

# miExpress™ microRNA synthetic mimics

## • Product Description:

GeneCopoeia's miExpress™ miRNA synthetic mimics are sequence-specific and chemically modified double-stranded RNA oligonucleotides that mimic mature endogenous miRNAs after transfection into cells. These mimics simulate the high level expression of mature miRNAs in cells to enhance the regulation of endogenous miRNA for gain-of-function studies.

## Applications:

- 1. Simulate miRNA activity to study gain-of-function effects
- 2. Identify and validate miRNA targets
- 3. Elucidate the role of miRNA in cellular processes and pathological pathways
- 4. Identification and validation of diagnostic and therapeutic miRNA biomarkers and targets

# Shipping and Storage:

miExpress<sup>TM</sup> miRNA synthetic mimics are shipped lyophilized at room temperature. Upon receipt, the miExpress<sup>TM</sup> miRNA synthetic mimics should be stored at -20°C or lower. Under these conditions, they are stable for at least 12 months.

#### • Precautions:

RNA oligonucleotides are susceptible to enzymatic degradation by nucleases and to chemical degradation by extreme pH and temperatures. We recommend wearing gloves, using RNase-free reagents, tubes, and pipette tips to maintain nuclease-free conditions when handling the oligonucleotides.

## • Resuspension:

- 1. Briefly centrifuge the tube containing the miExpress<sup>TM</sup> miRNA synthetic mimics to ensure that the oligonucleotide powder is collected at the bottom of the tube.
- 2. Resuspend the oligonucleotide in nuclease-free, sterile water to achieve the desired concentration (alternatively, an appropriate RNase-free buffer such as PBS may be used). We recommend a stock concentration of at least 20  $\mu$ M (adding 125  $\mu$ L sterile water to 2.5 nmol miRNA mimics will make a 20  $\mu$ M stock solution).
- 3. Gently pipette the solution up and down 3-5 times to resuspend.
- 4. Aliquot the miRNA mimics into small volumes to limit the number of thaw-freeze cycles.
- 5. Store at -20°C or lower. Avoid thaw-freezing more than 5 times. In this form, the oligonucleotide is stable for at least six months.

#### • Transfection:

Efficient delivery of miRNA mimics is critical for miRNA gain-of-function experiments. The transfection efficiency of miExpress<sup>TM</sup> miRNA synthetic mimics varies according to cell type and the transfection reagent used. It is important to select the appropriate transfection reagent for the cell

line under study. GeneCopoeia's EndoFectin™ Max transfection reagent (Catalog#: EF013) provides efficient delivery of small RNAs into various mammalian cells.

The following is a general protocol for use of EndoFectin<sup>TM</sup> Max transfection reagent to transfect miExpress<sup>TM</sup> miRNA synthetic mimics into cultured mammalian cells. The volumes and amounts used are for transfection in a 24-well plate.

#### 1. Plate cells

a) One day before transfection, plate cells in 0.5 mL of growth medium without anithiotics so that they will be 30-70% confluent at the time of transfection.

## 2. Prepare miRNA mimics/ EndoFectin<sup>TM</sup> Max complex

- a) Dilute the required amount of miRNA mimics stock solution into Opti-MEM I<sup>TM</sup> (Invitrogen) or other appropriate protein-free media for a final volume of 25 μL. miExpress<sup>TM</sup> miRNA synthetic mimics typically work well at a final concentrations of 10-50 nM, but a more extensive concentration range from 1-100 nM can be analyzed for optimization.
- b) Dilute 1-3  $\mu$ L of EndoFectin<sup>TM</sup> reagent with the same protein-free diluents for a final volume of 25  $\mu$ L.
- c) Add diluted EndoFectin<sup>™</sup> Max reagent drop-wise to the diluted miRNA mimics; mix by gently flicking the tube or pipetting.
- d) Incubate at room temperature for 10–25 min to allow the miRNA mimics/ EndoFectin™ Max to form.

#### 3. Transfect cells

- a) Add the miRNA mimics/ EndoFectin<sup>TM</sup> Max complex dropwise to the cells and rock the dish back and forth to evenly distribute the complexes.
- b) For transfection in the absence of serum, remove the normal growth medium and replace with serum-free medium, then add the DNA-EndoFectin<sup>TM</sup> Max complex. Add ½ volume of the growth medium containing 30% serum 3 hours after transfection.
- Incubate the cells under normal cell culture conditions for 24-48hr until they are ready for analysis.

To obtain the highest transfection efficiency with minimal cytotoxicity we recommend optimizing the transfection conditions empirically by adjusting the ratio of transfection reagent to RNA oligonucleotide, the cell density at the time of transfection and the length of exposure of cells to transfection reagent/miRNA mimic complex.

## Related Products

miExpress<sup>TM</sup> Precursor miRNA Expression Clones

Over-express miRNA for miRNA gain-of-function studies

miTarget<sup>TM</sup> miRNA 3'UTR Target Sequence Expression Clones

Do functional validation of predicted targets

OmicsLink<sup>TM</sup> Expression-Ready ORF cDNA Clones

Perform a variety of applications with expression-ready clones

All-in-One™ miRNA qRT-PCR Detection Kits

Accurately quantify miRNA expression

EndoFectin<sup>TM</sup> Transfection Reagents

Transfect efficiently and achieve reliable and reproducible results

Questions? Email us at <a href="mailto:support@qenecopoeia.com">support@qenecopoeia.com</a> or call 301-762-0888