Affymetrix® | USB® Products

PCR Tools

USB® VeriQuest™ Fast SYBR® Green qPCR Master Mix with Fluorescein (2X)

Applications:

- Amplification from genomic DNA or cDNA input
- Gene expression validation

Features:

- Uses fast mode thermal cycling conditions for results in one-third of the time compared to standard run protocols
- One-tube master mix with Fluorescein Reference Dye included—just add template, primers and water
- Reproducibility and consistency over a broad dynamic range: 8 orders of magnitude linear detection range (Fig. 1)
- High specificity with exceptional performance on challenging GC-rich regions (Fig. 2)
- Sensitivity and precision with limited targets (Fig. 3)
- Stable at room temperature for 72 hours in a pre-assembled reaction (Fig. 4)
- Contains dUTP and Uracil-DNA Glycosylase (UDG or UNG) for carry-over contamination prevention

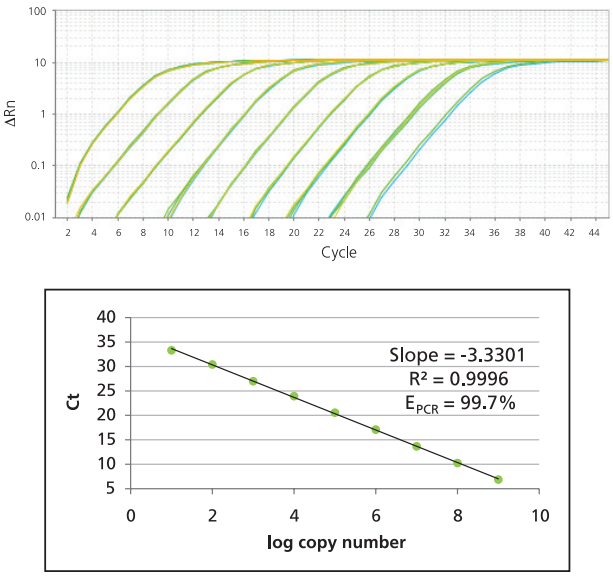
USB VeriQuest Fast SYBR Green qPCR Master Mix with Fluorescein is optimized for SYBR Green detection on all instruments that utilize Fluorescein as a passive reference dye. This master mix can be run using Fast mode cycling protocols, providing results in one-third of the time when compared to standard real-time PCR reagents. The 2X ready-to-use master mix contains hot start VeriQuest Taq DNA Polymerase, $MgCl_2$, ultrapure nucleotides with an optimized dUTP:dTTP ratio, Uracil-DNA Glycosylase (UDG or UNG), SYBR Green, and Fluorescein Passive Reference Dye in a proprietary reaction buffer. The hot start Taq polymerase has no polymerase activity prior to the initial heat activation step which allows reaction assembly at room temperature. The mix offers higher specificity and sensitivity, virtually eliminating non-specific primer amplification which can negatively affect the efficiency and accuracy of the data (Fig. 2). Since the mix contains dUTP and UDG, carry-over contamination prevention can be performed prior to amplification. VeriQuest Fast SYBR Green qPCR Master Mix with Fluorescein offers the same sensitivity and specificity found with our standard mode mixes with results in a fraction of the time.

VeriQuest Fast SYBR Green qPCR Master Mix with Fluorescein shows exceptional efficiency and high specificity on challenging templates such as high GC- and AT-rich regions (Fig. 2). The optimized formulation ensures no sacrifice in quality for increased speed. Performance comparisons highlight the sensitivity and reliable performance of this platform independent solution.

The sensitivity of the master mix allows for discrimination from a 1.33-fold difference in gene target amount detected. In Figure 2 a 1.33 to 10-fold dilution series of 10 ng to 1 ng of cDNA reverse-transcribed from HeLa total RNA were amplified with an efficiency of >99%.

VeriQuest Fast SYBR Green qPCR Master Mix with Fluorescein is highly stable and easy to work with. It remains stable at room temperature for 72 hours in a pre-assembled reaction and can be stored at 4°C for convenient handling. The mix also allows for room temperature reaction set-up. The speed and stability make VeriQuest Fast SYBR Green qPCR Master Mix with Fluorescein ideal for high-throughput handling.

Fig. 1. Linear detection range



Linear detection range of VeriQuest Fast SYBR Green qPCR Master Mix. Amplification plot and standard curve from real-time PCR for a dilution series of a synthetic target with starting amounts of 10^9 copies amplified in four replicate reactions using the ABI 7500 Real-Time PCR System and GAPDH primers. Reaction performed using fast mode cycling with 5 minutes activation at 95°C. The amplification process was linear over eight orders of magnitude.

