PR1MA™ qMAX cDNA Synthesis Kit

Description

High quality, qPCR ready DNA is quickly generated with the PR1MA gMax cDNA Synthesis kit. A blend of hexamer and oligo (dT) primers ensures unbiased and reproducible synthesis with true, relative cDNA representation. The exceptionally thermostable gMax Reverse Transcriptase combined with a potent RNase inhibitor allows for transcription to take place at higher temperatures (alleviating secondary structure problems) and maintains the integrity of the total RNA. The 5X buffer provides the perfect environment for highly efficient cDNA synthesis and greater yields. The buffer is optimized for superior data accuracy and reproducibility with limited starting material and low-copy templates.

-Unbiased, complete sequence representation for pg to 2µg of total RNA -Thermostable reverse transcriptase and optimized next generation buffer system provide consistent, high yield product

Upon receipt, immediately store at -20°C. Avoid excessive freeze/thaw cycles. When stored as directed, this product will retain its activity for 12 months from date of receipt. The product may also be stored at 4°C for up to one month.

Limitations of Use

For research purposes only. Not intended for therapeutic or diagnostic use.

Quality Control

PR1MA enzymes and reagents are tested under general assay conditions for activity, reproducibility, efficiency, heat activation, sensitivity, and absence of nuclease contamination and nuclease activity. This product is manufactured under a comprehensive quality management system, following ISO 9001:2008 standards.

MidSci is not responsible for consequential or incidental damages, direct or indirect, resulting from use of this product. MidSci guarantees the performance of this product as described when used in accordance with these instructions.

General Guidelines

Buffer

The cDNA Synthesis Kit is supplied with a specially formulated 5X buffer. This buffer has been optimized to work with the RT provided in the kit and contains a ratio of anchored oligo (dT) primers and random hexamers determined to produce a nonbiased population of cDNA. High quality cDNA can be produced with maximum efficiency using this buffer and RT. The use of other additives is not required or recommended.

Template

PR1MA qMax RT generates very high cDNA yields from small amounts of starting material. The cDNA Synthesis Kit works optimally with 0.5 μ g to 4.0 μ g of total RNA or oligo(dT) purified mRNA.

Reaction conditions

Most reactions can be carried out at a temperature of 42°C for 30 minutes. When working with templates that have a high GC content (above 65%), the temperature can be increased, up to 55°C, to alleviate any problems associated with secondary structure.

Analysis by qPCR

A cDNA synthesis reaction produces enough cDNA for analysis by real time PCR as well as other down stream processing reactions. The cDNA may be diluted 10X in PCR grade water prior to qPCR, however, the optimum dilution should be determined based on target gene abundance. For a 20µl qPCR reaction, using 2.0 to 5.0µl of the cDNA solution is recommended. cDNA may be stored at 4°C for 1 week or -20°C longer term.

Technical Support

For trouble-shooting and tech support, contact us at tech@midsci.com or call 800 227-9997.

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Reaction Setup

Thaw the 5X buffer and prepare your reaction in a 0.2ml tube as below:

Component	20 μl reaction	Final co	ncentration/Notes
5X cDNA Synthesis Reaction Buffe	r 4 μl	1X	
20X qMax Reverse Transcriptase	1μΙ	1X	Add before total RNA
Total RNA, 4pg - 0.5μg	Variable	Add up to 2	.0μg for low copy gene
PCR grade water	to final reaction volun	ne	

For other volumes, adjust the amount of each component accordingly.

Protocol

- 1. Incubate tube from above at 42°C for 30 minutes. For RNA with a high degree of secondary structure, incubate at 55°C.
- 2. Denature the RT by incubating at 85°C for 10 minutes.

The resulting cDNA can be diluted 10x in PCR-grade water prior to qPCR, however the optimum dilution should be determined based on target gene abundance. We recommend $2\mu l$ to $5\mu l$ of the cDNA solution per $20\mu l$ real-time PCR reaction. Alternatively, cDNA may be stored at 4°C for 1 week or -20°C for long term storage.

MidSci offers a full line of PCR enzymes and master mixes. Visit www.midsci.com for details.

Package contents and reordering

The PR1MA qMax cDNA Synthesis Kit is available in 25, 100 and 250 reaction packs. Kit includes 20X reverse transcriptase with RNase inhibitor and 5X reaction buffer with primers.

PR1MA qMax cDNA Synthesis Kit, sample pack, 5 reactions

Catalog number PR2100-C-S Includes 5µl of 20X RT and 20µl 5X buffer.

PR1MA qMax cDNA Synthesis kit, 25 rxns Catalog number PR2100-C-25 Includes 25μl of 20X RT and 100μl 5X buffer.

PR1MA qMax cDNA Synthesis kit, 100 rxns Catalog number PR2100-C-100 Includes 100µl of 20X RT in 25µl aliquots and 400µl 5X buffer in 100µl aliquots.

PR1MA qMax cDNA Synthesis kit, 250 rxns Catalog number PR2100-C-250 Includes 250μl of 20X RT in 25μl aliquots and 1ml 5X buffer in 100μl aliquots.





qMax cDNA Synthesis Kit PR2100-C

Reverse Transcriptase and Buffer Package contains: 25µl of 20X Reverse Transcriptase and 100µl of 5X cDNA Synthesis Buffer 25 reactions, Based on 20µl total reaction volume Store at -20°C upon receipt

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