AmpliTaq® 360 DNA Polymerase

360° Coverage for a Full Range of Targets

AmpliTaq® 360 DNA Polymerase

Benefits

- Optimized for the broadest range of targets (<3 Kb)—from everyday to challenging
- Unmatched sensitivity and yield
- Market-leading GC Enhancer for robust amplification of GC-rich sequences
- Achieves the highest quality sequencing data

Introduction

The new AmpliTaq® 360 products provide you with a solution that is designed to amplify the entire 360 degree "circle" of amplicon space—from everyday to more challenging templates. AmpliTaq 360 DNA Polymerase is available as a stand-alone enzyme or with the improved AmpliTaq 360 Buffer Kit, which includes the new AmpliTaq 360 Buffer, $MgCl_2$, and a supplementary 360 GC Enhancer.

The new AmpliTaq 360 products have elevated performance compared with other Taq DNA polymerases, including the original industry-leading AmpliTaq DNA Polymerase—the most referenced brand of DNA polymerase in the world.

One Enzyme, Any Target

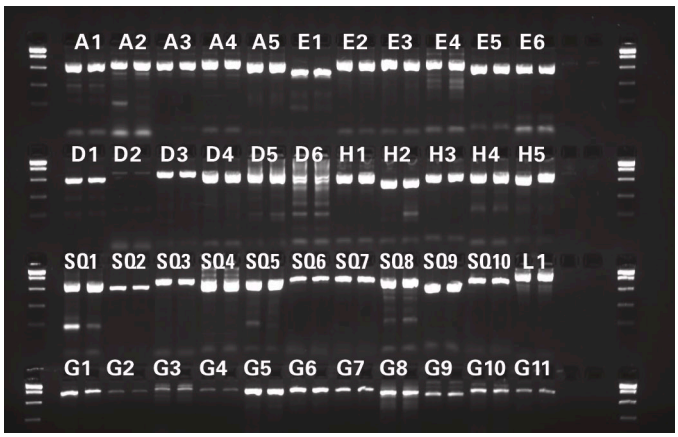
AmpliTaq 360 DNA Polymerase, when used with the new enhanced AmpliTaq 360 Buffer and the optional 360 GC Enhancer, amplifies a vast range of DNA sequence contexts.

AmpliTaq 360 DNA Polymerase has been extensively tested and optimized across a broad panel of difficult targets to achieve best-in-class performance for targets <3 Kb in length. Challenging targets include AT-rich, GC-rich, primer-dimer forming sequences, homopolymer repeats, and other targets that pose sequencing challenges. Targets that previously required significant optimization can now be amplified reproducibly with a single reagent under standardized conditions.

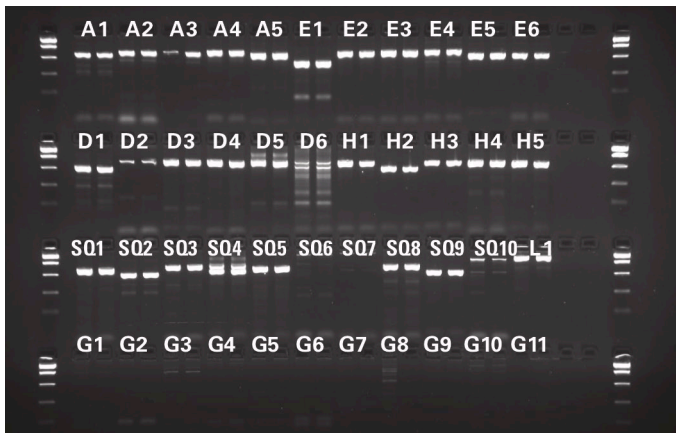
Competitive benchmarking across more than 40 target sequences distinguishes AmpliTaq® 360 DNA Polymerase as the best-performing enzyme, ensuring the highest probability of success for the amplification of both everyday and challenging targets. As shown in Figure 1, GC-rich regions are poorly amplified with other DNA polymerases while AmpliTaq 360 DNA Polymerase provides successful, robust amplification.

