

ThermaGo™

Enhanced Taq DNA Polymerase Specificity Throughout PCR

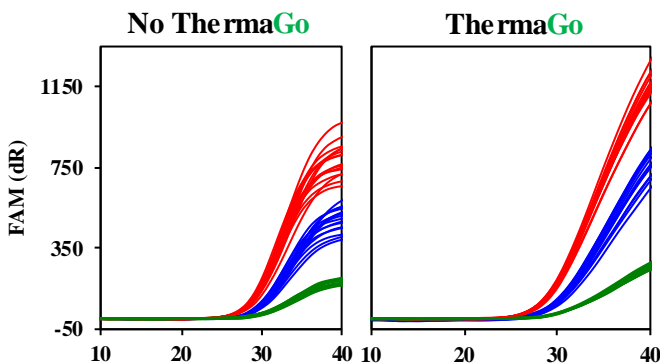


Figure 1 – ThermaGo reduces scatter, increases product signals, and allows greater separation of signals from homozygous and heterozygous genomes during real-time or end-point analyses. Lines represent probe signals from a Taqman probe against allele A.

Red lines, homozygous AA target.
 Blue lines, heterozygote Aa target.
 Green lines, homozygous aa target.

ThermaGo™ is a double-stranded, chemically modified nucleic acid that suppresses mis-priming during the annealing and extension steps of PCR. It increases the reproducibility of both real-time and end-point analyses. ThermaGo enhances the specificity of Taq DNA polymerase.

ThermaGo Suppresses Mispriming During PCR:

1. Improves the reproducibility of technical and biological replicates.
2. Increases detection of low-copy number targets by suppressing non-specific products.
3. Reduces signal scatter in standard and digital PCR.
4. Reduces mis-priming and chimera production prior to next-generation sequencing.
5. Improves end-point genotyping.

Applications of Interest:

Mispriming protection in all forms during PCR.

Detection of low copy target numbers (single-cell PCR, digital PCR, rare target detection in liquid biopsies/tumor samples).

Enhancing accuracy of highly multiplexed-PCR for targeted next-generation sequencing and other post-PCR applications.

Monoplex and multiplex endpoint SNP genotyping.

How to Prepare for Use:

ThermaGo is shipped as a dry reagent in 500 or 2500 units. To prepare 5 Units/ μ l ThermaGo, add 100 μ l molecular-grade 10 mM Tris-Cl, pH 8.3 to 500 units dry reagent (500 μ l 10 mM Tris-Cl, pH 8.3 to 2500 units dry reagent), vortex 1-2 minutes, then centrifuge briefly. Allow tube to sit at room temperature for 15 minutes with occasional mixing to ensure reagent is completely dissolved.

Recommended Storage:

Store ThermaGo at 4°C or -20°C in the dark or in light protected tubes. If frozen, divide stock into small volume aliquots to avoid freezing and thawing more than 5 times.

How To Use:

One unit of ThermaGo is defined as the amount required for maximum improvements in specificity, yield, and reproducibility in amplification reactions containing 1 unit of Taq DNA polymerase in a volume of 25 μ l. To use, begin by adding an equal number of units of ThermaGo and Taq DNA polymerase to a PCR master mix. ThermaGo works synergistically with ThermaStop™ to provide improved PCR performance before, during, and after amplification.

Typical 25 μ l PCR

Reagent	Final Concentration	Volume
10X PCR Buffer	1X	2.5 μ l
2 μ M primers	0.2 μ M	2.5 μ l
5 U/ μ l Taq	0.05 U/ μ l	0.25 μ l
5 U/ μ l ThermaGo	0.05 U/ μ l	0.25 μ l
Template		x μ l
H ₂ O		QS 25 μ l

To further optimize, test ThermaGo in the range of 0.5 to \geq 2.0 Units per Unit of Taq DNA polymerase. Note: very high levels of ThermaGo can inhibit amplification; thereby reducing product yield.

Expected Results:

Reactions containing optimal levels of ThermaGo will exhibit greater quantitative reproducibility due to suppression of non-specific products. Use of ThermaGo will produce higher yields of the intended products (e.g. stronger probe signals, correct bands on gels).

Please contact ThermaGenix Technical Support if you have further questions.