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HiDi Taq DNA polymerase

#9201
Store at -20°C.

Contents

HiDi (**Hi**gh **Di**scrimination) Taq DNA polymerase, 5 U/µl
HiDi reaction buffer, 10x

Description

HiDi Taq DNA polymerase is a highly selective DNA polymerase variant, specially evolved for all assays in which **Hi**gh **Di**scrimination is required, for instance in allele-specific PCRs, primer extensions or methylation-specific PCRs.
HiDi Taq DNA polymerase efficiently amplifies from primers that are matched at the 3'-end and discriminates primers that are mismatched. An aptamer-based hot-start formulation of the HiDi Taq DNA polymerase prevents false amplification. Temperatures above 50°-55°C cause the aptamer's secondary structure to melt and will set-free the polymerase.

HiDi Taq variant has a 5'-3'-nuclease activity and therefore can be used for hydrolysis probe-based real-time PCRs.

Quality Control Assays

PCR activity: HiDi Taq DNA polymerase is tested successfully for hydrolysis probe based real-time PCR. The product demonstrates linearity of amplification over a specified serial dilution of human genomic DNA.
DNA polymerase activity: HiDi Taq DNA polymerase activity is monitored and adjusted to a specific DNA polymerase activity using an artificial DNA template and a DNA primer.
Enzyme-concentration is determined by protein-specific staining. Please inquire more information at info@mypols.de for the lot-specific concentration.
No contamination has been detected in standard test reactions.

Safety

This product does not require a Material Safety Data Sheet because it does neither contain more than 1% of a component classified as dangerous or hazardous nor more than 0.1% of a component classified as carcinogenic. However, we generally recommend the use of gloves, lab coats and eye protection when working with these or any other chemical reagents. myPOLS Biotec takes no liability for damage resulting from handling or contact with this product. Further information can be found in the REGULATION (EC) No 1272/2008 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL.

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Applications

- SNP-detection by allele-specific amplification (ASA) / Allele-specific PCR
- Genotyping and genomic profiling
- Real-time PCR with fluorescence-based hydrolysis probes
- Real-time multiplex detection PCR

Recommendations for PCR/ Reaction Setup

PCR Mix

Component	Volume	Final concentration
Primer forward (10 µM)*	1 µl	0.2 µM (0.05-1 µM)
Primer reverse (10 µM)*	1 µl	0.2 µM (0.05-1 µM)
dNTPs (2 mM)	5 µl	200 µM
HiDi buffer (10x)	5 µl	1x
HiDi Taq polymerase 5 U/µl	0. 5 µl	2.5 U/reaction
Template/Sample extract	x µl	
Nuclease-free water		up to 50µl total vol.

* Primers should ideally have a GC content of 40-60% typically

Typical 3-step PCR protocol

Initial denaturation	95°C	2 min	
Denaturation	95°C	15 sec	25-40 cycles
Annealing*	54–72°C	10 sec	
Extension	72°C	30 sec/250 bp	
Hold	<10°C		

* Typically, the annealing temperature is about 3-5°C below the calculated melting temperature of the primers used.

Important notes

- Keep all components on ice.
- Spin down and mix all solutions carefully before use.
- HiDi 10x buffer is optimized for short amplicon length (about 60-200 bp, but also longer amplicon lengths are possible. The addition of additional Magnesium (+ 0.5 - 1.5 mM) might be needed in case of longer amplicons >500 bp.
- HiDi Taq DNA polymerase has a 5'-3'-nuclease activity and therefore can be used for hydrolysis probe-based assays.
- HiDi Taq DNA polymerase is not suitable for real-time PCRs using a real-time dye such as SYBR Green. In this case, HiDi DNA polymerase (#9001) is recommended.

References

HiDi Taq DNA polymerase is based on:
Variants of a Thermus aquaticus DNA Polymerase with Increased Selectivity for Applications in Allele- and Methylation-Specific Amplification. PLoS ONE 2014; 9(5): e96640. M. Drum, R. Kranaster, C. Ewald, R. Blasczyk, and A. Marx.

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Product source: recombinant protein expression in E.coli.