ChemStart™ DNA Polymerase

Catalog No.	Pack Size and Concentration	Components and Volume	
AZ-1405	500 units, 5 U/µL	ChemStart™ Polymerase - 1 x 100 µL	5x ChemStart™ Buffer – 2 x 1 mL
AZ-1410	1000 units, 5 U/µL	ChemStart™ Polymerase - 2 x 100 µL	5x ChemStart™ Buffer – 4 x 1mL
AZ-1450	5000 units, 5 U/µL	ChemStart™ Polymerase - 10 x 100 µL	5x Chemstart™ Buffer – 4 x 5 mL

Description

ChemStart™ DNA Polymerase is a chemically modified form of *Taq* DNA Polymerase which requires a heat-activation step prior to PCR. Upon thermal activation, a polymerase modifier is permanently released providing for rapid enzyme activation. The resulting hot start PCR enzyme delivers unparalleled sensitivity, yield and specificity compared with conventional DNA polymerase technology. ChemStart™ DNA Polymerase is a high-performance chemically-mediated hot start preparation which specifically circumvents the need for *Taq* monoclonal antibodies and therefore reduces the possibility of biological contamination. In addition, a proprietary enzyme purification process removes bacterial DNA sequences making ChemStart™ an ideal choice for microbial screening, pathogen detection and other assays in which the reduction of false positives is essential.

- Robust PCR performance across a wide range of DNA templates including low-copy number viral and bacterial targets.
- Low residual host *E. coli* DNA
- High-yields with amplicons up to 5 kb.
- Efficient 5x ChemStart™ buffer formulation includes optimal levels of $MgCl_2$ for maximum PCR efficiency.
- 5x ChemStart™ DNA Polymerase and 5x Buffer are compatible with conventional PCR and probe-based qPCR.

Storage

ChemStart™ DNA Polymerase is shipped on blue ice and should be stored at –20°C upon receipt. Excessive freeze/thawing should be avoided.

Important Guidelines

5x ChemStart™ Buffer: The 5x reaction buffer contains 15 mM $MgCl_2$ and PCR enhancers for success with most amplicons including low-copy DNA targets.

Template: For complex genomic DNA, we suggest the use of 5 ng - 500 ng per reaction; For cDNA or plasmid DNA, please use < 100 ng per reaction.

Primers: Primers should have a predicted melting temperature of around 60°C, using default Primer 3 settings (http://frodo.wi.mit.edu/primer3/). The final primer concentration in the reaction should be between 0.2 µM and 0.6 µM.

Annealing: We recommend performing a temperature gradient to determine the optimal annealing temperature. Alternatively, 58°C can be used as a starting point. For optimization, increase in 2°C increments. For example, if non-specific products are present or smearing is visible, a higher annealing temperature is required.

Extension: Optimal extension is achieved at 72°C. The optimal extension time is dependent on amplicon length and complexity. 30 seconds per kilobase (kb) is recommended for amplification from eukaryotic genomic DNA or cDNA.

Reaction setup: Allow reagents to thaw and mix well by inversion. Centrifuge prior to use.

1. Prepare a PCR master mix based on following table:

Component	25 µL Reaction	Final Concentration/Notes
5x ChemStart™ Buffer	5 µL	1x
100 mM dNTPs (25mM each)	0.25 µL	1mM
Forward Primer (10 µM)	1.0 µL	400 nM
Reverse Primer (10 µM)	1.0 µL	400 nM
Template DNA	<100 ng cDNA, <500 ng genomic DNA	variable
ChemStart™ DNA Polymerase (5 U/µL)	0.25 µL – 0.50 µL	variable
PCR-grade water	Up to 25 µL final volume	

* For alternative total reaction volumes (eg. 20 µL), scale all components proportionally to maintain final concentrations.

2. PCR cycling:

Cycles	Temperature & Time	Notes
1	95°C, 10 minutes	Initial Denaturation, enzyme activation
25 - 40	95°C, 15 seconds 60°C, 15 seconds 72°C, 30 seconds per kb	Denaturation Annealing (determined by user) Extension

Quality Control

ChemStart™ DNA Polymerase is tested extensively for robust activity, processivity, efficiency, heat activation, sensitivity, absence of nuclease contamination and absence of nucleic acid contamination. ChemStart™ DNA Polymerase is manufactured under a comprehensive quality management system, following ISO 9001:2015 standards.

Limitations of Use

This product is intended for research purposes only and is not intended for any animal or human therapeutic use.

Technical Support

For trouble-shooting and technical guidance, please contact us at tech@azuragenomics.com and provide PCR reaction conditions, cycling parameters, amplicon size, and screen grabs (gel images) if possible.

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