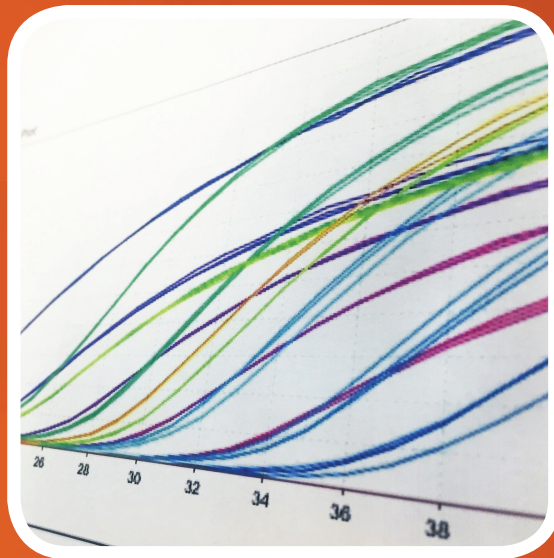


ISO
13485: 2003
13485: 2012

Real-time Tracking of Gene Expression



Next Generation Quantitative PCR

Real-Time PCR or quantitative PCR (qPCR) is a PCR-based technique that is able to simultaneously amplify and detect changes in the amplicon concentration. It is mostly used for quantitative analysis of gene expression in the research setting, but with more and more use in diagnostics and forensic testing nowadays. qRT-PCR, the coupling of a reverse transcription step in front of qPCR, allows quantification of RNA samples and detection of certain gene expressions, which can assist in diagnosing genetic diseases and detecting cancers.

Real-Time PCR or quantitative PCR (qPCR) is a PCR-based technique that is able to simultaneously amplify and detect changes in the amplicon concentration. Real-time PCR collects data during PCR amplification by utilizing fluorescence signal emitted by either DNA binding dyes such as SYBR Green Dye, or special probes such as TaqMan probe. **abm's** selection of BrightGreen qPCR MasterMixes and TaqProbe qPCR MasterMixes provides various options for both methods which also accommodates most qPCR thermal instrument models available on the market.

BrightGreen qPCR MasterMixes

abm's BrightGreen 2X qPCR MasterMix provides all ingredients necessary for quantitative PCR in a premixed and optimized format. A new generation of SYBR Green I, the BrightGreen Dye used in our BrightGreen qPCR MasterMixes works in a similar mechanism (Figure 1). Available with either ROX or fluorescein as the internal passive reference dye, **abm's** BrightGreen 2X qPCR MasterMix offers unparalleled performance in sensitivity, signal-to-noise ratio, and complete elimination of primer dimers.

Features:

- Streamlined protocol in a simple reaction set-up
- Accurate quantification of a variety of gene targets
- Reduced risk of contamination with less pipetting steps
- Compatibility with most real-time PCR instruments

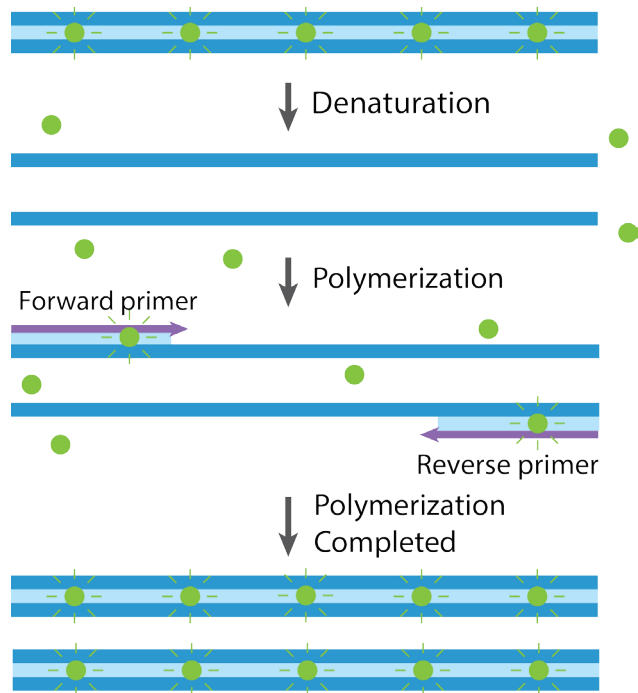


Figure 1: BrightGreen method: When the DNA polymerase synthesizes the complementary strand to the template, the intercalating dye, BrightGreen, incorporates itself into the dsDNA and emits a strong fluorescent signal which can be detected by the thermal cycler.