Superior Length and Sensitivity

AmpliTaq 360 DNA Polymerase reproducibly and efficiently amplifies long sequences (up to 3 Kb for hgDNA and 5 Kb for plasmid DNA). Figure 2 demonstrates high quality PCR amplification of long human and plasmid DNA. AmpliTaq 360 DNA Polymerase efficiently amplifies targets present at low copy number (Figure 3), even in the presence of high concentrations of complex DNA, making it especially suited for low-copy pathogen detection, and amplification of targets from degraded DNA samples. The extreme purity of the enzyme, along with its validated and optimized buffer system, contributes to its unmatched sensitivity.



Panel A. AmpliTaq® 360 DNA Polymerase



Panel B. Promega GoTaq® Green DNA Polymerase

Figure 1. AmpliTaq® 360 DNA Polymerase Amplifies a Broad Range of Targets. Panel A shows data produced with AmpliTaq® 360 DNA Polymerase while Panel B shows the same targets amplified using the Promega GoTaq® Green DNA Polymerase. PCR reactions were performed using 1 ng of template DNA and 1.25 units of enzyme in each 50 µL reaction. Annealing was uniform across the selected targets. Amplicons ranged from 300 to 1,400 base pairs (bp) in length with an average length of 553 bp. Each reaction was performed in duplicate. Amplicons are labeled as follows: E=Easy Amplification; A=High AT; G=High GC, D=Primer Dimer; H=Homopolymer; SQ=Sequencing Challenge. The high GC target (G) reactions included 2–10 µL of 360 GC Enhancer depending upon the target used.

