Parstous

Spidi ™ Pfu DNA polymerase

Cat: C101101 (250 U, 50 µL)
Cat: C101102 (500 U, 100 µL)
Store at -20 °C

Component	C101101	C101102
Spidi ™ Pfu DNA poly. 5 U/µl	250 U	500 U
$MgCl_2$ Solution 25 mM	0.5 ml	1ml
5X Buffer $MgCl_2$ free	1 ml	1.7 ml

Contents

Description:

Spidi ™ Pfu DNA Polymerase is a chimeric Pfu which has a DNA binding protein at the N-terminal portion of the gene. This enzyme keeps significant activity after exposure to 99 °C or repeated exposure to 98 °C with more processivity and extention rate than Pfu DNA polymerase. It catalyzes the polymerization of nucleotides into duplex DNA in the 5'_3' direction, resulting in blunt-ended PCR products **without 3'-dA overhangs.**

Spidi ™ Pfu DNA Polymerase exhibits 3'_5' exonuclease (proofreading) activity that enables the polymerase to correct the mis-incorporation of nucleotide, and lacks 5'_3' exonuclease activity. It is suitable for PCR and primer extension reaction that requires high fidelity when the PCR fragment is relatively **higher than 3 kb.**

The enzyme exhibits 3´>5´ proofreading activity, resulting in over 20-fold higher PCR fidelity than possible with Taq DNA Polymerases

Kit storage:

This kit should be stored at -20 °C. Under this conditions reagents are stable for two years from the date of production.

Component	Volume	Final Conc.
5X Reaction Buffer	4 µL	1X
$MgCl_2$ Solution 25 mM	1.2 µL	1.5 mM
40 mM dNTPs Mix (10 mM each)	0.5 µL	0.25 mM
Upstream Primer (10 pmol/ µL)	1 µL	0.5 pmoles/µL
Downstream Primer (10 pmol/ µL)	1 µL	0.5 pmoles/µL
Template DNA	Variable	10 fg~1 µg
PCR grade water	Variable	-
Spidi ™ Pfu poly. 5 U/µl	0.25 µL	
Total Volume	20 µL	-

General Reaction Protocol:

1. Thaw 5X reaction buffer, dNTP mixture.
2. Mix the master mix thoroughly and dispense appropriate volumes into PCR tubes or plates.
3. Add templates DNA to the individual PCR tubes or wells containing the master mix.
4. Program the PCR machine according to the program outlined.

Cycle	Time	Temp °C
1	1 min	98
30-35	10 sec	98
	30 sec	60
	30 sec	72
1	2 min	72

Notes:

∗ Longer extension time makes nonspecific bands
∗Extension rate for this enzyme is near 3000 bp/min.

Agarose gel Electrophoresis:

Run the total 5-7 µL of PCR products alongside 3 µL DNA marker on a 2% agarose gel containing Green viewer DNA safe stain.

Disclaimers and Addresses:

This product is for **Research Use Only** and should only be used by trained professionals.

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