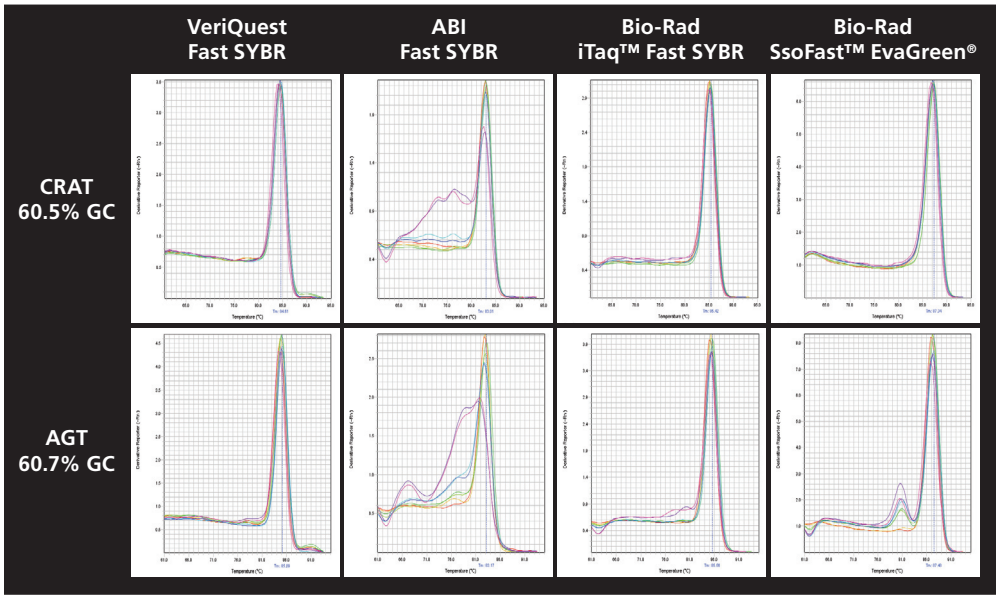
Components:

75675	100 reactions	1 ml
	500 reactions	5 ml
	1,000 reactions	2 x 5 ml
	2,500 reactions	5 x 5 ml
	5,000 reactions	10 x 5 ml

VeriQuest Fast SYBR Green qPCR Master Mix with Fluorescein (2X)

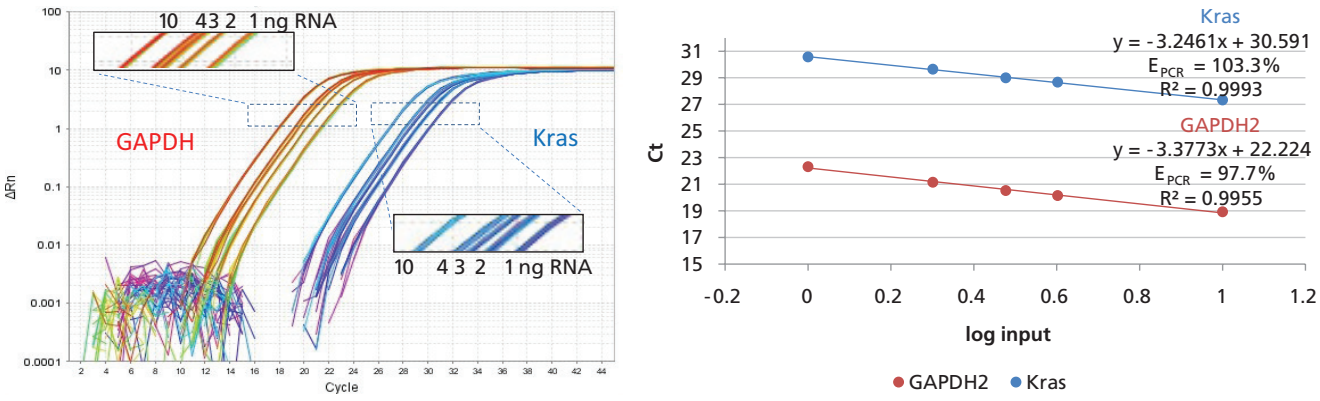
Product Code	Pack Size
75675	100 reactions
	500 reactions
	1,000 reactions
	2,500 reactions
	5,000 reactions