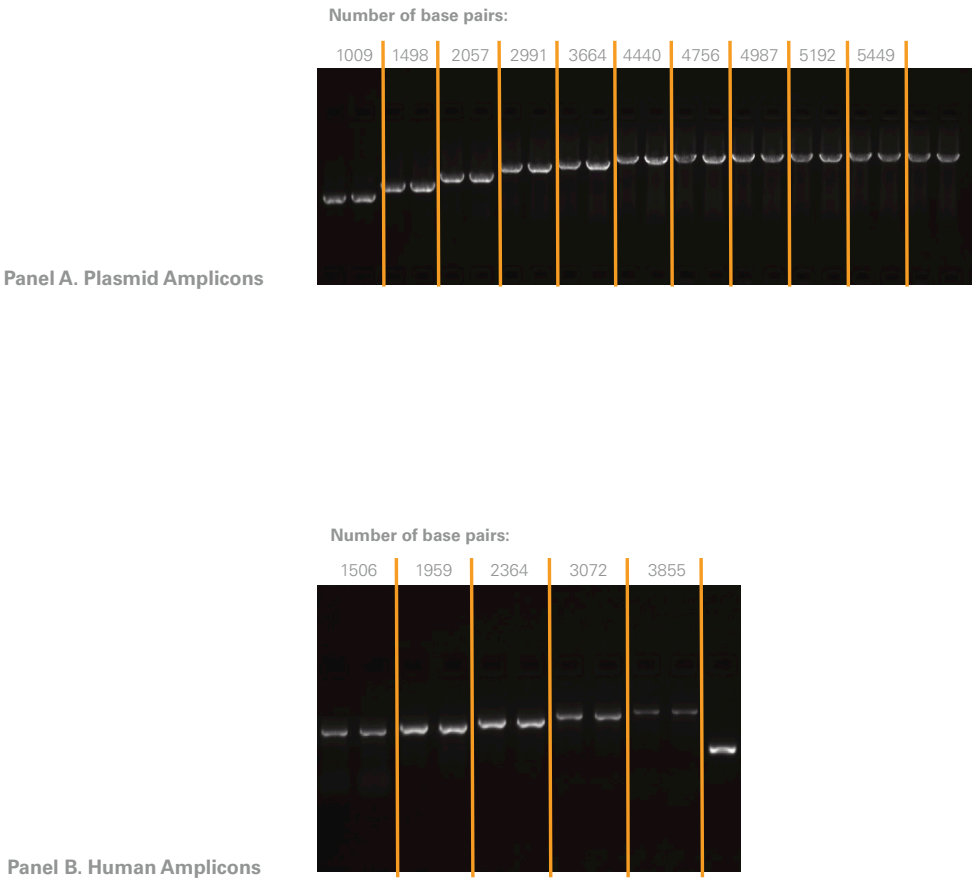


Figure 2. Amplification of Long Targets with AmpliTaq® 360 DNA Polymerase. Both plasmid (Panel A) and human (Panel B) targets, up to 5,459 bp long were efficiently and reproducibly amplified in duplicate reactions. Plasmid DNA was amplified using 2 mM $MgCl_2$ and standard thermal cycling conditions with a 6 minute extension time. Human Raji cell DNA was amplified using 2 mM $MgCl_2$ and standard thermal cycling conditions with a 4 minute extension time.

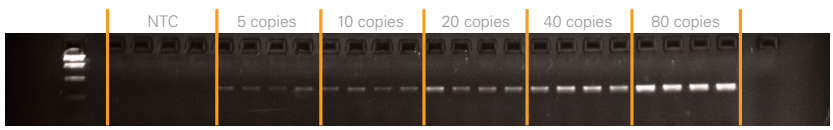


Figure 3. Sensitivity of AmpliTaq® 360 DNA Polymerase for Detection of Low Copy Targets. Amplification of the □-actin gene was performed for 0 to 80 starting copies of human genomic DNA, using 2.0 mM $MgCl_2$ and standard thermal cycling conditions with a 30 second extension time.

