

FastGene® BAC-Free HotStart Taq

Technical Data Sheet

Product Description

The FastGene® BAC-free HS Taq DNA Polymerase is based on the single-subunit, wild-type Taq DNA polymerase of the thermophilic bacterium Thermus aquaticus. It is however not a bacterial recombinant protein but purified from an eukaryotic expression system.

In the HotStart formulation, the enzyme is chemically inactivated at temperatures below 75 °C. This prevents nonspecific amplification during reaction setup, increases sensitivity, and improves reaction efficiency. PCR products generated with FastGene® Taq HotStart are A-tailed and may be cloned into TA cloning vectors.

Product Applications

FastGene® BAC-free HS Taq Mixes are ideally suited for:

- High throughput PCR of bacterial genomes
- Amplification of low copy DNA templates
- Multiplex PCR
- Specific amplification of complex templates
- 16S/23S screening
- RT-PCR

Limitation of Use

This product is for in vitro research only and not for clinical diagnostic.

Product Specifications

Shipping and Storage

FastGene® BAC-free HS Taq PCR kits are shipped on ice packs. Upon arrival, store kit components at -20 °C in a constant-temperature freezer. When stored under these conditions and handled correctly, full activity of the kit is retained until the expiry date indicated on the kit label. FastGene® BAC-free HS Taq Buffer contains isostabilizers and may not freeze solidly, even when stored at -20 °C. This will not affect the shelf-life of the product.

Handling

Always ensure that the product has been fully thawed and mixed before use. Reagents may be stored at 4 °C for short-term use. Return to -20 °C for long-term storage.

Quality Control

Each batch of FastGene® BAC-free HS Taq DNA Polymerase is confirmed to contain <1% contaminating protein (SDS-PAGE). The FastGene® BAC-free HS Taq PCR kits are subjected to stringent quality control tests, are free of contaminating exo- and endonuclease activity, as well as RNase-free and meet strict requirements with respect to DNA contamination levels.

Kit Codes and Components					
LS33S LS33	FastGene® BAC-Free HotStart Taq FastGene® BAC-Free HotStart Taq	50 Units 500 Units			
Related Products					
LS20 LS21 LS22 LS23 LS24 LS25 LS26 LS27 LS28 LS29 LS30 LS31	FastGene® Taq DNA Polymerase FastGene® Taq DNA Polymerase FastGene® Taq DNA Polymerase FastGene® HotStart Taq DNA Polymerase FastGene® HotStart Taq DNA Polymerase FastGene® HotStart Taq DNA Polymerase FastGene® Taq Ready Mix PCR FastGene® Taq Ready Mix PCR FastGene® Optima Polymerase Blend FastGene® Optima HotStart ReadyMix FastGene® Optima Polymerase Blend FastGene® Optima HotStart ReadyMix FastGene® Optima HotStart ReadyMix	100 Units 500 Units 2000 Units 100 Units 250 Units 1000 Units 50 x 50 µl rxns 250 x 50 µl rxns 250 Units 500 x 25 µl rxns 50 Units 20 x 25 µl rxns			

Quick Notes

- The FastGene® BAC-free HS Taq DNA Polymerase is produced in an eukaryotic expression system.
- The FastGene® BAC-free HS Taq DNA Polymerase is free of bacterial DNA.
- The FastGene® BAC-free HS Taq DNA Polymerase needs a 10 minute heat activation step at 95 °C.

Kit Content

- FastGene Bac free HS Tag
- 10 x Reaction buffer
- dNTP mixture (2 mM each)
- Sterile water

Contact & Support

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For technical support please contact: support@nippongenetics.eu



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FastGene® BAC-Free HotStart Taq Protocol

Step 1: Prepare the PCR master mix

- Ensure that all reagents are properly thawed and mixed.
- Prepare a PCR master mix containing the appropriate volume of all reaction components common to all or a subset of the reactions to be performed.
- Calculate the required volumes of each component based on the following table:

Component	20 μl rxn	Final conc.	
PCR-grade water	Up to 20 μl	N/A	
10 x Buffer	2 μΙ	1X	
FastGene® BAC-Free HotStart Taq*	0.2 μΙ	1 Unit	
dNTP (2 mM each)	2 μΙ	0.2 mM each	
Template (0.1 - 500 ng/ μl)**	1 μΙ	as required	
Forward Primer (10 µM)	0.5 μΙ	250 nM	
Reverse Primer (10 µM)	0.5 μΙ	250 nM	

^{*}Add the polymerase at the final step

Step 3: Run the PCR

• Perform PCR with the following cycling protocol:

Step	Temperature	Duration	Cycles	
Initial denaturation	95 °C	10 min¹	1	
Denaturation	95 °C	30 sec		
Annealing ²	60 - 65 °C	20 - 30 sec	25-35	
Elongation	72 °C	1 min / kB		
Final Elongation	72 °C	5 min	1	

 $^{^1}$ Initial denaturation for 10 min at 95 °C is recommended for most assays. 2 An annealing temperature 5 °C lower than the calculated melting temperature ($T_{\rm m}$) of the primer set is recommended as a first approach. If low yields and/or nonspecific amplification is obtained, an annealing temperature gradient PCR is recommended to determine the optimal annealing temperature of the primer pair.

Step 2: Set up individual reactions

- Transfer the appropriate volume of PCR master mix, template and primer to individual PCR tubes/wells or a PCR plate.
- Cap or seal individual reactions, mix and centrifuge briefly.

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^{**} Plasmid DNA 0.1 ng to 30 ng| genomic DNA 50 ng - 500 ng