

**[Cat. No.]** Please refer to the **Online Resources**

## Introduction

BIONEER's *AccuTarget™* miRNA Mimics are chemically synthesized double-stranded RNA oligonucleotides and are available for 875 human mature miRNAs in the miRBase Sequence Database. *AccuTarget™* miRNA Inhibitors are the single-stranded synthetic inhibitor targeting all human miRNAs in the miRBase Sequence Database. These miRNA mimics & inhibitors are available at 5, 10, and 20 nmol guaranteed yield. We also offer miRNA mimics and inhibitors library sets consisting of predesigned mimics or inhibitors at various small scaled (0.25, 0.5, 1, or 2 nmol) in a 96-well plate layout to meet the needs of individual customers. In addition, flexible miRNA library sets for customer-specified mimics & inhibitors are also available for the minimum order of 48 ea.

## Features & Benefits

- Ready-to-transfect miRNA mimics: Same activity as the endogenous miRNA actually present in the cell after transfection.
- Wide application: miRNA inhibitors for studying the loss-of-function of miRNAs by inhibiting the activity of target miRNAs.

## Specifications

- miRNA mimics and inhibitors can be ordered from the following: 5, 10, or 20 nmol guaranteed yields.
- A library set composed of predesigned miRNA mimics or inhibitors of various sizes (0.25, 0.5, 1, 2 nmol) is provided on a 96-well plate.
- For customers buying 48 or more orders, flexible miRNA library sets can be purchased.
- All miRNA mimics are provided in the form of double-stranded siRNA.
- All miRNA inhibitors are provided in the form of single-stranded miRNA, being an anti-sense strand to the target miRNA.
- An economical choice for a guaranteed high quality of miRNA mimics and inhibitors with the use of automated HPLC and Bio-RP purification.

## Quality Control

- All the sense and anti-sense miRNAs are inspected with MALDI-TOF analysis and annealed double-stranded miRNA is analyzed by non-denaturing PAGE to confirm proper annealing.

## 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.

## Precautions

- Wear gloves and a mask when handling *AccuTarget™* miRNAs mimics and inhibitors.
- Always use RNase free tubes and pipette tips.
- Only use DEPC-D.W. (or RNase free water) when diluting *AccuTarget™* miRNA Mimics and Inhibitors.

