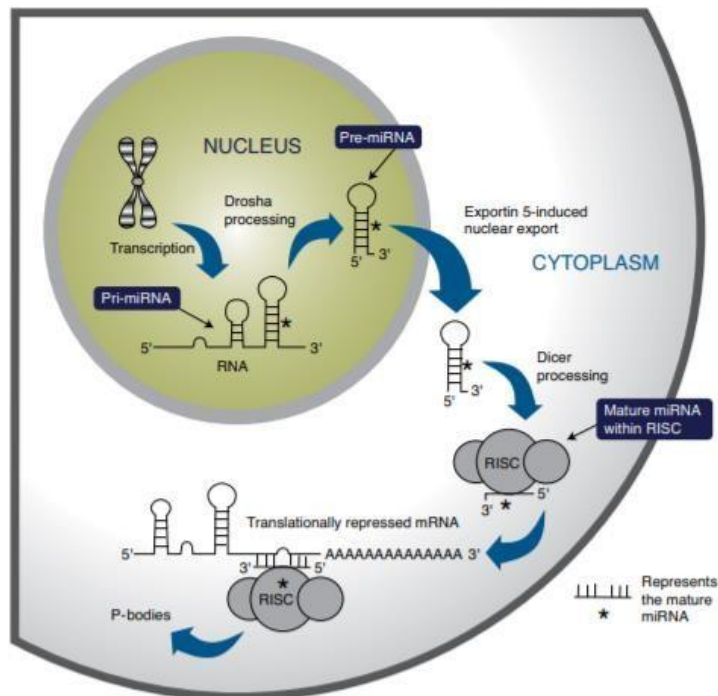


TaqMan® Assays for Primary, Precursor, and Mature MicroRNA

Thermo Fisher Scientific provides three different types of TaqMan® assays uniquely designed to detect the primary, precursor, and mature miRNA. This application note describes the difference and the intended target of these assays. The note also provides the guidance on how to obtain the assays from the company.

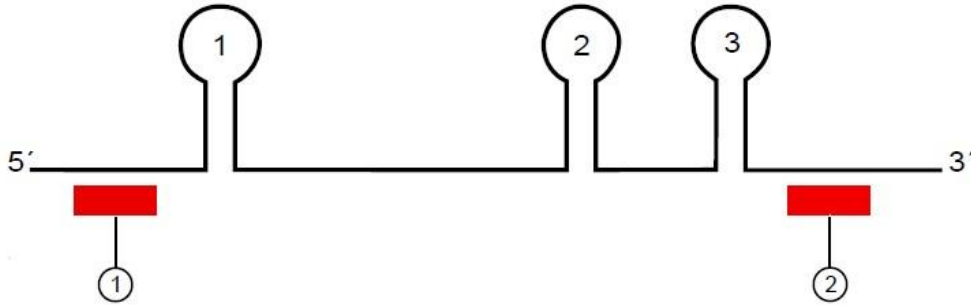
1. MicroRNA Biogenesis¹



MicroRNA genes are transcribed in the nucleus, giving rise to long primary miRNA transcripts (pri-miRNAs). A pri-miRNA sequence contains at least one (more commonly, multiple) stem-loop sequence containing mature miRNA sequences. The pri-miRNA transcript is processed in the nucleus by cleavage at the base of each stem-loop by Drosha. These precursor stem-loop molecules (pre-miRNAs) are then exported to the cytoplasm where they are processed by Dicer prior to the mature sequence being loaded into the RNA-induced silencing complex (RISC). Each of these maturation steps is potentially a target for regulation.

2. TaqMan® Pri-miRNA Assay²

TaqMan® Pri-miRNA Assays are designed with the same algorithms as TaqMan® Gene Expression Assays. The assay target sequence is near the stem-loops that are identified in the Sanger miRBase sequence repository. Each assay targets areas within 500 nucleotides on either side of the stem-loop. In a small portion of cases, stem-loop sequences are sufficiently close so as to prevent design to the intervening sequence. In these cases, we recommend using the nearest available assay.



- ① Stem-loop 1: detected by assay 1
- ② Stem-loops 2 and 3: detected by assay 2

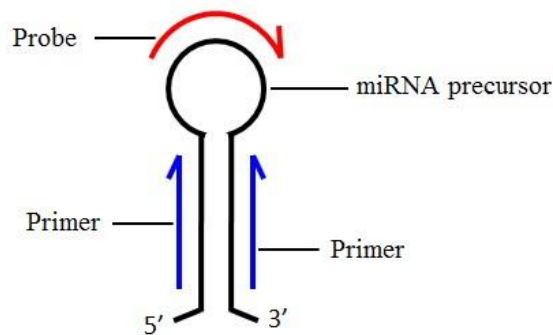
Predesigned TaqMan® pri-miRNA assay may be found by using the miRNA assay search tool on the ThermoFisher website. If there is no predesigned assay available, one can design a custom pri-miRNA assay by using the Custom TaqMan® Gene Expression Assay Design Tool on the website.

