

# Megaplex™ Primer Pools

For MicroRNA expression analysis with TaqMan® Array Cards

Pub. No. 4401697 Rev. F

 **WARNING!** Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from [thermofisher.com/support](http://thermofisher.com/support).

## Product description

Applied Biosystems™ Megaplex™ RT Primers streamline the workflow for quantitating up to 380 MicroRNAs in parallel by enabling reverse transcription of RNA to cDNA for all MicroRNAs in a single reaction. They are available for human and rodent (mouse and rat) species. A companion pool of Megaplex™ PreAmp Primers enables an optional preamplification step to be added to the workflow in situations where sample is limiting, or sensitivity is the most important factor. For comprehensive coverage, two pools each (Pool A and Pool B) of Megaplex™ RT Primers and Megaplex™ PreAmp Primers are available for human and rodent species. Real-time PCR can be performed with either individual TaqMan® MicroRNA Assays or with matched TaqMan® MicroRNA Assays on TaqMan® Array Cards.

## Contents and storage

Table 2 Megaplex™ Primer Pool sets

Contents	Cat. No.	Amount	Storage <sup>[1]</sup>
Megaplex™ Primer Pools, Human Pools Set v3.0 Includes: <ul style="list-style-type: none"> <li>• Megaplex™ RT Primers, Human Pool A v2.1</li> <li>• Megaplex™ RT Primers, Human Pool B v3.0</li> <li>• Megaplex™ PreAmp Primers, Human Pool A v2.1</li> <li>• Megaplex™ PreAmp Primers, Human Pool B v3.0</li> </ul>	4444750	50 reactions	Store at -20°C
Megaplex™ Primer Pools, Rodent Pools Set v3.0 Includes: <ul style="list-style-type: none"> <li>• Megaplex™ RT Primers, Rodent Pool A</li> <li>• Megaplex™ RT Primers, Rodent Pool B v3.0</li> <li>• Megaplex™ PreAmp Primers, Rodent Pool A</li> <li>• Megaplex™ PreAmp Primers, Rodent Pool B v3.0</li> </ul>	4444766	50 reactions	Store at -20°C

<sup>[1]</sup> See packaging for expiration date.

Megaplex RT Primers and Megaplex PreAmp Primers can be purchased separately.

Table 3 Megaplex™ RT Primers

Contents	Cat. No.	Amount	Storage <sup>[1]</sup>
Megaplex™ RT Primers, Human Pool A v2.1	4399966	RT primers MgCl <sub>2</sub> 50 reactions	Shipped at ambient temperature. Store at -20°C.
Megaplex™ RT Primers, Human Pool B v3.0	4444281		
Megaplex™ RT Primers, Human Pool Set v3.0 (Contains Pool A v2.1 and Pool B v3.0)	4444745		
Megaplex™ RT Primers, Rodent Pool A	4399970		
Megaplex™ RT Primers, Rodent Pool B v3.0	4444292		
Megaplex™ RT Primers, Rodent Pool Set v3.0 (Contains Pool A and Pool B v3.0)	4444746		

<sup>[1]</sup> See packaging for expiration date.

**Table 4** (Optional) Megaplex™ PreAmp Primers

Contents	Cat. No.	Amount	Storage <sup>[1]</sup>
Megaplex™ PreAmp Primers, Human Pool A v2.1	4399233	50 reactions	Shipped at ambient temperature. Store at -20°C.
Megaplex™ PreAmp Primers, Human Pool B v3.0	4444303		
Megaplex™ PreAmp Primers, Human Pool Set v3.0 (Contains Pool A v2.1 and Pool B v3.0)	4444748		
Megaplex™ PreAmp Primers, Rodent Pool A	4399203		
Megaplex™ PreAmp Primers, Rodent Pool B v3.0	4444308		
Megaplex™ PreAmp Primers, Rodent Pool Set v3.0 (Contains Pool A and Pool B v3.0)	4444747		

<sup>[1]</sup> See packaging for expiration date.

