AmpliTaq Gold® 360 DNA Polymerase

Insert PN 4398303 Rev. C

For Research Use Only. Not intended for any animal or human therapeutic or diagnostic use.

Product list

Part Number	Part	Storage Conditions
4398813	AmpliTaq Gold® 360 DNA Polymerase (100 U); AmpliTaq Gold® 360 Buffer, 10X (1 × 1.5 mL); 25 mM Magnesium Chloride (1 × 1.5 mL); 360 GC Enhancer (1 × 1.5 mL)	On receipt, store at -15 to -25 °C.
4398823	AmpliTaq Gold® 360 DNA Polymerase (250 U); AmpliTaq Gold® 360 Buffer, 10X (1 × 1.5 mL); 25 mM Magnesium Chloride (1 × 1.5 mL); 360 GC Enhancer (1 × 1.5 mL)	
4398833	AmpliTaq Gold® 360 DNA Polymerase (1000 U); AmpliTaq Gold® 360 Buffer, 10X (4 × 1.5 mL); 25 mM Magnesium Chloride (4 × 1.5 mL); 360 GC Enhancer (4 × 1.5 mL)	
4398892	AmpliTaq Gold® 360 DNA Polymerase (1500 U); AmpliTaq Gold® 360 Buffer, 10X (6 × 1.5 mL); 25 mM Magnesium Chloride (6 × 1.5 mL); 360 GC Enhancer (6 × 1.5 mL)	

Note: For safety and biohazard guidelines, refer to the "Safety" section in the *AmpliTaq Gold® 360 DNA Polymerase Protocol* (PN 4398943). For all chemicals, read the Safety Data Sheet (SDS) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

AmpliTaq® and AmpliTaq Gold® 360 DNA Polymerases ship at room temperature. Please note that if these enzymes are shipped with temperature sensitive products the entire shipment may be received on ice. Visit **http://www.appliedbiosystems.com/360wp** to review our stability studies and read more about the environmental impact of this change.

For detailed procedures to amplify DNA, refer to the *AmpliTaq Gold® 360 DNA Polymerase Protocol* (PN 4398943).

For detailed procedures to perform high resolution melting analysis, refer to the *Applied Biosystems High Resolution Melting Getting Started Guide* (PN 4393102). For brief procedures, refer to the product insert that accompanies the MeltDoctor™ HRM Dye.

AmpliTaq Gold® 360 DNA Polymerase

AmpliTaq Gold® 360 DNA Polymerase, when used with AmpliTaq Gold® 360 Buffer, 10X and the optional 360 GC Enhancer, amplifies a vast range of DNA sequence contexts. AmpliTaq Gold 360 DNA Polymerase is purified by an additional proprietary separation process to eliminate contaminating bacterial DNA sequences from the enzyme preparation. This ultra-pure enzyme, in addition to its hot-start capabilities, reduces false positives, amplifies low-level target sequences, and promotes amplification of a variety of templates including those from bacteria and the human genome. The enzyme is quality-control tested to verify that there are fewer than 10 copies of bacterial 16S ribosomal RNA gene sequences in a 5-unit aliquot.

Enzyme characteristics

- Storage Buffer Enzyme is supplied in 20 mM Tris, pH 9.0, 100 mM KCl, 0.1 mM EDTA, 1 mM DTT, 0.5% (w/v) Tween 20, 50% (v/v) glycerol
- Source: Recombinant, modified form of the *Thermus aquaticus* DNA polymerase gene expressed in *E. coli*
- Concentration: 5 U/µL

Reagent characteristics

The AmpliTaq Gold® 360 Buffer, 10X activates AmpliTaq Gold 360 DNA Polymerase resulting in highly specific, robust PCR amplification. The ionic strength and the pH of AmpliTaq Gold 360 Buffer, 10X are optimized for use with the AmpliTaq Gold 360 DNA Polymerase. The magnesium ion concentration that is required for optimal PCR amplification depends on the specific set of primers and template that are used. The 360 GC Enhancer is used for difficult templates, especially for templates with high GC content.

Recommended conditions to amplify DNA

Table 1 Recommended reaction component volumes to amplify DNA

Components	Volume for 50-µL reaction	Final concentration	Notes
AmpliTaq Gold® 360 Buffer, 10×	5 µL	1×	—
25 mM Magnesium Chloride	2 to 8 µL	1.0 to 4.0 mM	In most cases, 2.0 mM works well.
(Optional) 360 GC Enhancer	1 to 10 µL	N/A	For targets with 65 to 70% GC, start with 5 µL. For targets with >75% GC, start with 10 µL. In general, if increased specificity is required, add 1 to 2 µL per 50-µL reaction.
AmpliTaq Gold® 360 DNA Polymerase	0.25 µL	1.25 U/reaction	For some difficult-to-amplify targets, up to 5.0 U can be added.
dNTP Mix	4 µL	200 µM each	Applied Biosystems Part Number N8080260 contains a 10-mM solution of dNTP (2.5 mM each of dATP, dCTP, dGTP, and dTTP).
Primer 1 (10 µM)	1 to 5 µL	0.2 to 1.0 µM	Lowering the primer concentration reduces potential secondary products.
Primer 2 (10 µM)	1 to 5 µL	0.2 to 1.0 µM	Lowering the primer concentration reduces potential secondary products.
DNA	—	<1 µg/reaction	Typically use $>10^4$ copies
PCR-grade water	Variable	—	—
Total Volume	**50 µL**	**—**	**—**

The following thermal profile is recommended on a Veriti® Thermal Cycler or equivalent platform:

Table 2 Recommended thermal profile to amplify DNA

Stage	Step	Temperature (°C)	Time
Hold	Initial denaturation	95	10 min
Cycle (25 to 40 cycles)	Denature	95	30 sec
	Anneal	Primer T_m†	30 sec
	Extend	72	60 sec/kb
Hold	Final extension	72	7 min
Hold	Final Hold	4	∞

† Although any primer can be used with this product, Applied Biosystems recommends using primers with T_ms >55 °C. Use the Primer T_m calculator found on an Applied Biosystems thermal cycler, or go to **http://www.appliedbiosystems.com/support/techtools/calc**.

Note: For recommended conditions to perform high resolution melting analysis, refer to the product insert that accompanies the MeltDoctor™ HRM Dye.

Part Number 4398303 Rev. C 07/2010

applied biosystems by life technologies

Headquarters
5791 Van Allen Way | Carlsbad, CA 92008 USA
Phone 760.603.7200
www.appliedbiosystems.com

International Sales
For our office locations please call the division headquarters or refer to our Web site at
www.appliedbiosystems.com/about/offices.cfm