# Accuris™ Hot Start Tag DNA Polymerase

### **Description**

Engineered for controlled polymerase activity, Accuris Hot Start Tag is bound with a monoclonal antibody that blocks enzyme activity. This allows reactions to be set up at room temperature without the risk of non-specific amplification. Improved yield, activity, sensitivity and speed are achieved by modifying the polymerase with the addition of hydrophilic residues. A proprietary 5X buffer system with PCR enhancers and optimal levels of MgCl, has been created to partner with Accuris Hot Start Tag DNA Polymerase and is supplied in this package. Little, if any, optimization is needed.

- -Performs across a wide range of DNA templates including genomic DNA and GC-rich and AT-rich sequences
- Proprietary 5X reacton buffer includes enhancers for maximizing enzyme activity and reaction speed
- -Improved solubility and template affinity

### Storage

Upon receipt, immediately store at -20°C. Avoid excessive freeze/thaw cycles. When stored as directed, this product will retain its activity for 12 months from date of receipt. The product may also be stored at 4°C for up to one month.

#### **Limitations of Use**

For research purposes only. Not intended for therapeutic or diagnostic use.

## **Quality Control**

Accuris enzymes and reagents are tested under general assay conditions for activity, reproducibility, efficiency, heat activation, sensitivity, and absence of nuclease contamination and nuclease activity. This product is manufactured under a comprehensive quality management system, following ISO 9001:2008 standards.

#### **General Guidelines**

#### 1. Reaction Buffer

The 5X reaction buffer supplied with the Accuris Tag DNA polymerase has been formulated for maximum efficiency, sensitivity and successful PCR with a variety of difficult templates. Proprietary PCR enhancers and 15mM MgCl<sub>3</sub> are included in the buffer. Use of additional PCR enhancers may have a negative effect on the reaction.

### 2. Template

For PCR of complex genomic DNA, 5ng -500ng of template DNA may be added per reaction. Do not add more than 100ng of DNA for cDNA or plasmid DNA

#### 3. Primers

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

### 4. Annealing Temperature

An initial annealing temperature of 55°C is recommended. If nonspecific products appear, increase the temperature in 2°C increments. Alternately, a temperature gradient may be performed.

#### 5. Extension

The polymerase performs optimally at 72°C. Extension time is dependent upon amplicon complexity and length. Generally, 30 seconds per kb is recommended for eukaryotic genomic DNA and cDNA. A five second extension is sufficient for shorter amplicons. An initial extension time of 90 seconds is recommended for multiplex reactions to maximize yield and specificity.

### **Technical Support**

For trouble-shooting and tech support, contact us at info@accuris-usa.com or call 908 769-5555.

Accuris is not responsible for consequential or incidental damages, whether direct or indirect, resulting from use of this product. Accuris quarantees the performance of this product as described when used in accordance with these slj 09/21 instructions.

#### Reaction setup

Prenare the reaction as follows:

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Component	25 μl reaction	50 μl reaction	Final concentration
Accuris 5x Hot Start Taq Reaction Buffer	5 μl	10 μΙ	1X
100mM dNTPs (25mM each)	0.25 μΙ	0.5 μΙ	1mM
Forward Primer (10µM)	1.0 μΙ	2.0 μΙ	400 nM
Reverse Primer (10μM)	1.0 μΙ	2.0 μΙ	400 nM
Template DNA	<100ng cDNA,	<500ng genomic	variable
Accuris Hot Start Taq (5u/μl)	0.1μl - 0.5 μl	0.20 µl - 1 µl	variable
PCR-grade water	to final rea	ction volume	

For other volumes, adjust the amount of each component accordingly.

Gently mix the solution. If needed, spin briefly in a microcentrifuge to bring reaction mixture to the bottom of the tube. Transfer samples to a thermal cycler and begin cycling. Be sure to include an initial activation/denaturation step of 1-2 minutes.

### Routine PCR Cycling

Step	Temperature	Time
Enzyme activation	95°C	1-2 minutes
	95°C	15 seconds
25-40 cycles	55°C to 67°C*	15 seconds
	72°C	30 seconds per Kb

<sup>\*</sup>Annealing temperature to be determined by user

### Package contents and reordering

Hot Start Tag Polymerase is supplied at a concentration of 5 units/µl. Available in 500, 1000 and 6000 unit packages with 5X buffer.

## Accuris Hot Start Tag, Sample Pack

Catalog number PR1000-HS-S Includes 10 µl of enzyme and buffer.

## Accuris Hot Start Tag 500 units

Catalog number PR1000-HS-500 Includes 100 µl of enzyme and 4ml of 5X buffer in 1 ml aliquots.

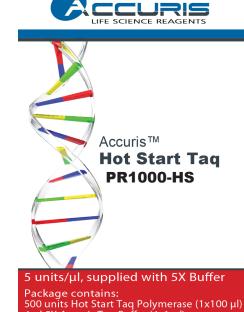
## Accuris Hot Start Tag, 1000 units

Catalog number PR1000-HS-1000 Includes 200 µl of enzyme in 100 µl aliquots, and 8ml of 5X buffer in 1ml aliquots.

### Accuris Hot Start Tag, 6000 units

Catalog number PR1000-HS-6000 Includes 1200 µl of enzyme in 100 µl aliguots, and 48ml of 5X buffer in1ml aliquots.

Visit www.accuris-usa.com for details on the entire PCR reagnet product line.



4ml 5X Accuris Tag Buffer (4x1ml) Store at -20°C upon receipt

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