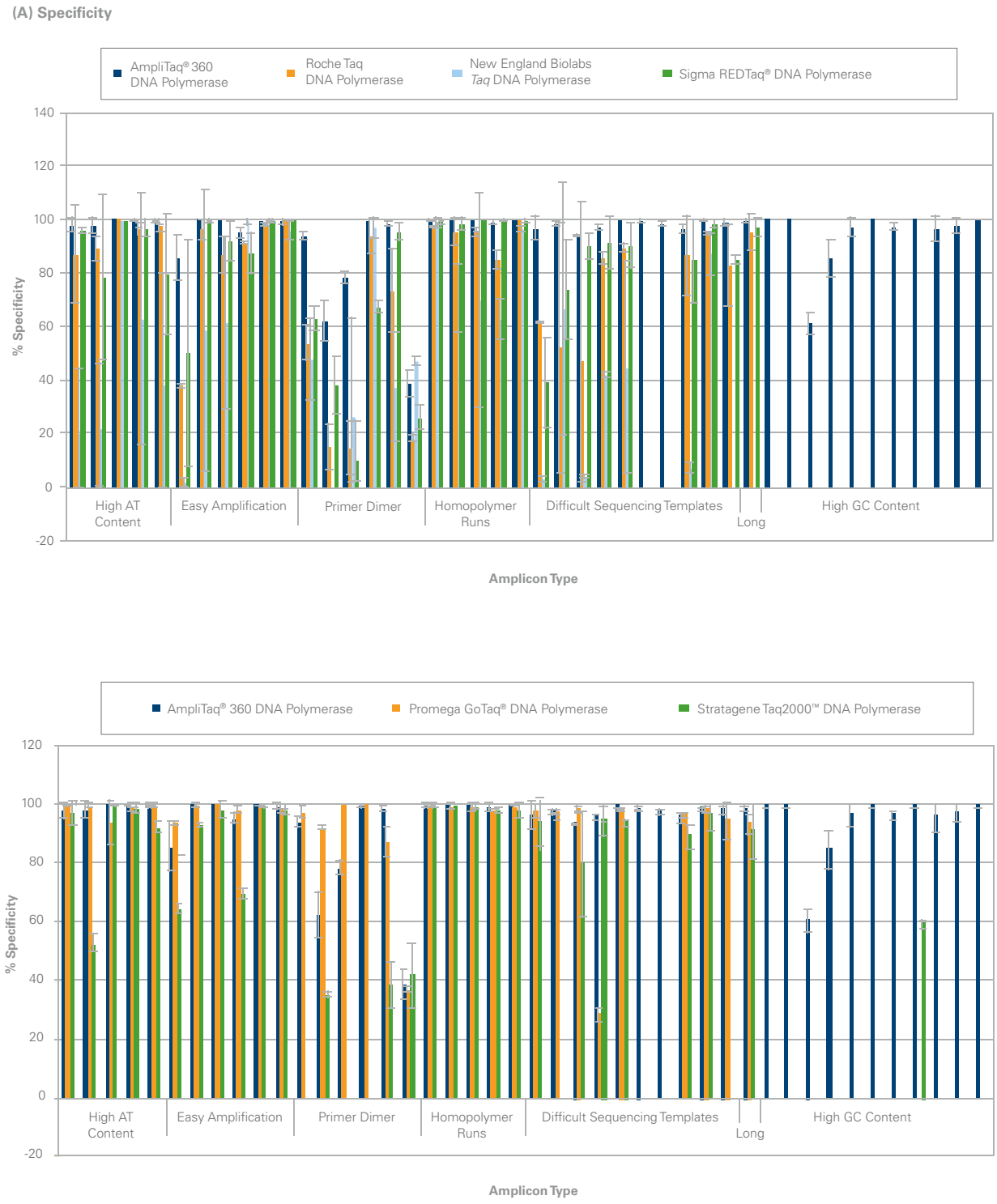
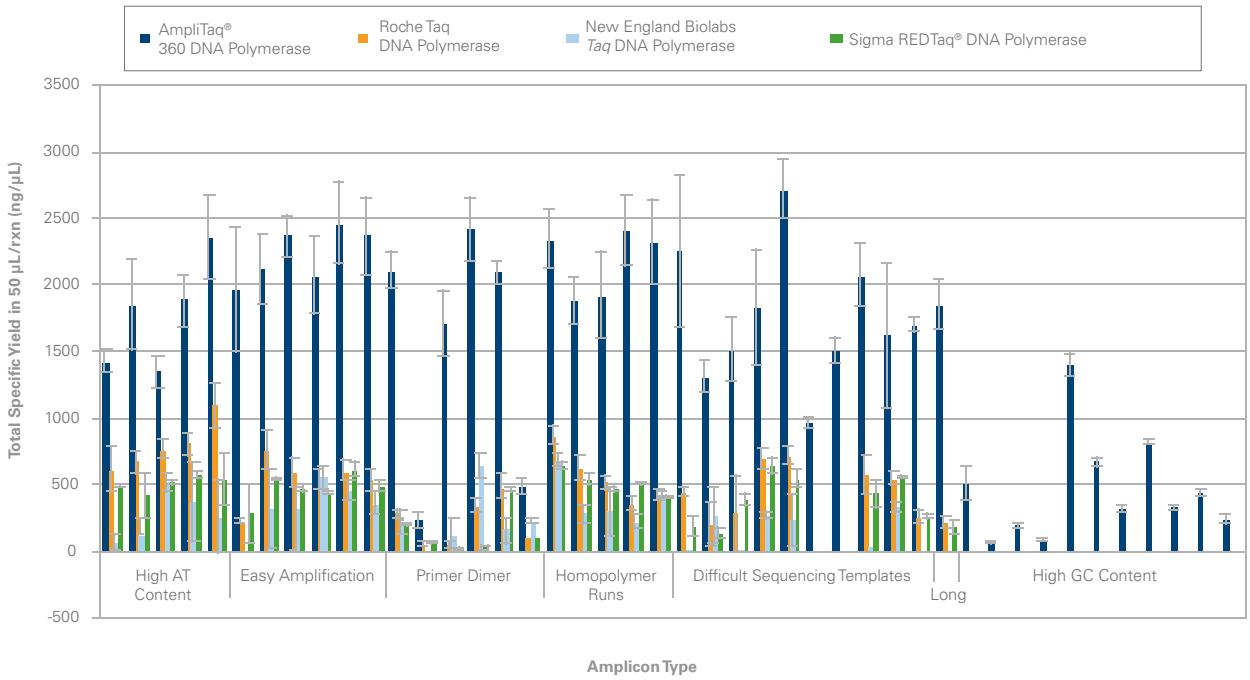