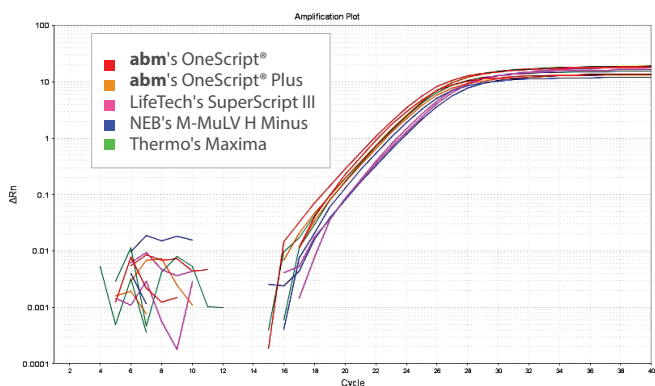


Figure 2A: With **abm's** BrightGreen qPCR MasterMix

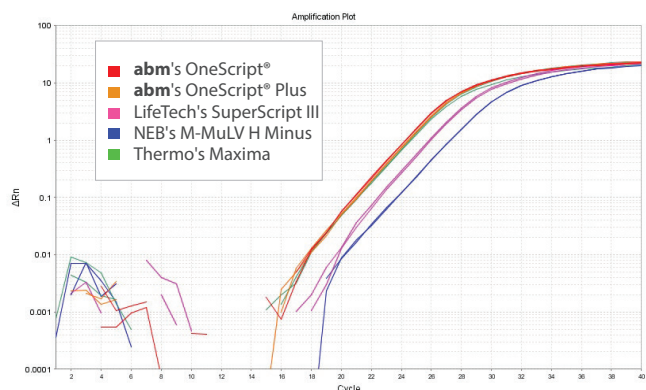


Figure 2B: With ABI Power SYBR Green Master Mix

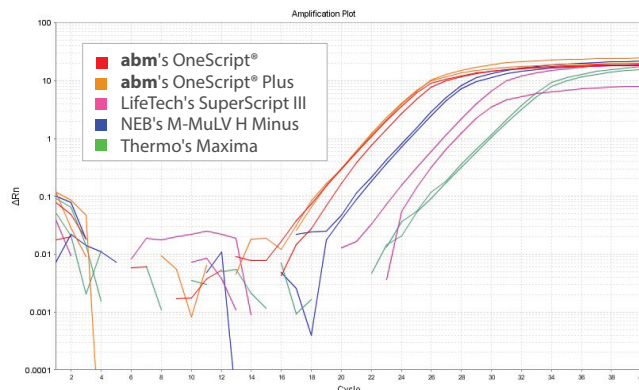


Figure 2C: With Biorad iQ SYBR Green Supermix

Reverse Transcriptase (RTase) is a well-known potent inhibitor of PCR. **abm's** BrightGreen 2X qPCR MasterMix outperforms competitors with its greater range of compatibility to commercially available RTases. Compared to other leading brands, our OneScript® and OneScript® Plus RTases also demonstrate the least amount of inhibition to downstream qPCR applications (Figure 2A-E).