## Online Resources



Korean



English

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

## 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



Caution



Consult  
Instructions  
For Use


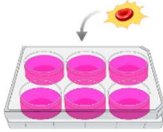



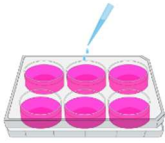

Do not  
Re-use



Use-by  
Date

**Experimental Procedures**

Steps		Procedure Details																					
<b>Resuspension Protocol</b>																							
1	 <p><b>Resuspension of miRNA</b></p>	<ol style="list-style-type: none"> <li>Briefly centrifuge tubes (or multi-well plates) containing miRNA mimics or inhibitors to ensure that the miRNA pellet is located at the bottom of the tube.</li> <li>Dissolve miRNAs to a convenient stock concentration using the recommended volume of DEPC-D.W. (or RNase-free water) shown in Table 1.</li> <li>Mix the miRNAs by pipetting or vortexing briefly and spin down.</li> <li>Store at -20°C in small aliquots and avoid repeated freeze and thaw cycles.</li> </ol> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th rowspan="2">Amount of miRNA (nmol)</th> <th colspan="2">Volume of DEPC-D.W. for desired final concentration</th> </tr> <tr> <th>100 µM stock</th> <th>20 µM stock</th> </tr> </thead> <tbody> <tr> <td style="text-align: center;">1.0</td> <td style="text-align: center;">10</td> <td style="text-align: center;">50</td> </tr> <tr> <td style="text-align: center;">5.0</td> <td style="text-align: center;">50</td> <td style="text-align: center;">250</td> </tr> <tr> <td style="text-align: center;">10.0</td> <td style="text-align: center;">100</td> <td style="text-align: center;">500</td> </tr> <tr> <td style="text-align: center;">20.0</td> <td style="text-align: center;">200</td> <td style="text-align: center;">1000</td> </tr> </tbody> </table> <p><b>Table 1. Recommended volumes and concentrations for miRNA resuspension.</b></p>	Amount of miRNA (nmol)	Volume of DEPC-D.W. for desired final concentration		100 µM stock	20 µM stock	1.0	10	50	5.0	50	250	10.0	100	500	20.0	200	1000				
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<b>Transfection Protocol</b>																							
<ul style="list-style-type: none"> <li>We used Lipofectamine® RNAiMAX (Thermo Fisher) and a HeLa cell for the validation of our miRNA.</li> <li>This protocol is optimized for transfection in a 6-well culture plate format.</li> <li>To perform transfection in different cell culture formats, refer to Table 2 and the manufacture's Lipofectamine® RNAiMAX protocol.</li> </ul>																							
1	 <p><b>Preparation of cells</b></p>	<ol style="list-style-type: none"> <li>One day before transfection, plate <math>0.25\text{-}1 \times 10^6</math> cells (adherent cells) in each well with 2 ml of growth medium without antibiotics so that they will be 60-80% confluent at the time of transfection.</li> </ol> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th>Culture Vessel</th> <th>Relative surface area*</th> <th>Volume of plating medium</th> </tr> </thead> <tbody> <tr> <td style="text-align: center;">96-well</td> <td style="text-align: center;">0.2 cm<sup>2</sup></td> <td style="text-align: center;">100 µl</td> </tr> <tr> <td style="text-align: center;">48-well</td> <td style="text-align: center;">0.4 cm<sup>2</sup></td> <td style="text-align: center;">250 µl</td> </tr> <tr> <td style="text-align: center;">24-well</td> <td style="text-align: center;">1 cm<sup>2</sup></td> <td style="text-align: center;">500 µl</td> </tr> <tr> <td style="text-align: center;">6-well</td> <td style="text-align: center;">5 cm<sup>2</sup></td> <td style="text-align: center;">2 ml</td> </tr> <tr> <td style="text-align: center;">60 mm</td> <td style="text-align: center;">10 cm<sup>2</sup></td> <td style="text-align: center;">5 ml</td> </tr> <tr> <td style="text-align: center;">100 mm</td> <td style="text-align: center;">30 cm<sup>2</sup></td> <td style="text-align: center;">10 ml</td> </tr> </tbody> </table> <p><b>Table 2. Relative surface area of <i>in vitro</i> cell culture dish and culture media volume.</b></p> <p>* Surface area may vary according to manufacturers.</p> <ol style="list-style-type: none"> <li>Remove the growth medium from the cells.</li> <li>Add 500 µl of fresh growth medium without serum.</li> </ol>	Culture Vessel	Relative surface area*	Volume of plating medium	96-well	0.2 cm <sup>2</sup>	100 µl	48-well	0.4 cm <sup>2</sup>	250 µl	24-well	1 cm <sup>2</sup>	500 µl	6-well	5 cm <sup>2</sup>	2 ml	60 mm	10 cm <sup>2</sup>	5 ml	100 mm	30 cm <sup>2</sup>	10 ml
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<p>2</p>	 <p><b>Preparation of mixture</b></p>	<p>4. For each well to be transfected, prepare miRNA-Lipofectamine® RNAiMAX complexes as follows.</p> <p>4-1. Dilute 3 µl of miRNA (10 µM stock) in 150 µl of growth medium without serum (or Opti-MEM® I Reduced Serum medium) and mix gently.</p> <p>4-2. Prepare diluted Lipofectamine® RNAiMAX before use. Add 9 µl of Lipofectamine® RNAiMAX in 150 µl of growth medium without serum (or Opti-MEM® I Reduced Serum medium). Incubate for 5 min at room temperature.</p> <p>4-3. Combine the diluted miRNA duplex with diluted Lipofectamine® RNAiMAX (1:1 ratio). Gently mix and incubate for 20 min at room temperature.</p>
<p>3</p>	 <p><b>Add mixture and incubate cells</b></p>	<p>5. Add 250 µl of the mixture (miRNA duplex with Lipofectamine® RNAiMAX) to each well of 6-well plate containing cells. The final volume in each well is 1 ml. The amount of miRNA used per well is 25 pmol. Mix gently with hands by rocking the plate back and forth.</p> <p>6. Incubate the cells for 5-6 hrs at 37°C in CO<sub>2</sub> incubator.</p>
<p>4</p>	 <p><b>Analyze transfected cells</b></p>	<p>7. Change the medium with a fresh one containing serum and incubate the cells 24-48 hrs until you are ready to analyze for miRNA functional studies.</p>