Product information

Product Name: Taq DNA Polymerase

Catalog No: VN160E, VN161E

Packing Size: 125 µl and 50 µl

Shipping Condition: Ambient temperature

Storage Condition: -20^{0}C

Product Description:

Taq DNA Polymerase is a highly thermostable DNA polymerase derived from the thermophilic bacterium Thermus aquaticus. It catalyzes the 5'→3' synthesis of DNA, has low 5'→3' exonuclease activity, and lacks detectable 3'→5' exonuclease (proofreading) activity. Taq DNA Polymerase also exhibits deoxynucleotidyl transferase activity, which can result in the addition of extra adenines at the 3'-end of PCR products.

Recombinant Taq DNA Polymerase is the ideal enzyme for standard PCR amplification of purified DNA templates. It can also be used in real-time PCR assays, including those with DNA binding dyes, such as SYBR Green and Eva Green, as well as TaqMan assays that require 5'-exonuclease activity.

10X Reaction Buffer:

500 mM Tris-HCl pH 9.1, 160 mM ammonium sulfate, 0.25% Brij-58, and 25 mM magnesium chloride.

Protocol:

PCR setting for a 25 µl reaction

Reagent	Volume	Final Concentration
10x Taq Mutant Buffer	2.5 µl	1x
dNTP Mix (10 mM)	0.5-1.0 µl	200-400 µM each
Left Primer	Variable	0.2-0.4 µM
Right Primer	Variable	0.2-0.4 µM
DNA template†	Variable	0.1-100 ng
PCR Enhancer Cocktail (optional)*	Titration	Variable
Taq Polymerase**	0.05-0.25 µl	
De-ionized Distilled H_2O	Adjust final volume to 25 µl	-

The amount of DNA required for PCR depends mainly on the genome size and the copy number of the target gene.

For optimal performance, we recommend using one of our PCR Enhancer Cocktails (PEC) if your samples contain PCR inhibitors. Please note that the PEC is not included with the enzyme and should be ordered separately if needed. We also suggest performing a titration of the PEC to determine the optimal amount to use. Please refer to the PEC datasheet for further details.

To determine the optimal enzyme concentration for a specific target, we strongly recommend conducting an enzyme titration test. A good starting amount of enzyme for a 25 µl reaction with purified DNA is 0.05 µl. However, targets larger than 1 kb may require more enzyme or the use of the LA version.

Typical Cycling Parameters

Initial denaturation	95^{0}C	2 min
25-40 cycles		
Denaturation	94^{0}C	10 to 30 sec
Annealing	50^{0}C to 68^{0}C	20 to 60 sec
Extension	70^{0}C	1-2 min / 1kb target
Final Extension	70^{0}C	5 min
Hold	4^{0}C	

Troubleshooting guide

No PCR products	Please check your PCR settings to ensure that all necessary components are included in the PCR mix. It's also recommended to perform a gradient annealing temperature experiment to determine the optimal temperature for your specific target. This can help to improve PCR efficiency and specificity, resulting in more reliable and consistent results.
The bands in agarose gel are smear	Enzyme titration for a particular target. It involves testing a range of enzyme concentrations to find the concentration that produces the most efficient and accurate results. It's important to note that using too much enzyme can actually inhibit the PCR reaction or overamplify the target, especially when working with short and simple target genes. Additionally, it's important to check for DNA and primer degradation as degraded DNA or primers can also negatively impact the PCR results.
Low yield of products	You can try the following strategies to improve PCR performance: • Conduct an enzyme titration experiment to optimize the enzyme concentration for your specific target. • Increase the extension time to ensure complete amplification of the target DNA. • Try a gradient annealing temperature experiment to identify the optimal annealing temperature for your primers. • Consider redesigning your primers if the amplification is not specific or efficient enough.
Non-specific products are observed	If you are observing non-specific products in your PCR reaction, there are several strategies you can try: • Perform a gradient annealing temperature experiment to identify the optimal temperature for your primers, which can help to reduce non-specific products. • Check the GC content of your target sequence. If it's above 65%, it may be necessary to use our PEC-GC. • Evaluate your primers and redesign them if necessary to improve specificity and reduce non-specific products.

Note: This product is for R&D use only

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