ELISABETH PHARMACON Ltd.
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EliZyme™ HS FAST

Intended use:

For Research Use Only. Not for use in diagnostic procedures.

Storage:

Upon arrival store components at -20 °C. Avoid prolonged exposure to light. When stored under these conditions, the kit will retain full activity until the expiration date indicated on the kit label. Reagents may be stored at 4 °C up to 1 month.

Product description

EliZyme HS FAST uses hot-start technology to inactivate the enzyme below 65 °C preventing primer-dimer formation and non-specific amplification. DNA polymerase in **EliZyme HS FAST** is inactivated until the initial activation step at 95 °C. **EliZyme HS FAST** is suited for difficult PCR templates. The mix is resistent to PCR inhibitors allowing direct PCR from unprocessed samples including bacterial culture, bacterial colonies, blood and urine.

EliZyme HS FAST is a robust enzyme system suited for routine PCR, multiplex PCR, amplification of DNA for Sanger sequencing and other genotyping applications. The enzyme system is characterised by enhanced PCR speed, yield and specificity. **EliZyme HS FAST** delivers exceptional PCR performance on complex templates including GC-rich and AT-rich sequences. **EliZyme HS FAST Taq DNA Polymerase** has 5'-3' exonuclease activity but no 3'-5' exonuclease (proofreading) activity. The error rate is approximately 1 error per 2.0 x 10^5 nucleotides incorporated. PCR products generated with **EliZyme HS FAST** are A-tailed and may be cloned into TA vectors.

For higher comfort is **EliZyme HS FAST** also available in a 2x ready mix. **EliZyme HS FAST MIX Red** contains a red dye for tracking during agarose gel electrophoresis. It is suitable for direct loading onto agarose gel.

Content

	Ref. No.	Content	Size
EliZyme HS FAST	EZ5505	1x0.1 ml 5 U/µl + 4x1 ml buffer	500 U
	EZ5510	2x0.1 ml 5 U/µl + 1x8 ml buffer	1000 U
	EZ5520	4x0.1 ml 5 U/µl + 2x8 ml buffer	2000 U
EliZyme HS FAST MIX	EZ5720	5x1 ml mix	200 rxns
	EZ5760	2x7.5 ml mix	600 rxns
EliZyme HS FAST MIX Red	EZ5620	5x1 ml mix	200 rxns
	EZ5660	2x7.5 ml mix	600 rxns

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	Buffer/MIX	Content
EliZyme HS FAST	5x buffer	15 mM $MgCl_2$, 5 mM dNTPs
EliZyme HS FAST MIX	2x mix	6 mM $MgCl_2$, 2 mM dNTPs
EliZyme HS FAST MIX Red	2x mix Red	6 mM $MgCl_2$, 2 mM dNTPs

Additional $MgCl_2$ is not necessary. The buffer composition has been optimised to maximise PCR success rates.

Primers

Primers should have a predicted melting temperature of around 60 °C. Primers should be designed to eliminate the possibility of primer-dimer formation and nonspecific amplification. The final primer concentration in the reaction should be between 0.2 µM and 0.6 µM.

PCR

It is not recommended to use fast cycle conditions for multiplex PCR. To increase yield, 90 second final extension time should be used.

For colony PCR use a sterile tip to pick a bacterial colony and resuspend into a 50 µl reaction as described below. From liquid culture add 5 µl of overnight culture to the final mix. Initial denaturation time is increased to 10 minutes.

For direct PCR add 2 µl of blood or urine to 50 µl reaction as described below.

Reaction setup

EliZyme HS FAST

After thawing, briefly vortex 5x EliZyme Reaction Buffer and shortly spin.

Reagent	50 µl reaction	Final conc.
5x EliZyme Reaction Buffer	10 µl	1x
Forward primer (10 µM)	2 µl	400 nM
Reverse primer (10 µM)	2 µl	400 nM
Template DNA	5 – 500 ng genomic DNA, < 100 ng cDNA	Variable
EliZyme HS Taq DNA Polymerase (5 U/µl)	0.25 – 1 µl	
PCR grade water	Up to 50 µl	

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EliZyme HS FAST MIX

After thawing, briefly vortex the mix and shortly spin.

Reagent	50 µl reaction	Final conc.
2x EliZyme HS Taq MIX	25 µl	1x
Forward primer (10 µM)	2 µl	400 nM
Reverse primer (10 µM)	2 µl	400 nM
Template DNA	5 – 500 ng genomic DNA, < 100 ng cDNA	Variable
PCR grade water	Up to 50 µl	

EliZyme HS FAST MIX Red

After thawing, briefly vortex the mix and shortly spin.

Reagent	50 µl reaction	Final conc.
2x EliZyme HS Taq MIX Red	25 µl	1x
Forward primer (10 µM)	2 µl	400 nM
Reverse primer (10 µM)	2 µl	400 nM
Template DNA	5 – 500 ng genomic DNA, < 100 ng cDNA	Variable
PCR grade water	Up to 50 µl	

PCR cycling profile

Step	Temperature	Time	Cycles
Initial denaturation	95 °C	1 – 2 min*	1
Denaturation	95 °C	15 s	40
Annealing	55 – 65 °C	15 s	
Extension	72 °C	1 – 90 s**	
Final extension	72 °C	90 s***	1

*For PCR from bacterial colonies extend initial denaturation time to 10 minutes
**15 s/kb, for amplicons shorter than 1 kb may be used 1 second extension
***Only for multiplex PCR

Manufacturer:

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REF Catalog number	Upper limit of temperature
LOT Batch code	Manufacturer
Use by (last day of month)	Σ Contains sufficient "N" tests