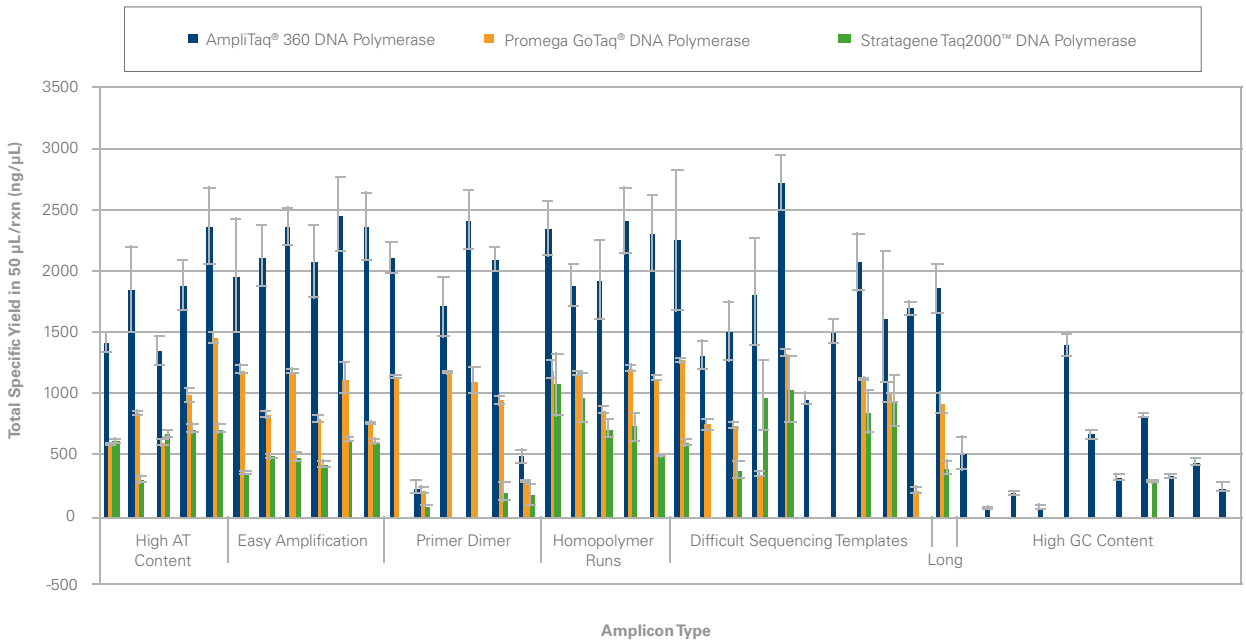