Search TaqMan® Assays and Arrays

TaqMan Assays miRNA	*	<input type="button" value="Search"/>	Advanced Search
Format Single-tube assays	miRNA Products Pri-miRNA Assays	Species	<input type="button" value="Enter multiple targets"/> <input type="button" value="Enter target sequence"/>

3. TaqMan® Precursor miRNA assay³

TaqMan® Precursor miRNA Assays are designed with the same algorithms as TaqMan® Gene Expression Assays. The assay target sequence is the precursor miRNA which belongs to the non-coding RNA category.



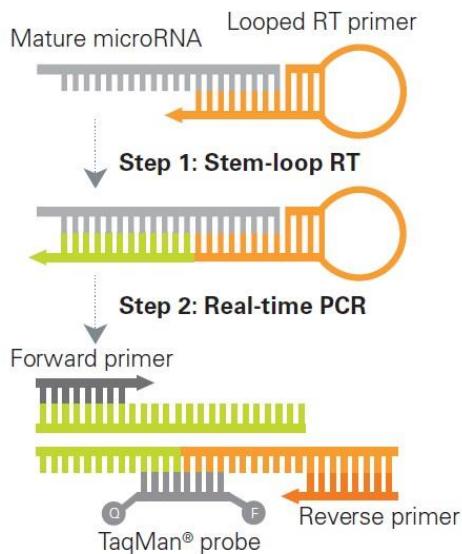
Predesigned TaqMan® precursor miRNA assay may be found by using the TaqMan® gene expression assay search tool on the ThermoFisher website. If there is no predesigned assay available, one can design a custom precursor miRNA assay using Custom TaqMan® Gene Expression Assay Design Tool on the website. The target sequence for designing the assay is the precursor miRNA sequence.

TaqMan Assays Gene Expression	Search TaqMan® Assays and Arrays	<input type="text" value="Search"/>
Format Single-tube assays	Assay Type All Gene Express...	Species
		Enter multiple targets

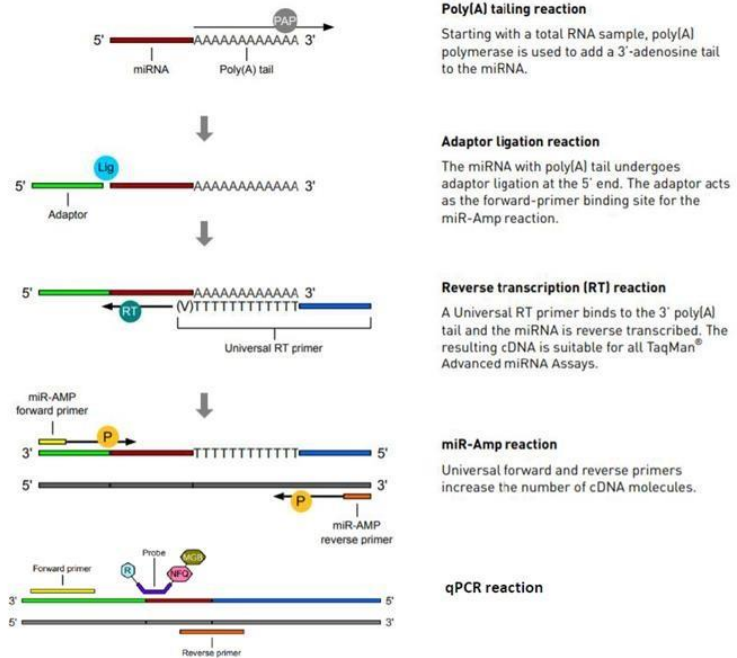
4. TaqMan® mature miRNA assay^{4,5}

There are two types of TaqMan® mature miRNA assay: TaqMan® miRNA assay and TaqMan® advanced miRNA assay. TaqMan® miRNA assay uses miRNA-specific reverse transcription primer to make cDNA and TaqMan® advanced miRNA assay uses universal reverse transcription primer to make cDNA.

TaqMan® miRNA Assay



TaqMan® advanced miRNA Assay



TaqMan® miRNA assay and TaqMan® advanced miRNA assay may be found by using the TaqMan® miRNA assay search tool on the ThermoFisher website. For designing custom TaqMan® miRNA assay, one can use the Custom TaqMan® Small RNA Assay Design Tool on the website. For designing custom TaqMan® advanced miRNA assay, please contact ThermoFisher Scientific's technical support.

TaqMan Assays miRNA	Search TaqMan® Assays and Arrays	<input type="text" value="Search"/>
Format Single-tube assays	miRNA Products miRNA Assays a...	Species
		Enter multiple targets

References

1. TaqMan® Pri-miRNA Assays. Product Brochure, Applied Biosystems (2016).
2. TaqMan® Pri-miRNA Assays User Guide, Rev. E. Applied Biosystems (2019)
3. TaqMan® Gene Expression Assays-single-tube assays User Guide, Rev. Q. Applied Biosystems (2018)
4. TaqMan® Small RNA Assays User Guide, Rev. G. Applied Biosystems (2019)
5. TaqMan® Advanced miRNA Assays User Guide, Rev. C. Applied Biosystems (2016)