# *AccuPower*® *GreenStar*™ RT-qPCR PreMix (V4/2022-05-02)

## [Cat. No.] K-6400

## Introduction

*AccuPower*® *GreenStar*™ RT-qPCR PreMix is a one-step RT-qPCR product based on intercalating dye method using thermostable reverse transcriptase (RTase) and HotStart PCR technology. By applying BIONEER's *RocketScript*™ RTase which is enhanced thermal stability and processivity, efficient reverse transcription (RT) of RNA molecules with complex secondary structures is possible. In real-time PCR, antibody-based HotStart *Taq* DNA Polymerase provides superior priming accuracy and reduced non-specific reactions such as mis-priming and primer dimer during PCR at a low temperature. This product contains vacuum-dried all components for real-time PCR, except for template RNA and target-specific primers. By just adding template RNA and target-specific primers, reproducible results with high sensitivity and specificity can be obtained.

## Applications

- Low copy viral/bacterial pathogen load determination in an earlier stage
- Low copy mRNA amplification
- Low copy target RNA quantification
- RNA amplification for microarray and/or for NGS

## Features & Benefits

- High sensitivity: Amplification of target gene present in a miniscule amount of 1 pg template RNA.
- High specificity: Minimized experimental errors by non-specific amplification and effective amplification of template RNA existing in a small amount by using HotStart *Taq* DNA Polymerase and thermostable RTase.
- Advanced performance: Comprehensive choice of template RNA for RT-qPCR, even complex secondary structure, by using thermostable *RocketScript*™ RTase, capable of performing RT at high temperature.
- Convenience: Reactants are individually packaged in each of the PCR tubes, it allows any user simply perform one-step RT-qPCR by adding template RNA and target-specific primers.
- Reproducibility: Mass production under ISO 9001 quality system allows minimized deviation between lots and reproducible results in replicated tests performed under same conditions and variation.

## Components

| Components | K-6400 |
|---|---|
| Tube/Plate | 96 tubes |
| DEPC-D.W. | 1.8 ml x 4 ea |

## Composition

| Composition | Concentration |
|---|---|
| HotStart *Taq* DNA Polymerase | 1 U |
| *RocketScript* Reverse Transcriptase | 200 U |
| Intercalating dye | 0.4X |
| Reaction buffer with 1.5 mM $MgCl_2$ | 1X |
| dNTPs (dATP, dCTP, dGTP, dTTP) | 1 mM |

## Specifications

| HotStart *Taq* DNA Polymerase | |
|---|---|
| 5' to 3' exonuclease activity | Yes |
| 3' to 5' exonuclease activity | No |
| 3'–A overhang | Yes |

## 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

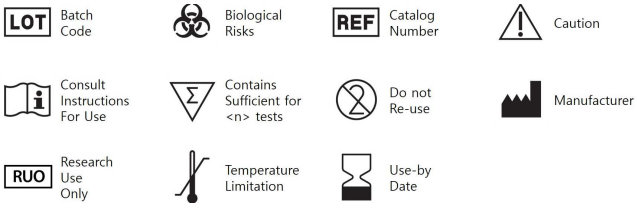
## Ordering Information

| Description | | | | | Cat. No. |
|---|---|---|---|---|---|
| Exicycler | 8-tube strips | 50 µl | optical film included | 96 rxn | K-6400 |

## 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

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## Experimental Procedures

<table>
<tr><th colspan="2">Steps</th><th>Procedure Details</th></tr>
<tr><td>1</td><td>  <br>Preparation of reaction mixture</td><td>
1. Add template RNA, target-specific primers, 50X ROX dye (optional, not provided), and DEPC-D.W. into AccuPower® GreenStar™ RT-qPCR PreMix tubes to make a total volume of 50 µl. Do not include the dried pellet.<br><br>
• Preparation of reaction mixture
<table>
<tr><th>Components</th><th>50 µl reaction</th></tr>
<tr><td>Template RNA (10 pg-100 ng)</td><td>Variable</td></tr>
<tr><td>Forward primer (10 pmol/µl)</td><td>0.5-3 µl</td></tr>
<tr><td>Reverse primer (10 pmol/µl)</td><td>0.5-3 µl</td></tr>
<tr><td>(Optional) 50X ROX dye*</td><td>1 µl</td></tr>
<tr><td>DEPC-D.W.</td><td>Variable</td></tr>
<tr><td>Total volume</td><td>50 µl</td></tr>
</table>
* Note: ROX dye is used for normalization of intensity by background subtraction. The use of ROX dye is recommended for Applied Biosystems 7500 Real-Time PCR System (Applied Biosystems), but not required for Exicycler™ 96 Real-Time PCR System (BIONEER) and CFX96 Real-Time PCR System (Bio-Rad).<br><br>
2. Seal real-time PCR tubes with adhesive optical sealing film (Cat. No. 3111-4110, provided).<br><br>
3. Dissolve the vacuum-dried pellet by vortexing, and briefly spin down.
</td></tr>
<tr><td>2</td><td>  <br>RT-qPCR</td><td>
4. 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>50-70°C†</td><td>15 min</td><td>1 cycle</td></tr>
<tr><td>Pre-denaturation</td><td>95°C</td><td>3-5 min</td><td>1 cycle</td></tr>
<tr><td>Denaturation</td><td>95°C</td><td>5-30 sec</td><td rowspan="2">40-45 cycles</td></tr>
<tr><td>Annealing & Extension</td><td>55-60°C</td><td>5-30 sec</td></tr>
</table>
* Note: Users can adjust the protocol according to their instrument and template sequences to get optimal results.<br>
† For cDNA synthesis, it is recommended to start your reaction at least at 50°C, but not exceed at 70°C.<br><br>
5. After the reaction is completed, analyze the results.
</td></tr>
</table>

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