

GeneCopoeia miRNA Functional Analysis Tools

Comprehensive

GeneCopoeia miRNA functional analysis tools provide solutions for investigating miRNA gene expression and regulation as well as target identification and validation in virtually all cell types, tissues and animal models.

- ♦ Sequenced verified miRNA clones in viral and non-viral-based vectors
- ♦ Genome-wide miRNA 3' UTR target sequences
- ♦ Efficient transfection reagents
- ♦ Expression-ready ORF cDNA clones
- ♦ Optimized miRNA qRT-PCR detection kit and validated qPCR primers

All-in-One miRNA qRT-PCR Kits and Primers

Robust

Detect and quantify mature miRNA with co-optimized qRT-PCR SYBR® Green detection kits and validated qPCR miRNA primers. A robust formulation provides more universal reaction conditions along with other benefits including:

- ♦ Specific — detect single-base differences
- ♦ Sensitive — detect miRNA in 100 pg of total RNA
- ♦ Practical — detect only mature miRNA
- ♦ Convenient — easy, fast and cost-effective

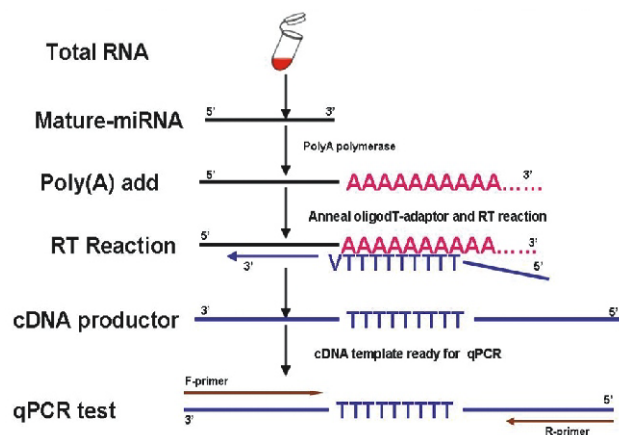


Figure 1. Overview of steps in the All-in-One miRNA qRT-PCR Detection Kit.

All your applications are possible

- Study the regulation of mRNA individually or collectively by transducing or transfecting precursor miRNA expression clones into target cells.
- Identify and validate miRNA targets using miRNA target validation expression constructs.
- Study miRNA expression profiles with validated qPCR primers and qRT-PCR detection kit.
- Restore gene function with expression-ready OmicsLink ORF cDNA clones.

miExpress™ Precursor miRNA Expression Clones

Complete

**Validated All-in-One
miRNA qPCR primers get
the job done.**

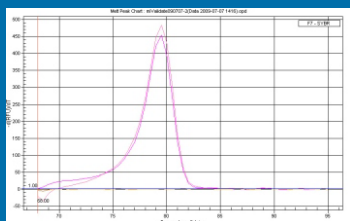
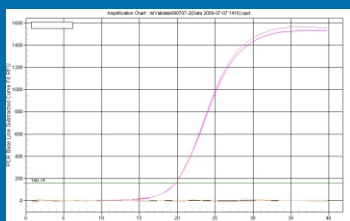


Figure 2. All-in-One miRNA qPCR primers are validated to generate a single amplification of the correct size for the targeted miRNA and to yield a single dissociation curve peak. A cDNA pool, containing reverse transcribed products from 10 human tissue total RNA samples was used as the validation template. qPCR was performed using 0.2 μ M primer with 2 \times All-in-One qPCR Mix. The upper panel shows a validated result for melting curve with the lower showing validated result for amplification curve.

miExpress™ precursor miRNA expression clones are available in non-viral and viral-based vector systems allowing stable or transient transduction of miRNA into virtually all cell types including difficult-to-transfect and non-dividing cells.

- ◆ miExpress miRNA clone libraries cover all known human, mouse and rat miRNAs available in the miRBase database.
- ◆ All clones are fully sequenced.
- ◆ Optimization of expression cassettes allows high expression of precursor miRNA and the maturation of miRNA inside cells.
- ◆ Expression cassettes of all miRNA expression constructs are completely sequenced.
- ◆ Lentiviral-based expression constructs allow efficient transduction of miRNA into non-dividing and difficult-to-transfect target cells.
- ◆ A neomycin or puromycin selection marker enables regulation studies of both long-term and transient expression.
- ◆ Transduction efficiencies are monitored with fluorescent reporter protein.