## Workflow

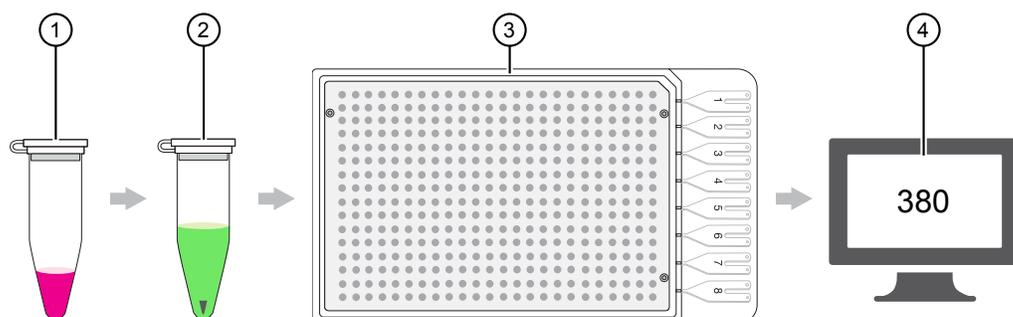


Figure 1 Workflow using Megaplex™ Primer Pools and TaqMan® Array MicroRNA Card

- ① cDNA synthesis using Megaplex™ RT Primers
- ② (Optional) Pre-amplification using Megaplex™ PreAmp Primers
- ③ Real-time PCR using TaqMan® Array Cards
- ④ Data analysis (380 targets)

## Data normalization

Each TaqMan® Array Card of the two-card set (either human or rodent) contains identical candidate endogenous control assays that provide a variety of options for normalization.

Endogeneous control	Species	Notes
Mammalian U6	Human, mouse, and rat	Repeated 4x on card enabling monitoring of assay reproducibility
RNU48	Human	Small nucleolar RNA
RNU44	Human	Small nucleolar RNA
snoRNA135	Mouse	Small nucleolar RNA
snoRNA202	Mouse	Small nucleolar RNA
U87	Rat	Small nucleolar RNA
Y1	Rat	Small non-coding RNA
ath-miR159a	<i>Arabidopsis</i>	Not detectable in mammalian species For use as a negative or processing control

## Megaplex™ assay performance

Megaplex™ RT Primers and Megaplex™ PreAmp Primers are highly multiplexed pools designed to significantly streamline the workflow when profiling many miRNA targets in a single experiment. When combined in a large multiplex pool, a small subset of assays exhibit lower no-template-control (NTC  $C_t$  values <35) values than when run individually. This is not an assay design issue. In the interest of providing the broadest coverage possible, customers have requested that we include these assays on the profiling array, but with the expectation that they are to be considered semi-quantitative. As a result, fold-change measurements using these assays may be less than the true value. To aid data interpretation, the identity of these assays is provided in the Megaplex™ Assay Performance File. If accurate fold-change measurements for these assays is required, we recommend that changes detected using the Megaplex™ assay workflow be validated using the corresponding individual TaqMan® MicroRNA Assay.

## Megaplex™ Assay Performance File

The Megaplex™ Assay Performance File is found in the **Product literature** section of each TaqMan® Array Card page at [thermofisher.com](http://thermofisher.com).

File	TaqMan® Array Card Cat. No.
Megaplex™ Assay Performance File Human Card A	4398965
Megaplex™ Assay Performance File Human Card B	4444910
Megaplex™ Assay Performance File Rodent Card A	4398967
Megaplex™ Assay Performance File Rodent Card B	4444899

## Related documentation

Publication	Pub. No.
<i>Megaplex™ Primer Pools for MicroRNA expression analysis User Guide</i>	4399721
<i>Megaplex™ Primer Pools for MicroRNA expression analysis Quick Reference</i>	4399813

## Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at [www.thermofisher.com/us/en/home/global/terms-and-conditions.html](http://www.thermofisher.com/us/en/home/global/terms-and-conditions.html). If you have any questions, please contact Life Technologies at [www.thermofisher.com/support](http://www.thermofisher.com/support).



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**Revision history:** Pub. No. 4401697

Revision	Date	Description
F	1 August 2018	Updated products. Updated workflow diagram.
E	21 March 2016	Format, style, and legal updates.
D	09 June 2010	Baseline for this revision history.

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