

At Taq DNA Polymerase (Hot Start)



Lot :  
 Expiry Date :  
 Concentration : 5u/μl  
 Supplied with : 2ml of 10X ViBuffer A  
 1ml of 10X ViBuffer S  
 1ml of 50mM MgCl<sub>2</sub>

Store at - 20°C



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Description:

At Taq DNA Polymerase is a complex of specific anti-Taq monoclonal antibody with top quality thermostable Taq DNA Polymerase for automatic "hot start" amplification, resulting in greatly enhanced amplification specificity, sensitivity and yield. At Taq DNA Polymerase catalyzes the polymerization of nucleotides into duplex DNA in the 5' to 3' direction in the presence of Mg<sup>2+</sup> and has the 5' to 3' exonuclease activity.

Features:

- Ultra pure recombinant protein which is reversibly complex with anti-Taq monoclonal antibody that blocks replication activity of the enzyme at moderate temperatures.
- Carefully selected anti-Taq antibodies have high thermal stability, providing protection against non-specific primer extension from room temperature to 70°C.
- Formation of complexes between Taq DNA Polymerase and anti-Taq antibody forms a basis for automatic "Hot Start" amplification, which allows for the assembly of amplification reactions at room temperature.
- High stability of the complexes allows for the enormous increase in amplification specificity, sensitivity and yield in comparison to the conventional amplification assembly method.
- Increased specificity as a result of reduced amplification artefacts such as primer-dimer formation and mispriming in multiplex amplification.

Unit Definition :

1u is defined as amount of enzyme that required to catalyze the incorporation of 10nmoles of dNTP into acid-insoluble material in 30 minutes at 74°C.

Reaction Buffer:

10X ViBuffer A (without MgCl<sub>2</sub>):

500mM KCl, 100mM Tris-HCl (pH9.1 at 20°C) and 0.1% Triton™X-100. The buffer is optimized for use with 0.1-0.2mM of each dNTP.

10X ViBuffer S:

160mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 500mM Tris-HCl (pH 9.2 at 22°C), 17.5mM MgCl<sub>2</sub> and 0.1% Triton™X-100. The buffer is optimized for use with 0.35mM of each dNTP.

Storage Buffer:

20mM Tris-HCl (pH 8.0 at 22°C), 100mM KCl, 0.5% Tween™ 20, 0.5% Nonidet-P40, 0.1mM EDTA, 1mM DTT and 50% glycerol.

Quality Control:

All preparations are assayed for contaminating endonuclease, exonuclease, and non-specific DNase activities. Functionally tested in DNA amplification.

SUGGESTED INITIAL PCR CONDITIONS FOR VARIOUS PCR PRODUCT SIZES WITH VIVANTIS DNA POLYMERASES (#PL1201 - 06 / #PL2201 - 06 / #PL3201 - 06 / #PL4201 - 06)  
 REACTION MIX (FINAL CONCENTRATION) :

|   |                                |                          |           |            |
|---|--------------------------------|--------------------------|-----------|------------|
| Primers : 0.2 - 1μM<br><br>Template: Plasmid (0.02-0.2ng)<br>Lambda (0.1 - 150ng)<br>Genomic (0.05-5μg) | Product Size                   | 100bp - 5kb              | 5kb - 8kb | 8kb - 20kb |
|   | dNTP Mix                       | 100μM                    | 200μM     | 300μM      |
|   | ViBuffer (1X)                  | A                        | A         | S          |
|   | Ultrapurpure DMSO or formamide | -                        | 3%        | 3%         |
| DNA Polymerase  |                                | Refer to below Table (A) |           |            |

|                 |                 |                 |                 |
|-----------------|-----------------|-----------------|-----------------|
| Product Size    | 100bp - 5kb     | 5kb - 8kb       | 8kb - 20kb      |
| Denaturation    | 94°C, 2 min     | 94°C, 2 min     | 94°C, 2 min     |
| Denaturation    | 94°C, 30 s      | 94°C, 30 s      | 94°C, 12 s      |
| Annealing*      | 50 - 68°C, 30 s | 50 - 68°C, 30 s | 50 - 68°C, 30 s |
| Extension / 1kb | 72°C, 30 s      | 72°C, 45 s      | 68°C, 1 min     |
| Cycles          | 25 - 35         | 25 - 35         | 25 - 35         |
| Final Extension | 72°C, 7 min     | 72°C, 7 min     | 68°C, 7 min     |

\* Primer dependent

TABLE (A) : RECOMMENDED UNITS FOR SPECIFIC VIVANTIS DNA POLYMERASES PER 50μL REACTION VOLUME :

|              |                    |                        |                       |                          |
|--------------|--------------------|------------------------|-----------------------|--------------------------|
| Product Size | Taq (#PL1201 - 06) | Max Taq (#PL2201 - 06) | At Taq (#PL3201 - 06) | ATMax Taq (#PL4201 - 06) |
| 0.1 - 5.0kb  | 2.0                | 2.0                    | 2.0                   | 2.0                      |
| 5.0 - 8.0kb  | 2.5                | 2.0                    | 2.5                   | 2.0                      |
| 8.0 - 20.0kb | 2.5                | 2.0                    | 2.5                   | 2.0                      |
| >20.0kb      | -                  | 2.0                    | -                     | 2.0                      |

\* This protocol is subjected to changes depending on the template DNA