Heat-labile Uracil-N-Glycosylase (UNG)

Product Description

Heat-labile Uracil-N-Glycosylase (UNG) is a thermosensitive UNG enzyme obtained by recombinant expression in *Escherichia coli*. This enzyme catalyzes the release of free uracil from uracil-containing single-stranded and double-stranded DNA and exhibits no activity against RNA. Compared with conventional *E. coli*-derived UNG enzymes, Heat-labile UNG possesses higher enzymatic activity at low temperatures ranging from 20℃ to 37℃. Featuring prominent heat lability, the enzyme can be readily inactivated at 50℃. This property effectively prevents the degradation of dUTP-containing amplification products at ambient temperature, which is commonly triggered by the residual activity of conventional UNG after incomplete inactivation. Therefore, Heat-labile UNG is highly applicable to carryover contamination prevention in PCR reactions. It is well compatible with conventional RT-PCR amplification protocols and is suitable for contamination control in RT-PCR systems.

Components

Components	BR1A808-01 100U	BR1A808-02 500U
Heat-labile Uracil-N-Glycosylase (UNG)	0.1 mL	0.5 mL

Unit Definition

One unit of activity (U) refers to the amount of enzyme required to degrade 1 μg of single-stranded DNA containing dU bases within 1 h at 37℃.

Storage Buffer

20 mM Tris-HCl (pH 7.5), 100 mM NaCl, 0.1 mM EDTA, 1 mM DTT, Stabilizer, 50% Glycerol.

Storage

Store at -20±5℃.

Notes

1.For Research Use Only. Not for use in diagnostic procedures.

2.Heat-labile Uracil-N-Glycosylase (UNG) has a relatively low optimal reaction temperature. Optimize enzyme amount and reaction time within 20℃-37℃ using 0.1-0.5 U for 5-10 min. The enzyme can be inactivated during reverse transcription.

3.This product is suitable for carryover-contamination control in PCR and RT-PCR.

4.Avoid repeated freeze-thaw cycles. Do not expose the product to environments with large temperature fluctuations.

5.The efficiency of dUTP utilization and sensitivity to UNG enzyme varies for different genes to be amplified. If the detection sensitivity is reduced due to the use of the UNG system, adjust and optimize the reaction system accordingly. If technical support is required, please contact our company.

Quality Control

1.SDS-PAGE electrophoresis purity no less than 98%.

2.Amplification sensitivity, batch-batch difference, and stability.

3.No exogenous nuclease activity, no exogenous endonuclease or exonuclease contamination.

Prepare Reaction Mix

Component	Volume per Reaction	Concentration in Master Mix
10×PCR Buffer (Mg^{2+} free)	5 μL	1×
dNTPs (dATP, dGTP, dCTP)	—	200 μM
dUTP (replaces dTTP)	—	200-600 μM
25 mM $MgCl_2$	2-8 μL	1-4 mM
5 U/μL Taq	0.25 μL	1.25 U
1 U/μL Heat-labile Uracil-N-Glycosylase (UNG)	0.5 μL (0.1-0.5 μL)	0.5 U (0.1-0.5 U)
25×Primer Mix*	2 μL	1×
Template	—	< 1 μg/reaction
ddH_2O	To 50 μL	—

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* If used for qPCR or RT-qPCR, add a fluorescent probe to the reaction system. A final primer concentration of 0.2 μM usually gives good results. If reaction performance is poor, adjust the primer concentration within 0.2-1 μM. Probe concentration is usually optimized within 0.1-0.3 μM. Gradient concentration experiments can be performed to determine the optimal primer and probe combination.