Biozym Red HS Taq Master Mix

Product information

Store at -20°C.
The kit may be stored at 4°C for short term use (up to 1 month). Multiple freeze/thaw cycles (up to 30) are possible with no loss of activity.

Component	331126X 50 rxn/20 µl	331126S 100 rxn/20 µl	331126L 500 rxn/20 µl	331126XL 2500 rxn/20 µl
Biozym Red HS Taq 2x Mix	1 x 0.5 ml	1 x 1.0 ml	5 x 1.0 ml	25 x 1.0 ml

For research use only.

1. Introduction

Biozym Red HS Taq Master Mix is designed for use in all routine and high throughput PCR applications including multiplex PCR, genotyping, screening, library construction, colony PCR and direct PCR. The robust enzyme ensures consistent results on a broad range of templates.

Using latest technology the polymerase enables short extension times, enhanced specificity and improved yields – ideal for complex templates and multiplex PCR. The used hot-start technology inhibits DNA polymerase activity at ambient temperature, thus preventing amplification of non-specific products.

Biozym Red HS Taq Master Mix is a convenient 2x mix containing HS Taq Polymerase, nucleotides and optimized reaction buffer including $MgCl_2$. Only template and primer need to be added by the user.
The master mix also include a density reagent and tracking dye for directly loading onto agarose gels.

2. Notes

- Biozym HS Taq DNA Polymerase produces A-tailed ends.
- Error rate: Approximately 1 error per 2.0 x 10^5 nucleotides incorporated.
- Biozym Red HS Taq Master Mix is particularly resistant to PCR inhibitors. Direct PCR could be performed from unprocessed samples including bacterial culture, bacterial colonies, blood and urine. (see 5.8)

3. Technical support

For technical support please contact support@biozym.com

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4. Reaction conditions for PCR

Carefully mix and centrifuge the tube before opening to ensure homogeneity and improve recovery.

Table 1. Pipetting instructions

Component	50 µl reaction	20 µl reaction	Final conc.	Notes
Biozym Red HS Taq Master Mix	25 µl	10 µl	1x	
Forward primer (10 µM)	2.0 µl	0.8 µl	400 nM	See 5.2 for primer design
Reverse primer (10 µM)	2.0 µl	0.8 µl	400 nM	
Template DNA cDNA gDNA	 <100 ng <0.5 µg	 <40 ng <0.2 µg	Variable	See 5.3 for template
PCR grade water	Up to 50 µl reaction volume	Up to 20 µl reaction volume		

Table 2. Cycling instructions

Cycles	Temperature	Time	Notes
1	95°C	1 - 2 min	Initial denaturation and enzyme activation. For colony PCR increase to 10 min.
40	95°C 55 to 65°C 72 °C	15 seconds 15 seconds 1 - 90 seconds	Denaturation Annealing Extension (15 s/kb). For multiplex PCR use 90 seconds

5. Notes about reaction components and cycling conditions

5.1. Biozym Red HS Taq Master Mix

The 2x mix contains Biozym HS Taq DNA Polymerase, 6mM $MgCl_2$, 2mM dNTPs, enhancers, stabilizers and a red dye for tracking during agarose electrophoresis. We do not recommend to adding further PCR enhancers or $MgCl_2$.

5.2. 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.

5.3. Template

For eukaryotic DNA use between 5 ng and 500 ng per reaction, for cDNA use below 100 ng per reaction.

5.4. Annealing

Use an annealing temperature equal to the Tm of the lower Tm primer. If non-specific products are present increase in 2°C increments. Alternatively use a temperature to experimentally find the optimal annealing temperature.

5.5. Extension

Extension should be performed at 72°C. The optimal extension time is dependent on amplicon length and complexity of template. We recommend an extension time of 15 seconds per kilobase (kb) for amplification from eukaryotic DNA (for amplicons between 1 kb and 6 kb).

5.6. Multiplex PCR

When first performing multiplex PCR it is recommended to run a temperature gradient around the calculated annealing temperatures. The annealing temperature representing best specificity should be used in subsequent experiments. Fast cycling conditions should not be used. Initially a 90 second extension time is recommended. To increase yield this time may be extended further.

5.7. Colony PCR

Use a sterile tip to pick a bacterial colony and resuspend into a 50µl reaction as described in table 1 above. From liquid culture add 5µl of overnight culture to the master mix. For lysis of cells initial denaturation time should be 10 minutes.

5.8. Direct blood/urine PCR

Add 2µl mammalian blood or urine to a 50µl reaction as described in table 1.

5.9. Tracking dye migration

The 2x mix contains a red dye for tracking during gel electrophoresis. In a 2% agarose TAE gel the dye migrates at a rate equivalent to 350bp of DNA, in a 1% agarose TAE gel equivalent to 600bp of DNA.