

TransStart® FastPfu Fly DNA Polymerase

Please read the datasheet carefully prior to use.

Cat. No. AP231

Version No. Version 5.0

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

Concentration: 2.5 units/μl

Description

TransStart® FastPfu Fly DNA Polymerase is a hot start, ultra high-fidelity DNA Polymerase used for fast PCR. Compared with TransStart® FastPfu DNA Polymerase, TransStart® FastPfu Fly DNA Polymerase has higher extension rate (≤ 5 kb fragments can achieve 12 kb/min extreme amplification, > 5 kb fragments can achieve 6 kb/min high-speed amplification), featuring higher amplification efficiency, higher yield, higher fidelity and higher sensitivity. 2×TransStart® FastPfu Fly Reaction Mix already contains dNTPs. When DNA is amplified, just add template, primer, water and TransStart® FastPfu Fly DNA Polymerase. The Reaction can be carried out with the concentration of Reaction Mix being 1×. PCR products are blunt end and can be cloned directly into pEASY®-Blunt series of vectors.

- Offers 108-fold fidelity as compared to EasyTaq® DNA Polymerase.
- PCR products are blunt end and can be cloned directly into pEASY®-Blunt series of 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 ultra high-fidelity.
- High sensitivity.
- High yield.

Applications

- Complex templates, GC/AT-rich templates.
- Ultra high-fidelity and fast PCR, blunt end cloning, site-directed mutagenesis.
- Long fragment amplification.

Kit Contents

Component	AP231-21	AP231-22	AP231-23
TransStart® FastPfu Fly DNA Polymerase	250 U×1	500 U×1	500 U×6
2×TransStart® FastPfu Fly Reaction Mix	1 ml×3	1.2 ml×5	1.2 ml×30
50 mM MgSO ₄	200 μl×1	400 μl×1	1 ml×1
6×DNA Loading Buffer	500 μl×1	1 ml×1	1 ml×2
PCR Stimulant	200 μl×1	400 μl×1	1 ml×1

Recommended PCR System and Conditions (taking 50 μl reaction system 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
2×TransStart® FastPfu Fly Reaction Mix	25 μl	1×
TransStart® FastPfu Fly DNA Polymerase	1 μl	2.5 units
Nuclease-free Water	Variable	-
Total volume	50 μl	-



Optimized parameters (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	98°C	1 min
30-35 cycles	98°C	10 sec
	Tm-5°C	5 sec
	72°C	6 or 12 kb/min *
1 cycle	72°C	1 min

*For fragments of 5 kb and below, select 12 kb/min; for fragments above 5 kb, select 6 kb/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×-2.0×.

Notes

- It is recommended to add *TransStart® FastPfu* Fly DNA Polymerase to the reaction system in the last step.
- Please thaw 2×*TransStart® FastPfu* Fly Reaction Mix thoroughly before use. If there is 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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