TransStart® FastPfu DNA Polymerase

Please read the datasheet carefully prior to use.

Cat. No. AP221

Version No. Version 2.0

Storage: at -18°C or below for two years

Concentration: 2.5 units/μl

Description

TransStart® FastPfu DNA Polymerase is a hot start high-fidelity DNA polymerase used for fast PCR. Offering a high amplification efficiency and high extension rate (4 kb/min, it is 8 times that of ordinary Pfu enzymes), *TransStart*® FastPfu DNA Polymerase amends the defects of low amplification efficiency, low yield and low extension rate (0.5 kb/min) in common Pfu polymerase, and greatly shortens reaction time.

• Offers 54-fold fidelity as compared to *EasyTaq*® DNA Polymerase.

• Blunt-end PCR products can be directly cloned into *pEASY*®-Blunt vectors.

• Amplification of genomic DNA fragment up to 15 kb.

• Amplification of plasmid DNA fragment up to 20 kb.

Features

• Hot start, high specificity.

• High amplification efficiency.

• Fast and high fidelity.

Applications

• Amplifies complex and high GC/ AT templates.

• High fidelity and fast PCR, blunt end cloning, site-directed mutagenesis.

• Amplifies long fragment.

Kit Contents

Component	AP221-01/11	AP221-02/12	AP221-03/13
TransStart® *FastPfu* DNA Polymerase	250 U×1	500 U×1	500 U×6
5×*TransStart*® *FastPfu* Buffer	1.2 ml×1	1.2 ml ×2	1.2 ml ×12
2.5 mM dNTPs	-/ 500 μl×1	-/ 1 ml ×1	-/ 1 ml ×6
6×DNA Loading Buffer	500 μl×1	1 ml ×1	1 ml ×2
50 mM $MgSO_4$	200 μl×1	400 μl×1	1 ml ×1
Complimentary Component	200 μl×1	400 μl×1	1 ml ×1
PCR Stimulant			

Storage Buffer

50 mM Tris-HCl pH 8.2, 0.1 mM EDTA, 1 mM DTT, Stabilizers, 50% glycerol

5×*TransStart*® FastPfu Buffer (with Mg^{2+})

100 mM Tris-SO_4 (pH 9.2), 200 mM KCl, 50 mM $(NH_4)_2SO_4$, 10 mM $MgSO_4$, 10% Glycerol, others

Recommended PCR System and Condition (50 µl reaction as an example)

Component	Volume	Final Concentration
Template	Variable	as required
Forward Primer (10 µM)	1 µl	0.2 µM
Reverse Primer (10 µM)	1 µl	0.2 µM
5×*TransStart*® *FastPfu* Buffer	10 µl	1×
2.5 mM dNTPs	4 µl	0.2 mM
TransStart® *FastPfu* DNA Polymerase	1 µl	2.5 units
Nuclease-free Water	Variable	-
Total volume	50 µl	-

Suggested Conditions (50 µl reaction volume)

Template	Input
Genomic DNA	10-500 ng
Plasmid DNA	1-30 ng
cDNA	1-2 µl cDNA from RT reaction (50-500 ng RNA for RT reaction)

PCR

Number of Cycles	Temperature	Time
1 cycle	95℃	2 min
30-35 cycles	95℃	20 sec
	Tm-5℃	20 sec
	72℃	4 kb/min
1 cycle	72℃	5 min

PCR Stimulant

PCR Stimulant is used to optimize the amplification of complex templates or high GC/ AT templates. The amplification of the Pfu series of enzymes is enhanced significantly. The concentration of the storage solution is 5×, and the concentration of the working solution can be adjusted between 0.5×-2.5×.

Notes

- For GC-rich templates, the recommended denaturation temperature is 98°C.
- To ensure high fidelity, we recommend using high quality dNTPs. dNTPs containing dUTP cannot be used.
- It is recommended to add *TransStart*® FastPfu DNA Polymerase to the reaction system in the last step.
- If 5×*TransStart*® FastPfu Buffer has a small amount of precipitation after thawing, please heat it in a 37°C water bath and mix it for use.

For research use only, not for clinical diagnosis.

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