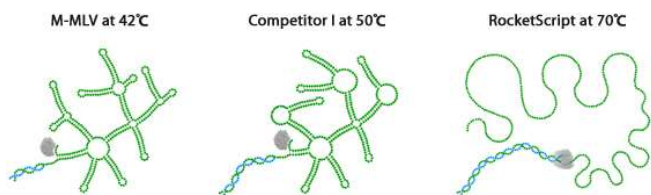


[Cat. No.] **K-2205, K-2206, K-2207**

## Introduction

AccuPower® RocketScript™ Cycle RT PreMix (dN<sub>6</sub>) is a next generation cDNA synthesis kit that uses RocketScript™ Reverse Transcriptase (RocketScript™ RTase) and Cyclic Temperature Reverse Transcription (CTRT) technologies 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. In addition, the CTRT not only can increase the efficiency, but also effective for full-length cDNA synthesis. The CTRT reaction is composed of 2 or 3 steps. The step 1 is performed at 15-25°C, at which short primers are fully annealed. Then, the step 2 is performed at 42-48°C for cDNA synthesis. The step 3 is performed at high temperature 50-55°C at which secondary structure of template RNA obstructing reverse transcription is released. This product contains vacuum-dried components essential for cDNA synthesis including RocketScript™ RTase, reaction buffer, DTT, dNTPs, RNase inhibitors, and Oligo dN<sub>6</sub>. It simplifies preparation of reverse transcription reaction mixture by adding template RNA and nuclease-free water without any extra process.



**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

- First-strand synthesis of cDNA from RNA molecules (RT)
- Random priming reaction
- cDNA library construction
- Probe labeling
- mRNA 5'-end mapping by primer extension analysis
- PCR
- Real-time PCR

## Features & Benefits

- **Sensitivity & Efficiency:** RocketScript™ RTase and CTRT technologies provide enhanced sensitivity synthesizing cDNA from low concentration RNA and increased efficiency by applying circulating temperature reaction.
- **Thermostable activity:** RocketScript™ RTase has excellent thermal resistance even at 70°C, so it is suitable for cDNA synthesis of secondary RNA structures. The cDNA synthesis temperatures can be set at various temperatures (42-70°C) according to user's experimental conditions, allowing effective cDNA synthesis.
- **Flexible Reaction:** As well as CTRT, reverse transcription reaction is possible at a single temperature within 22-55°C.

## 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 inhibitors	1 U
Oligo dN <sub>6</sub>	100 pmol

## Specifications

RocketScript™ Reverse Transcriptase	
DNase activity	No
RNase activity	No
Fragment size	Up to 10 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

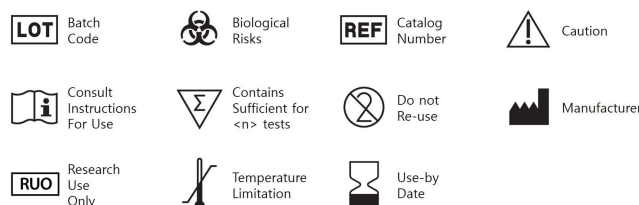
## Ordering Information

Description	Cat. No.
0.2 ml thin-wall 8-tube strips with attached cap	20 μl/rxn K-2205
	50 μl/rxn K-2207
	480 tubes 20 μl/rxn K-2206




