Gloria U 2X Mix for NGS

RK20723

Product components

Components	Cat.No	Size-1 1 mL	Size-2 5 × 1 mL
Gloria U 2X Mix for NGS	RM20410	1 mL	5 × 1 mL

Product Description

Gloria U hot-start DNA polymerase is a typical B-family polymerase, a modified version of Gloria Nova DNA polymerase. Due to the inactivation of inactivation of uracil-binding capacity of Gloria U polymerase, it can efficiently amplify bisulfite-treated DNA, and the enzyme shows the same high yield as Gloria Nova hot-start DNA polymerase. The enzyme exhibits the same high yield, low GC bias, and coverage uniformity as the Gloria Nova hot-start DNA polymerase. Gloria U 2X Mix for NGS has 5′-3′ sustained synthetic activity and 3′→5′ exonuclease (correction) activity and can be used for uracil-containing template amplification or uracil-containing library amplification.

Storage

Upon receipt, store all components at -20°C.

Standard Protocol

All components should be mixed and centrifuged instantaneously before use. After mixing the other reaction components, Gloria U 2X Mix for NGS can be added to prevent the degradation of primers by its 3 -5 exonuclease activity.

It is recommended to prepare the reaction system on ice and quickly transfer it to a PCR instrument pre-warmed at 98 °C.

Recommended Reaction

Take 50 μL reaction systems as examples.

Components	50 μL	Total Concentration
Gloria U 2X Mix for NGS	25 μL	1X
Forward Primer (5 μM)	1-2 μL	0.4-0.8 μM
Reverse Primer (5μM)	1-2 μL	0.4-0.8 μM
DNA Template*	Variable	
Nuclease-free Water	to 50 μL	N/A

Note: Gently mix the reaction mixture. Centrifuge the reaction mixture to ensure all components are at the bottom of the tube. To prevent evaporation, please overlay the sample with mineral oil if using a PCR machine without a heated lid..

*** , Note: Different DNA templates have different optimal reaction concentrations, please refer to the basic principles. Templates here refer to templates or libraries containing uracil.**

E-mail:cn. market@abclonal.com
TEL:400-999-6126

Recommended PCR Program

Step	Temp	Time	Cycles
Pre-denaturation	98 ℃	45 s	1
Denaturation	98 ℃	10 s	
Annealing	55-65 ℃	30s-1 min	Recommended number of
Extension	72 ℃	30-60 s / kb	cycles are based on input
Post-extension	72 ℃	1-5 min	1
Hold	4-12 ℃	∞	1

Choose different cycles depending on the amount of input

Input Bisulfite gDNA	PCR cycles
5ng	15-16
25ng	11-12
50ng	9-10
100ng	8-9
200ng	7-8
Input FFPE DNA	**PCR cycles**
5ng	16-17
25ng	12-13
50ng	10-11
100ng	9-10
200ng	8-9
Input cfDNA	**PCR cycles**
10ng	16-17

Application

1. Achieve superior amplification of bisulfite-converted

Sodium bisulfite treatment converts unmethylated cytosines to uracil, while methylated cytosines are not. In subsequent PCR amplification, thymine replaces uracil, making it possible to precisely methylate DNA to the base pair level by sequencing. Gloria U 2X Mix for NGS provides high yields of bisulfite-transformed DNA with stable amplification.

2.Achieve superior amplification of damaged DNA (e.g., FFPE)

Cytosine deamination occurs on its own over a longer period of time. Deamination accelerates at elevated temperatures and leads to accumulation of uracils in DNA and free nucleotides. When other corrective enzymes are ineffective, Gloria U 2X Mix for NGS can perform high-fidelity amplification from damaged DNA templates containing uracil.

E-mail： cn.market@abclonal.com
TEL：400-999-6126