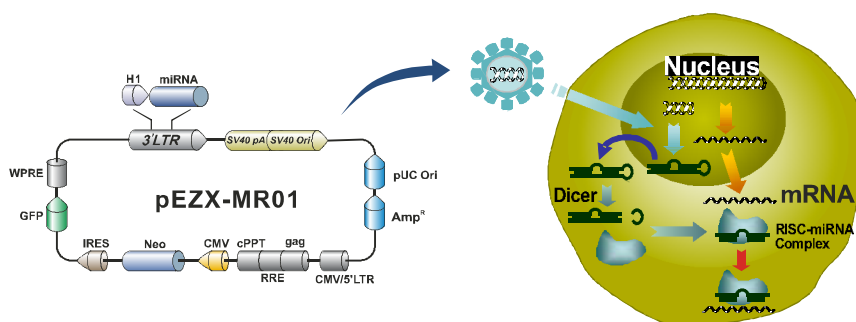


Figure 3. Precursor miRNA constructs expressed with a lentiviral-based vector and their involvement in vector-mediated miRNA gene regulation.

miTarget™ miRNA Target Sequence 3' UTR Expression Clones

Uncomplicated

GeneCopoeia functional analysis tools facilitate miRNA studies with genome-wide miTarget™ miRNA target sequence 3' UTR expression clones. miTarget clones are available in two mammalian expression vector systems.

- ♦ All 3' UTR sequences were obtained from public domain databases.
- ♦ 3' UTR sequences are inserted downstream of the coding sequences.
- ♦ The firefly luciferase (non-secreted) and *Gaussia* luciferase (secreted) reporter genes are controlled by an SV40 promoter.
- ♦ Luciferase expression is regulated by binding of the targeting miRNA to the 3' UTR target sequence; luciferase activity is quantified with a colorimetric assay.
- ♦ Vectors include *Renilla* luciferase or alkaline phosphatase which can be used as a normalization reporter gene.

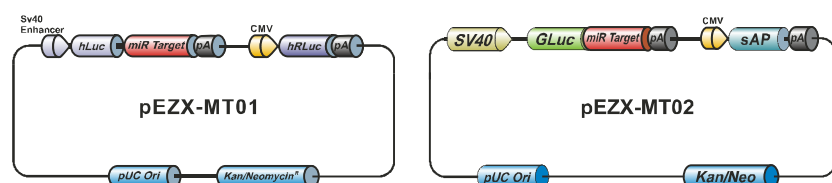


Figure 4. Vector features for human miRNA target sequence expression clones.

The vectors offer different combinations of reporter and tracking genes to match different assay requirements.

Vector	Reporter gene	Tracking gene	Advantage
pEZX-MT01	Firefly luciferase	<i>Renilla</i> luciferase	Assays can be performed on cell lysates.
pEZX-MT02	<i>Gaussia</i> luciferase	Alkaline phosphatase	Target cells are not destroyed to perform enzymatic assays.

Discovery simplified

- The regulatory effect of a particular miRNA on its potential target is assessed with an enzymatic assay for firefly or *Gaussia* luciferase.
- The miRNA 3' UTR expression constructs transcribe chimeric mRNAs consisting of a luciferase coding sequence and a 3' UTR target sequence.
- The reporter enzymatic assays are conducted with either firefly luciferase (non-secreted) or *Gaussia* luciferase (secreted).

Luc-Pair™ miR Luciferase Assay Kit

Ideal

The Luc-Pair™ miR Luciferase Assays were optimized for use with GeneCopoeia miRNA 3' UTR target sequence expression clones and provide an efficient system for measuring firefly and *Renilla* luciferase sequentially.

- ♦ High sensitivity in a dynamic range
- ♦ Successive reporter system
- ♦ Consistent reproducibility
- ♦ Simple assay format; suitable for micro-plates
- ♦ Suitable for high-throughput screening

Luc-Pair assays are easy to use with less hands-on time and provide quick results. Order with your miTarget 3' UTR clone for ideal assay results.

GeneCopoeia	Contents	Description
miExpress™ Precursor miRNA Expression Clones	750 human 450 mouse 270 rat	Study the regulatory and functional effect of miRNAs on corresponding genes and proteins
miTarget™ miRNA Target Validation Expression Clones	25,000 human 25,000 mouse	Cross validate data using luciferase reporter genes
Luc-Pair™ miR Luciferase Assay Kit	Substrates and buffers	Successive measurement of firefly and <i>Renilla</i> luciferase for miRNA target validation
All-in-One™ miRNA Real-Time qPCR Kits	SYBR® Green-based qRT-PCR	Accurate quantification of miRNA expression
All-in-One™ miRNA Real-Time qPCR Primers	700 human, mouse and rat primers	Perform miRNA sequence-specific assays
OmicsLink™ Expression-Ready ORF cDNA Clones	20,000 human 15,000 mouse	Perform gain-of-function studies with expression-ready clones

Visit us online for detail kit information and to search for clones and primers.

Find your Expressway to Discovery™ with GeneCopoeia comprehensive miRNA solutions.

To order

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