Fig. 2. Specificity with amplification of GC rich targets



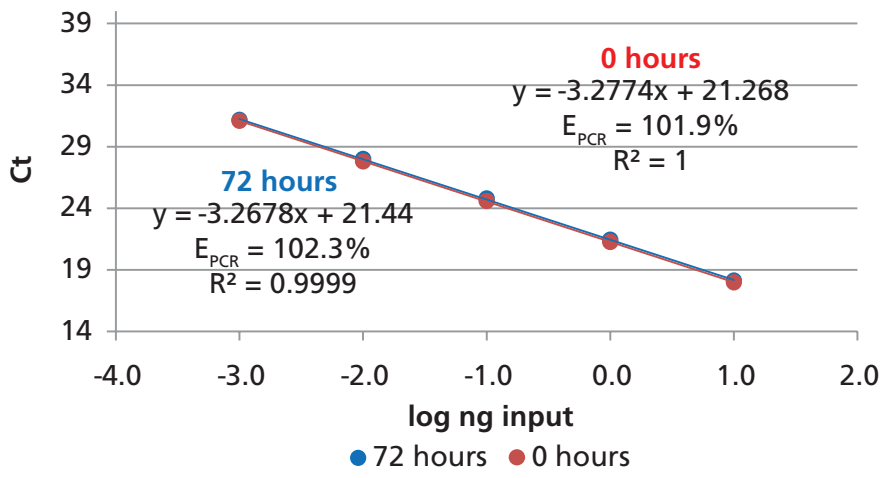
Melt curve analysis showing amplification results of CRAT, Carnitine O-Acetyltransferase, and AGT, Angiotensinogen. Results show comparisons against competitor master mixes.

Fig. 3. High sensitivity and precision in limited target quantification



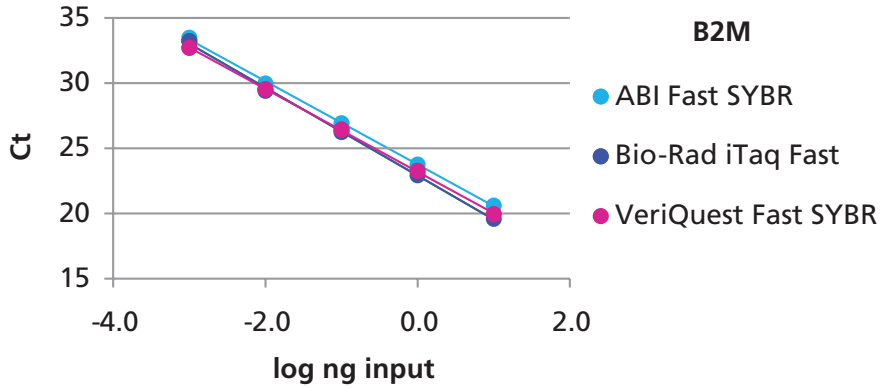
Amplification plot (left) and standard curve (right) from real-time PCR for a 1.33 to 10-fold dilution series of 10 ng to 1 ng of cDNAs reverse-transcribed from HeLa total RNA.

Fig. 4. VeriQuest Fast SYBR Green qPCR Master Mix stability

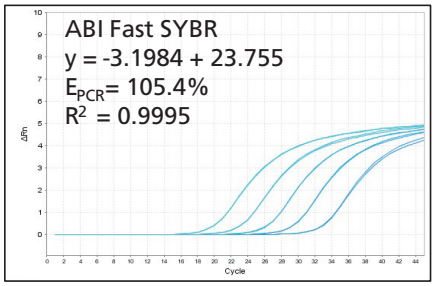


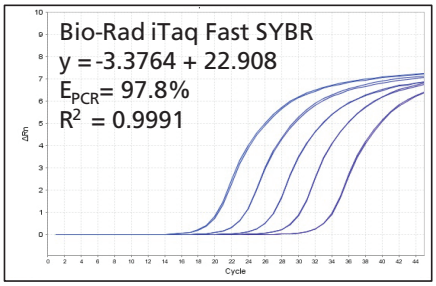
GAPDH was detected from pre-assembled PCR reactions incubated at room temperature for 72 hours. Results were compared to the freshly prepared mix (0 hours).

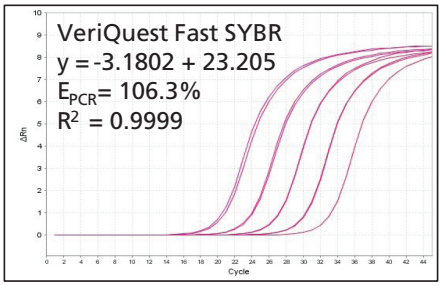
Fig. 5. Exceptional results with performance comparison



Standard curves from real-time PCR for a 10-fold dilution series of 10 ng to 1 pg cDNA reverse-transcribed from HeLa total RNA amplified in duplicate reactions with VeriQuest Fast SYBR Green qPCR Master Mix, Bio-Rad iTaq Fast SYBR Green Supermix with ROX™, and ABI Fast SYBR Green Master Mix.







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