Figure 4. AmpliTaq® 360 DNA Polymerase Demonstrates Overall Superior Specificity and Yield. Summary of 2 replicates for each of 40 targets indicating the average specificity for each target and the standard deviation. PCR reactions were performed using 1 ng of template DNA and 1.25 units of enzyme in each 50 µL reaction. Annealing temperatures were uniform across the selected targets. Amplicons ranged from 300 to 1400 base pairs (bp) in length with an average length of 553 bp. Specificity and specific yield averages are summarized in Table 1.

(B) Specific Yield





Template Description	Specific Yield (ng)		Specificity (%)	
	AmpliTaq® 360 DNA Polymerase	Promega GoTaq®	AmpliTaq® 360 DNA Polymerase	Promega GoTaq®
Avg w/o GC-rich Amplicons	1912.48	943.10	94.23	93.44
Avg GC-rich Amplicons	658.87	14.85	95.20	6.82
Avg all Amplicons	1513.61	647.75	94.53	65.88

Table 1. Average Specific Yield and Specificity for AmpliTaq® 360 DNA Polymerase Compared with Promega GoTaq® Green DNA Polymerase. PCR reactions were performed using 1 ng of template DNA per reaction using cycling conditions according to each manufacturer's recommendations. All enzyme concentrations were standardized at 0.025U/µL. Annealing and extension times and temperatures were specific to each primer set. Amplicons ranged from 300 to 1400 base pairs (bp) in length with an average length of 553 bp. Promega GoTaq® Green DNA Polymerase was chosen for this comparison because it was the next best competitive product when compared with AmpliTaq® 360 DNA Polymerase.

Superior Sensitivity, Reproducibility and Yield

Compared to the original AmpliTaq® DNA Polymerase, AmpliTaq® 360 DNA Polymerase is purified by an additional proprietary separation process to eliminate contaminating bacterial DNA sequences from the enzyme preparation. When used with the enhanced AmpliTaq 360 Buffer, this ultra-pure enzyme reduces false positive results, amplifies low-level target sequences, and promotes the amplification of a variety of templates including those from bacterial and human genomes. This provides excellent specificity, reproducibility and yield across a broad range of targets (Figure 4).

Optimized Buffer and GC Enhancer

Applied Biosystems offers the best PCR buffer formulation combined with a superior GC-rich enhancer for the overall best performing product across the entire amplicon space. The 360 GC Enhancer can be added to your reactions with difficult templates, especially for templates with a high percentage of GC content. The bottom row of amplicons in Figure 1 demonstrates the successful amplification of GC-rich sequences that competitor enzymes are unable to amplify.

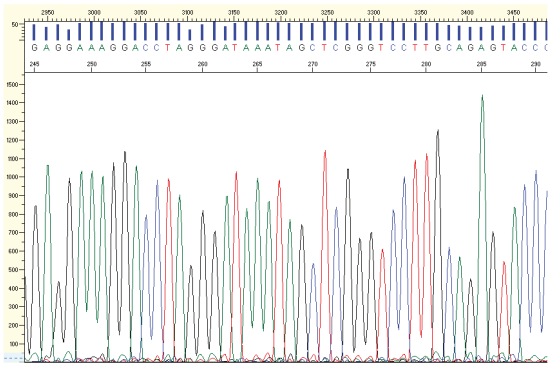
Obtain High Quality Sequencing Data

Achieve equivalent or better sequencing results compared to the original AmpliTaq DNA Polymerase. Quality parameters including read length, KB-QV30, and peak-under-peak were compared for sequencing data obtained from both AmpliTaq 360 DNA Polymerase and the original AmpliTaq DNA Polymerase (Figure 5). Equivalent, or in the case of high-GC containing sequences, better sequencing results were consistently obtained.

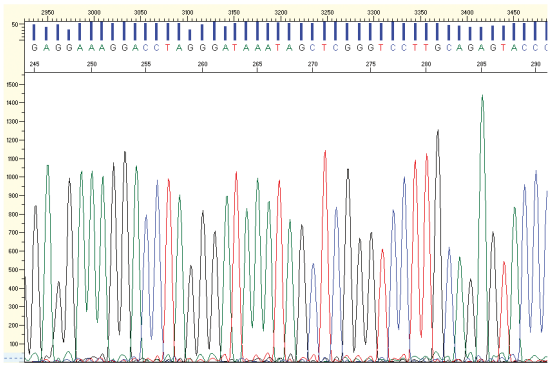
Unparalleled PCR Technical Support

Applied Biosystems has ongoing R&D programs that focus on refining the PCR process and developing new applications and complementary products. We offer technical seminars, PCR user-group meetings, and internet sites to keep you up-to-date on PCR protocols and new applications. In addition, our experienced staff of PCR specialists is always available to assist you. For a list of national and international technical support phone numbers, visit our Web site at **www.appliedbiosystems.com**.

