Datasheet

Features:
- Enzyme with hot start capability increases reaction specificity and sensitivity, optimized
- DFS-Taq PLUS DNA polymerase activation requires not more than 5 min heating
- High selectivity and reaction yield
- The mix is colored for easy pipetting
- Reduced preparation time

Applications:
- Real-time PCR with oligonucleotide probes
- Conventional PCR
- High-throughput PCR
- Genotyping
- For detection of bacterial DNA

Description:
Bio-Star qPCR-Mastermix (2×) NO-ROX for probes is developed for quantitative real-time PCR with fluorescent dye SYBR Green I. The Mastermix contains all components, except template and primers, for successful PCR.

- The mix is optimized for efficient and reproducible hot-start real-time PCR of genomic, plasmid and viral DNA samples.
- The solution contains substances that increase half-life and processivity of DFS-Taq PLUS DNA polymerase by enhancing its stability during PCR.
- It includes components that influence primer annealing temperature and characteristics of template melting thus enabling to increase the specificity of PCR and use templates with complicated structure.
- DFS-Taq Plus DNA polymerase is inactive at room temperature because of monoclonal antibodies.
- The inert dye allows control when using multi-well plates. Use of the kit saves time and minimizes contamination risk due to reduced number of pipetting steps.

Components and Mixture
2X Bio-Star qPCR-Mastermix no-ROX for probes contains:

- 100 mM Tris-HCl (pH 8.5 at 25 °C), 100 mM KCl, 0.4 mM each of dNTP, 10 mM MgCl2, 0.1 U/µl DFS-Taq DNA Polymerase, 0.025% Tween 20, stabilizers and enhancers,
- Tube of MgCl2 100 mM
- Water Mol.Bio Grade 2 x 1,25 ml

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Storage and transportation: at -20 °C; not more than 50 thawing-freezing cycles. shipping with blue ice or at room temperature

Storage terms: up to 24 mounts

Stability tests / Quality control / Comparison
Exodeoxyribonuclease activity: DNA was stable after incubation of 1 µg fragment of phage lambda DNA in the presence of 25 µl of ***Bio-Star qPCR Mastermix (2×)*** in 50 µl reaction solution at 37 °C and 70 °C for 4 h.

Test for bacterial DNA contamination

Test for Hot -Start (we test activity with or without MAB at various temperatures

Exodeoxyribonuclease activity: DNA was stable after incubation of 1 µg fragment of phage lambda DNA in the presence of 25 µl of BioMaster HS-qPCR (2×) in 50 µl reaction solution at 37 °C and 70 °C for 4 h.

Ribonuclease activity: Absence of ribonuclease activity was confirmed after incubation of 1 µg of 5′-[P32]-labeled RNA fragment in the presence of 25 µl of BioMaster HS-qPCR (2×) in 50 µl reaction solution at 37 °C for 4 h.

Stability: One month at room temperature does not reduce PCR efficiency

Amplification protocol
1. Defrost the reaction mixture and stir thoroughly.
2. Add the following components into the thin-wall PCR tubes considering the final volume of a reaction mixture equal to 50 µl:

Component	Volume	Final concentration
2x Mastermix	25 µl	1x
Forward Primer	variable	0.1 - 600 nM
Reverse Primer	variable	0.1 - 600 nM
DNA Template	variable	10 pg - 1 µg
Sterile Water	up to 50 µl	

3. Gently vortex and remove droplets by centrifugation.
4. Perform PCR using temperature conditions recommended below:

Step	Temp.°C	Incubation time	Number of Cycles
Preliminary denaturation	95	3-7 min	1
Denaturation	95	15 sec	25-40
Annealing	50-68	10-30 sec	25-40
Elongation	58-72	30-60 sec	25-40

or alternatively:

Step	Temp.°C	Incubation time	Number of Cycles
Preliminary denaturation	95	3-7 min	
Denaturation	95	15 sec	30-50
Annealing/Elongation	50-68	1 min	30-50

5. The PCR results are displayed as amplification curve

Order Information

Prod. No.	Description	Quantity
P9120	Bio-Star qPCR-Master mix for probes (no ROX)	100 rcs (2,5 ml)