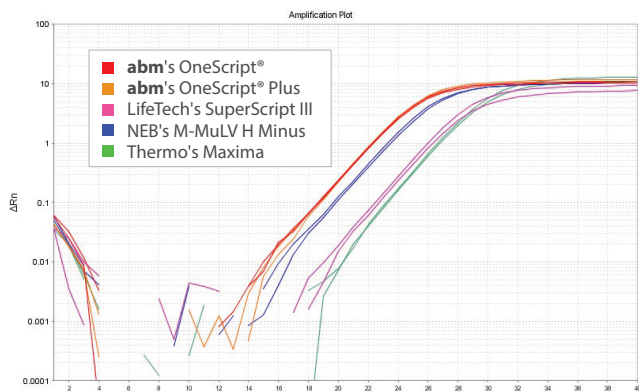


Figure 2D: With Takara SYPR Premix Ex Taq II

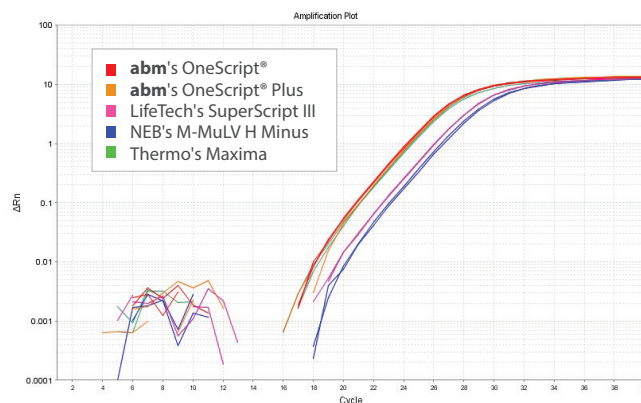


Figure 2E: With Roche FastStart SYBR Green Master

Figure 2 (A-E): 20 μ l RT reactions (100 pg of Human RNA per reaction) were performed with various RTases available on the market. 1 μ l of each RT product was then used directly as template for GAPDH amplification using qPCR MasterMixes from various leading brands.

abm's BrightGreen 2X MasterMix is the ultimate solution for reducing non-specific amplification in qPCR. Its optimized buffer system works in tandem with a highly efficient HotStart DNA polymerase to minimize signal-to-noise ratio (Figure 3A-E).

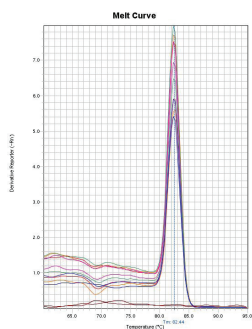


Figure 3A: abm's BrightGreen qPCR MasterMix

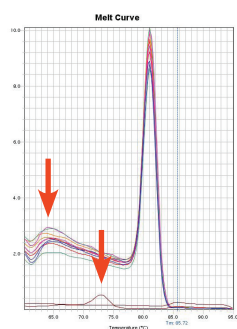


Figure 3B: ABI Power SYBR Green MasterMix

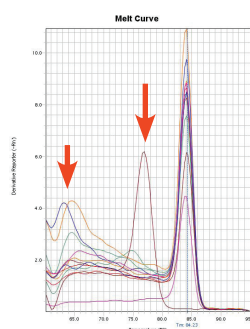


Figure 3C: Biorad iQ SYBR Green SuperMix

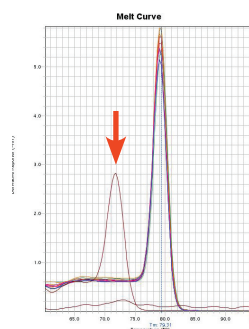


Figure 3D: Takara SYPR Premix Ex Taq II

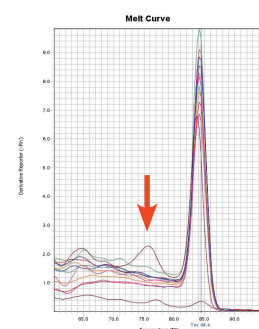


Figure 3E: Roche FastStart SYBR Green Master

Figure 3 (A-E): Melt curves of the corresponding MasterMixes from the reactions shown in Figure 3A-3E.

TaqProbe qPCR MasterMixes

abm's TaqProbe 2X qPCR MasterMix is designed for high throughput quantitative PCR using TaqMan[®] probe-based chemistry (Figure 4). Available with either ROX or fluorescein as the internal passive reference dye, **abm's** TaqProbe 2X qPCR MasterMix offers superb performance in sensitivity and signal-to-noise ratio. The multiplex formulation supports quantitative amplification and detection of up to four targets simultaneously with consistent and reliable results.

Features:

- Streamlined protocols with its single tube set-up
- Accurate quantification of a variety of gene targets
- Reduced contamination risks with less pipetting steps
- Compatibility with most real-time PCR instruments