## 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**

Steps		Procedure Details																																																			
1	 <b>Preparation of reaction mixture</b>	<p>1. Add template RNA and nuclease-free water into <i>AccuPower® Rocketscript™</i> Cycle RT PreMix (dN<sub>6</sub>) tubes to make a total volume of 20 µl or 50 µl. Do not include the dried pellet.</p> <ul style="list-style-type: none"> <li>Amount of template RNA</li> </ul> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th colspan="2">Components</th> <th>20 µl reaction</th> <th colspan="2">50 µl reaction</th> </tr> </thead> <tbody> <tr> <td rowspan="2" style="text-align: center;">Template RNA</td> <td style="text-align: center;">Total RNA</td> <td style="text-align: center;">0.01-5 µg</td> <td colspan="2" style="text-align: center;">0.01-5 µg</td> </tr> <tr> <td style="text-align: center;">Poly(A) RNA</td> <td style="text-align: center;">0.01-5 µg</td> <td colspan="2" style="text-align: center;">0.01-5 µg</td> </tr> </tbody> </table> <p>2. Dissolve the vacuum-dried pellet by pipetting or vortexing, and briefly spin down.</p>				Components		20 µl reaction	50 µl reaction		Template RNA	Total RNA	0.01-5 µg	0.01-5 µg		Poly(A) RNA	0.01-5 µg	0.01-5 µg																																			
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2	 <b>cDNA synthesis</b>	<p>3. Perform the reaction under the following conditions.</p> <p>3-1. CTRT reaction (Example 1)</p> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th>Step</th> <th>Temperature</th> <th>Time</th> <th>Cycles</th> </tr> </thead> <tbody> <tr> <td>Primer annealing</td> <td style="text-align: center;">15°C</td> <td style="text-align: center;">20 sec</td> <td></td> </tr> <tr> <td>cDNA synthesis</td> <td style="text-align: center;">50°C</td> <td style="text-align: center;">4 min</td> <td style="text-align: center;">10 cycles or more</td> </tr> <tr> <td>Melting secondary structure &amp; cDNA synthesis</td> <td style="text-align: center;">60°C</td> <td style="text-align: center;">20 sec</td> <td></td> </tr> <tr> <td>Heat inactivation</td> <td style="text-align: center;">95°C</td> <td style="text-align: center;">5 min</td> <td style="text-align: center;">1 cycle</td> </tr> </tbody> </table> <p>3-2. CTRT reaction (Example 2)</p> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th>Step</th> <th>Temperature</th> <th>Time</th> <th>Cycles</th> </tr> </thead> <tbody> <tr> <td>Primer annealing</td> <td style="text-align: center;">15°C</td> <td style="text-align: center;">1 min</td> <td></td> </tr> <tr> <td>Melting secondary structure &amp; cDNA synthesis</td> <td style="text-align: center;">42-70°C</td> <td style="text-align: center;">4 min</td> <td style="text-align: center;">10 cycles or more</td> </tr> <tr> <td>Heat inactivation</td> <td style="text-align: center;">95°C</td> <td style="text-align: center;">5 min</td> <td style="text-align: center;">1 cycle</td> </tr> </tbody> </table> <p>* <b>Note:</b> For difficult or high GC-content templates, perform cDNA synthesis at 55°C.</p> <p>3-3. Single temperature reaction (Example 3)</p> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th>Step</th> <th>Temperature</th> <th>Time</th> <th>Cycles</th> </tr> </thead> <tbody> <tr> <td>cDNA synthesis</td> <td style="text-align: center;">22-55°C</td> <td style="text-align: center;">30-60 min</td> <td style="text-align: center;">1 cycle</td> </tr> <tr> <td>Heat inactivation</td> <td style="text-align: center;">95°C</td> <td style="text-align: center;">5 min</td> <td style="text-align: center;">1 cycle</td> </tr> </tbody> </table> <p>* <b>Note:</b> Recommended temperature is range of 42-48°C.</p> <p>4. After the reaction, maintain the reaction mixture at 4°C. The samples can be stored at -20°C until use.</p>				Step	Temperature	Time	Cycles	Primer annealing	15°C	20 sec		cDNA synthesis	50°C	4 min	10 cycles or more	Melting secondary structure & cDNA synthesis	60°C	20 sec		Heat inactivation	95°C	5 min	1 cycle	Step	Temperature	Time	Cycles	Primer annealing	15°C	1 min		Melting secondary structure & cDNA synthesis	42-70°C	4 min	10 cycles or more	Heat inactivation	95°C	5 min	1 cycle	Step	Temperature	Time	Cycles	cDNA synthesis	22-55°C	30-60 min	1 cycle	Heat inactivation	95°C	5 min	1 cycle
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	 <b>Option</b>	<ul style="list-style-type: none"> <li>If PCR is needed, transfer 2-5 µl of reaction mixture (synthesized cDNA) into <i>AccuPower®</i> PCR PreMix tubes (Cat. No. K-2012, not provided), and perform the reaction under the following conditions.</li> </ul> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th>Step</th> <th>Temperature</th> <th>Time</th> <th>Cycles</th> </tr> </thead> <tbody> <tr> <td>Pre-denaturation</td> <td style="text-align: center;">95°C</td> <td style="text-align: center;">5 min</td> <td style="text-align: center;">1 cycle</td> </tr> <tr> <td>Denaturation</td> <td style="text-align: center;">95°C</td> <td style="text-align: center;">20 sec</td> <td></td> </tr> <tr> <td>Annealing</td> <td style="text-align: center;">45-65°C</td> <td style="text-align: center;">20 sec</td> <td style="text-align: center;">25-35 cycles</td> </tr> <tr> <td>Extension</td> <td style="text-align: center;">72°C</td> <td style="text-align: center;">0.5-1 min/kb</td> <td></td> </tr> <tr> <td>Final extension</td> <td style="text-align: center;">72°C</td> <td style="text-align: center;">3-5 min</td> <td style="text-align: center;">1 cycle</td> </tr> </tbody> </table> <p>* <b>Note:</b> For maximum yield and specificity, temperatures and cycling times should be optimized for each new template DNA or primers.</p>				Step	Temperature	Time	Cycles	Pre-denaturation	95°C	5 min	1 cycle	Denaturation	95°C	20 sec		Annealing	45-65°C	20 sec	25-35 cycles	Extension	72°C	0.5-1 min/kb		Final extension	72°C	3-5 min	1 cycle																								
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