E.coil UDG

Catalog # P061-01

Version 5.1

Vazyme biotech co., ltd.

Introduction

E. coli uracil-DNA glycosylase (UDG) catalyzes uracil-containing DNA and releases uracils. UDG hydrolyzes uracils effectively from single or double-stranded DNA, but not from oligonucleotide with less than 6 bases.

Package Information

Components	P061-01 500U
E. coli UDG (5 U/μl)	100 μl

Storage Buffer

10 mM Tris-HCl, pH 7.4@ 25°C
50 mM KCl
0.1 mM EDTA
1 mM DTT
0.1 mg/ml BSA
50% Glycerol (v/v)

Storage

Store at -20℃

Origin

Recombinant *E. coli* strain cloned with UDG gene of psychrophilic marine bacterium

Unit Definition

One unit (U) is defined as the amount of enzyme that releases 60 pmol of uracil from the double-stranded DNA (containing dU) per minute. The activity is determined through the [^{3}H]-uracil amount released from the reaction system, which includes 50 μl of solution containing 0.2 μg of DNA (10^4 - 10^5 CPM/dg) and is performed at 37°C for 30 minutes.

Quality Control

Exonuclease residue detection: DNA electrophoresis bands do not change when 10 U of this enzyme and 0.6 μg of λ-Hind III are incubated at 37°C for 16 hours.
Endonuclease residue detection: DNA electrophoresis bands do not change when 10 U of this enzyme and 0.6 μg of Supercoiled pBR322 DNA are incubated at 37°C for 4 hours.
RNase residue detection: RNA electrophoresis bands do not change when 10 U of this enzyme and 1 μg of total RNA of HeLa cell are incubated at 37°C for 1 hours.
E. coli DNA residue detection: *E. coli* genome residue of 200 U of this product should be less than 10 copies in TaqMan qPCR detection specified with *E. coli* 16s rDNA .

Protocol

1. Recommended reaction mixture for PCR:

ddH_2O	to 50 μl
10 × Taq Buffer(with 20 mM $MgCl_2$)	5 μl
25 mM $MgCl_2$[a]	Optional
dUTP[b]	0.6 mM
dATP/dCTP/dGTP	0.2 mM each
Template DNA	Optional
Primer 1 (10 μM)	2 μl
Primer 2 (10 μM)	2 μl
Taq DNA Polymerase (5 U/μl)	0.5 μl
E. coli UDG (1 U/μl)[c]	0.2 μl

a.The final concentration of Mg^{2+} can be adjusted between 2.0 and 3.0 mM according to experiment needs.

b.The final dUTP concentration can be adjusted to 0.2 - 0.6 mM according to experiment demands.

c.According to the requirements of the experiment, the general amount of the *E. coli* UDG in the 50 μl reaction system is 0.1 - 1 U.

2. PCR conditions:

37 ℃	10 min	U-containing template degradation
95 ℃	2 min	UDG inactivation, template degeneration
PCR Reaction		