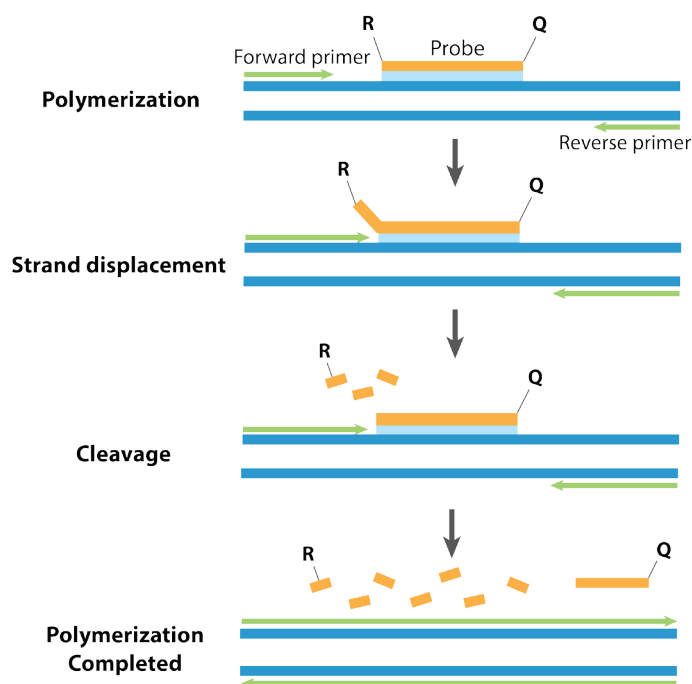


Figure 4: TaqProbe method: 5' to 3' exonuclease activity of Taq DNA polymerase displaces the reporter dye of the TaqMan Probe, and cleaves the reporter at the 5' end from the quencher at the 3' end to emit fluorescence.

One-Step BrightGreen/TaqProbe qRT-PCR Kit

One-Step qRT-PCR Kit uses a combination of high-quality enzymes in a proprietary buffer system to deliver precise and accurate sample analysis in a high-throughput format. This kit offers ultimate convenience to the end-user in addition to guaranteed performance with respect to high sensitivity, superb signal-to-noise ratio, and complete elimination of primer dimers. **abm** provides the One-Step qRT-PCR Kit with a selection of reference dyes compatible with a range of qPCR machines.

- Improved fidelity and yield for reverse transcription
- Prevention of template (RNA) degradation with RNaseOFF Ribonuclease Inhibitor
- Superb performance with respect to sensitivity and signal-to-noise ratio
- Significant reduction in non-specific PCR amplification by utilizing HotStart Taq DNA polymerase
- Fully optimized for detection of low-copy genes — high-quality, full-length cDNA from as little as 0.01pg of RNA
- Streamlined protocol in a simple single-tube reaction set-up
- Reduced risk of contamination with less pipetting steps

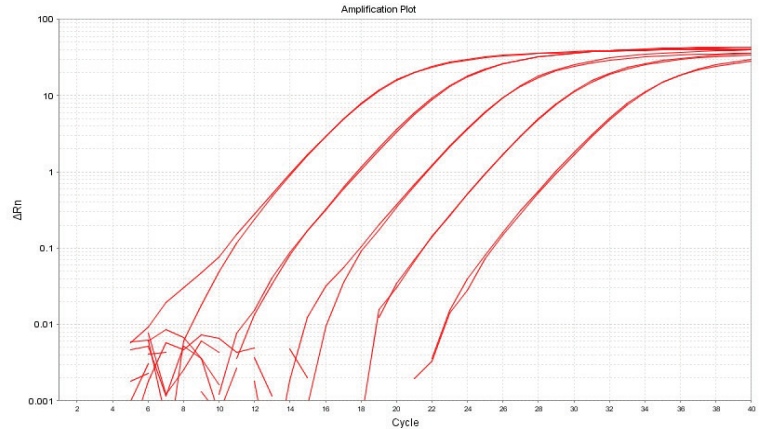


Figure 5: 20 μ l One-Step qRT-PCR reactions (20 ng, 2 ng, 200 pg, 20 pg or 2 pg of RNA per reaction) were carried out with **abm**'s One-Step BrightGreen qRT-PCR kit for GAPDH amplification.

ExCellenCT One-Step BrightGreen/TaqProbe qRT-PCR Kit

abm's ExCellenCT Lysis Kit provides an unique and effective alternative method to extract and process RNA templates directly from cultured cells in less than 15 minutes. **abm**'s proprietary qRT-PCR Enzyme Mix contains stabilizers and enhancers to optimize the two reactions in a real-time "single step". Coupled together, this complete system provides the ultimate convenience in generating consistent, reproducible, and accurate results from 10^1 - 10^5 of cultured cells.

BrightGreen Express qPCR MasterMixes

abm's BrightGreen Express 2X qPCR MasterMix provides all ingredients necessary for extremely rapid quantitative PCR in a premixed and optimized format. Our proprietary TaqFast DNA Polymerase shortens reaction time to 1/5 of standard qPCR time. Available with either ROX or fluorescein as the internal passive reference dye, **abm**'s BrightGreen Express 2X qPCR MasterMix offers unparalleled sensitivity, signal-to-noise ratio, and complete elimination of primer dimers.

