1. Schematic diagram of 5' UTR of a gene, with complex secondary structure, at three different temperatures.
RocketScript™ Reverse Transcriptase is fully active at 70°C, so it can synthesize complete gene sequences that *M-MLV* and other reverse transcriptase can not synthesize.

Applications

- Multiplex RT-PCR
- Low copy detection
- Gene-expression analysis

Features & Benefits

- Multiplex RT-PCR: Simultaneous cDNA synthesis and amplification up to 10 target genes.
- Specificity: Minimized non-specific amplification and maximized PCR efficiency by using HotStart *Taq* DNA Polymerase.
- Ease-of-use: Reactants are individually packaged in each of the PCR tubes, it allows any user simply perform cDNA synthesis and PCR by adding template RNA and primers. In addition, tracking dye and sedimentation agents are included in products for electrophoresis.

Composition

Composition	Concentration
RocketScript™ Reverse Transcriptase	200 U
5X Reaction buffer	1X
DTT	0.25 mM
dNTPs (dATP, dCTP, dGTP, dTTP)	Each 250 μM
RNase inhibitor	1 U
HotStart *Taq* DNA Polymerase	1 U
Stabilizer and tracking dye	1X

Specifications

HotStart *Taq* DNA Polymerase	
5' to 3' exonuclease activity	Yes
3' to 5' exonuclease activity	No
3'–A overhang	Yes
Fragment size	Up to 1 kb

Storage

Store at -20°C. If stored in the recommended temperature, this product will be stable until the expiration date printed out on the label.

Online Resources

Korean **English**

Visit our **product page** for additional information and protocols

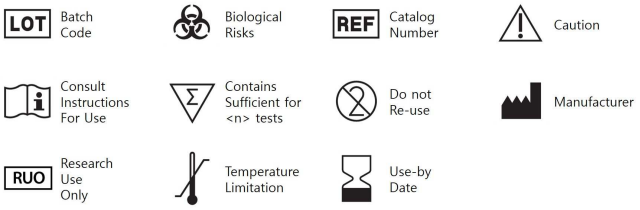
Ordering Information

Description			Cat. No.
0.2 ml thin-wall 8-tube strips with attached cap	96 tubes	20 μl/rxn	K-2211
		50 μl/rxn	K-2213
	480 tubes	20 μl/rxn	K-2212
		50 μl/rxn	K-2214

Notice

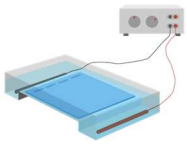
BIONEER corporation reserves the right to make corrections, modifications, improvements and other changes to its products, services, specifications or product descriptions at any time without notice.

Explanation of Symbols



Experimental Procedures

<table>
<tr><th colspan="2">Steps</th><th>Procedure Details</th></tr>
<tr><td>1</td><td> 
Preparation of reaction mixture</td><td>1. Add template RNA, primers and nuclease-free water into AccuPower® RocketPlex™ RT-PCR PreMix tubes to make a total volume of 20 µl or 50 µl. Do not include the dried pellet.

• Amount of template RNA and primers
<table><tr><th colspan="2">Components</th><th>20 µl reaction</th><th>50 µl reaction</th></tr><tr><td rowspan="2">Template RNA</td><td>Total RNA</td><td>10 pg-5 µg</td><td>10 pg-5 µg</td></tr><tr><td>Poly(A) RNA</td><td>10 pg-5 µg</td><td>10 pg-5 µg</td></tr><tr><td>Primers</td><td>Gene specific primer</td><td>5-10 pmol</td><td>5-10 pmol</td></tr></table>
2. Dissolve the vacuum-dried pellet by pipetting or vortexing, and briefly spin down.</td></tr>
<tr><td>2</td><td> 
RT-PCR</td><td>3. Perform the reaction under the following conditions.
<table><tr><th>Step</th><th>Temperature</th><th>Time</th><th>Cycles</th></tr><tr><td>cDNA synthesis</td><td>42-70°C</td><td>10-60 min</td><td>1 cycle</td></tr><tr><td>Pre-denaturation</td><td>95°C</td><td>5 min</td><td>1 cycle</td></tr><tr><td>Denaturation</td><td>95°C</td><td>10-30 sec</td><td></td></tr><tr><td>Annealing</td><td>50-65°C</td><td>10-30 sec</td><td>30 cycles</td></tr><tr><td>Extension</td><td>72°C</td><td>1 kb/min</td><td></td></tr><tr><td>Final extension</td><td>72°C</td><td>5 min</td><td>1 cycle</td></tr></table>* Note: Recommended temperature of reverse transcription reaction is 50°C.</td></tr>
<tr><td>3</td><td> 
Analyze with gel electrophoresis</td><td>4. After the reaction, maintain the reaction mixture at 4°C. The samples can be stored at -20°C until use.

5. Load samples on agarose gel without adding a loading-dye mixture, and perform gel electrophoresis for analysis.</td></tr>
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