Panel A.
Example of Sequencing Results from a Standard Template.



AmpliTaq® DNA Polymerase



AmpliTaq® 360 DNA Polymerase

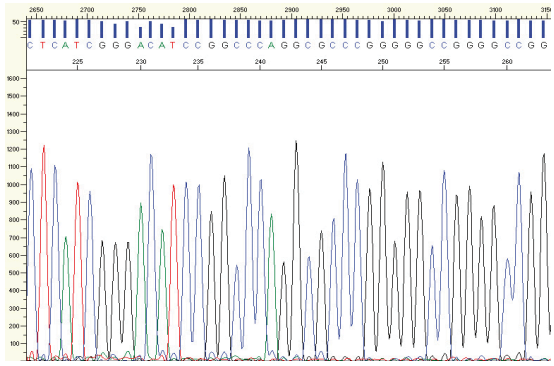
	Length of Read (bp)	KB-QV30	Peak-under-Peak
AmpliTaq® DNA Polymerase	553	543	0.0420
AmpliTaq® 360 DNA Polymerase	555	539	0.0507

Amplicon Length: 634 bp
GC Content: 57.4%

Panel B.
Example of Sequencing Results from a High-GC Content Template.

Does not amplify

AmpliTaq® DNA Polymerase



AmpliTaq® 360 DNA Polymerase

	Length of Read (bp)	KB-QV30	Peak-under-Peak
AmpliTaq® DNA Polymerase	Does not amplify	Does not amplify	Does not amplify
AmpliTaq® 360 DNA Polymerase	542	535	0.0510

Amplicon Length: 582 bp
GC Content: 78.2%

Figure 5. High Quality Overall Sequencing Data Obtained from AmpliTaq® 360 DNA Polymerase-Generated PCR Products. Amplification products from standard (Panel A) and high GC content (Panel B) targets, generated by AmpliTaq® 360 DNA Polymerase and the original AmpliTaq DNA Polymerase, were sequenced (10 ng PCR product per sequencing reaction). Samples were cycle-sequenced using BigDye® Terminator v3.1 and standard thermal cycling conditions. BigDye® XTerminator™ Purification Kit was used to clean up the cycle sequencing reaction, and the samples were injected into the Applied Biosystems 3730*xl* DNA Analyzer with POP-7™ polymer.

ORDERING INFORMATION

Description	Size	P/N
AmpliTaq® 360 DNA Polymerase *Includes AmpliTaq® 360 DNA Polymerase, AmpliTaq 360 Buffer, 25 mM $MgCl_2$ and 360 GC Enhancer*	100U	4398808
	250U	4398818
	1000U	4398828
	1500U	4398891
	2x1500U	4398893
	5x1000U	4398895
	25x1000U	4398897
	25000U	4398899
AmpliTaq® 360 DNA Polymerase, standalone	25,000U	4398838
AmpliTaq® 360 Buffer Kit *Includes AmpliTaq 360 Buffer, 25 mM $MgCl_2$ and 360 GC Enhancer*	1.5 mL	4398848
	6x1.5 mL	4398858
	150 mL	4398868
AmpliTaq® 360 DNA Polymerase Protocol	1 protocol	4398942
AmpliTaq® 360 DNA Polymerase Quick Reference Card	1 card	4398952

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Applied Biosystems

Headquarters
850 Lincoln Centre Drive | Foster City, CA 94404 USA
Phone 650.638.5800 | Toll Free 800.345.5224
www.appliedbiosystems.com

International Sales
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