- Extremely rapid activation — only 30 seconds!
- Ultra-fast qPCR thermal cycling — 10 second annealing/elongation time
- Streamlined protocol with a simple reaction set-up
- Ultra-fast quantification of a variety of gene targets
- Reduced risk of contamination with less pipetting steps
- Compatibility with most real-time PCR instruments

KiloGreen qPCR MasterMixes

abm's KiloGreen 2X qPCR MasterMix is designed for amplification of complex DNA fragments (i.e. high GC content and secondary structure) ranging from 300bp to 1.3kb in length. Available with either ROX or fluorescein as the internal passive reference dye, **abm**'s KiloGreen 2X qPCR MasterMix enables reliable amplification as well as unmatched sensitivity and signal-to-noise ratio.

- Streamlined protocol in a simple reaction set-up
- Accurate quantification of a variety of gene targets
- Reduced risk of contamination with less pipetting steps
- Compatibility with most real-time PCR instruments

BrightGreen miRNA qPCR MasterMixes

Designed for quantitative real-time analysis of miRNA samples, **abm's** BrightGreen miRNA qPCR MasterMix offers superb performance in sensitivity, signal-to-noise ratio, and reduction of non-specific PCR amplification. The proprietary formulation of our MasterMix is optimized for the amplification of cDNA generated from **abm's** miRNA cDNA synthesis kits. Available with either ROX or fluorescein as the internal passive reference dye, BrightGreen miRNA qPCR MasterMixes are compatible with a range of qPCR machines.

qPCR Lentivirus Titration Kit

Designed with titer primers for all HIV-1 based vector detection, **abm's** Lentivirus Titration Kit enables fast and simple lentivirus titration. Its rapid RNA extraction method and qRT-PCR based viral RNA quantification allow for assay completion in just 2 hours. Along with our online titer calculator, titration of lentiviral preparations has never been easier.

- No RNA purification — saves time and eliminates inaccuracies
- Ready-to-use reagent mix — reduces variability
- qRT-PCR in one step — more sensitive and accurate than other methods
- No NTC amplification — the only one on the market that eliminates NTC signals due to our unique primer design

qPCR AAV Titration Kit

The qPCR Adeno-Associated Virus Titration Kit provides a fast and simple way to perform Adeno-Associated Virus titration. **abm's** kit has demonstrated itself to be superior in both sensitivity and specificity compared to similar products on the market, while keeping non-specific background signals to a minimum.

- No RNA purification — saves time and eliminates inaccuracies
- Ready-to-use reagent mix — reduces variability
- Rapid set-up, qPCR assay can be run in under 2 hours

abm's qPCR Product List

Cat. No.	Product Name	Size
MasterMix-R, LR, iC, S	BrightGreen 2X qPCR MasterMix	500 reactions
G471-R, LR, iC, S	One-Step BrightGreen qRT-PCR Kit	100 reactions
G917-R, LR, iC, S	ExCellenCT One-Step BrightGreen qRT-PCR Kit	100 reactions
MasterMix-P, PL, PC, PS	TaqProbe 2X qPCR MasterMix	500 reactions
G493-P, PL, PC, PS	One-Step TaqProbe qRT-PCR Kit	100 reactions
G918-P, PL, PC, PS	ExCellenCT One-Step TaqProbe qRT-PCR Kit	100 reactions
MasterMix-ER, EL, EC, ES	BrightGreen Express 2X qPCR MasterMix	500 reactions
MasterMix-KR, KL, KC, KS	KiloGreen 2X qPCR MasterMix	500 reactions
MasterMix-mR, mL, mC, mS	BrightGreen miRNA qPCR MasterMix	500 reactions
LV900	qPCR Lentivirus Titration Kit	100 reactions
G931	qPCR Adeno-Associated Virus Titration Kit	100 reactions

More Resources

For more information about Quantitative PCR, visit our Knowledge Base and YouTube Channel!

Knowledge Base

https://www.abmgood.com/marketing/knowledge_base.php

YouTube Channel

www.youtube.com/c/abmgood

Polymerase Chain Reaction - An Introduction

<https://youtu.be/matsiHSuoOw>

Polymerase Chain Reaction - Variations of DNA Polymerase

<https://youtu.be/oqeV72oYfD0>

qPCR Products Overview

<https://youtu.be/nSEgjidPUFo>

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All **abm** PCR, RT-PCR, and qPCR products are ISO 13485:2003 and 13485:2012 certified as diagnostic grade and in compliance with all regulatory requirements for the design and manufacture of medical devices, as outlined by the International Organization for Standardization (ISO). For technical questions, please email us at technical@abmgood.com or visit our website at www.abmGood.com.

