Thermo SCIENTIFIC

PRODUCT INFORMATION

Uracil-DNA Glycosylase

#__ __

Lot: ___ Expiry Date: _

Concentration: 1 u/µl

Supplied with: _ ml of 10X Reaction Buffer

Store at -20°C

In total _ vials.

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Description

Uracil-DNA Glycosylase catalyzes the hydrolysis of the N-glycosylic bond between uracil and sugar, leaving an apyrimidinic site in uracil-containing single or double-stranded DNA. Shows no activity on RNA (1).

Applications

- Control of carry-over contamination in PCR (2).
- Glycosylase mediated single nucleotide polymorphism detection (GMPD) (3).
- Site-directed mutagenesis (4).
- As a probe for protein-DNA interaction studies (5).
- SNP genotyping.
- Cloning of PCR products (6).
- Generation of single strand overhangs of PCR products and cDNA.

Source

E.coli K12 cells.

Molecular Weight

25.6 kDa monomer.

Definition of Activity Unit

One unit of the enzyme catalyzes the release 1 nanomole of uracil from uracil-containing DNA template in 60 min at 37°C.

Storage Buffer

The enzyme is supplied in: 30 mM Tris-HCl (pH 7.5), 150 mM NaCl, 1 mM EDTA, 1 mM DTT, 0.05% (v/v) Tween 20 and 50% (v/v) glycerol.

10X Reaction Buffer

200 mM Tris-HCl (pH 8.2 at 25°C), 10 mM EDTA, 100 mM NaCl.

Inhibition and Inactivation

- Inhibitors: Ugi protein from the *Bacillus subtilis* phage PBS2, protein p56 from the *Bacillus subtilis* phage phi29 (7).
- Inactivated by heating at 95°C for 10 min. Enzyme activity is partially restored at temperatures lower than 55°C. Therefore put PCR products on ice after PCR and load directly on a gel.

Note

- The abasic sites formed in DNA by Uracil-DNA Glycosylase may be subsequently cleaved by heat, alkali-treatment or endonucleases that cleave specifically at abasic sites.
- UDG is active in the presence or absence of divalent cations.

CERTIFICATE OF ANALYSIS

Endodeoxyribonuclease Assay

No conversion of covalently closed circular DNA to nicked DNA was detected after incubation of 10 units of Uracil-DNA Glycosylase with 1 µg of pUC19 DNA for 4 hours at 37°C.

Ribonuclease Assay

No contaminating RNase activity was detected after incubation of 10 units of Uracil-DNA Glycosylase with 1 µg of [^{3}H]-RNA for 4 hours at 37°C.

Labeled Oligonucleotide (LO) Assay

No degradation of single-stranded and double-stranded labeled oligonucleotide was observed after incubation with 2 units of Uracil-DNA Glycosylase for 4 hours at 37°C.

Quality authorized by:  Jurgita Zilinskiene

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References

1. Lindahl, T., et al., DNA N-Glycosidases, J. Biol. Chem., 252, 10, 3286-3294, 1977.
2. Longo, M.C., et al., Use of uracil DNA glycosylase to control carry-over contamination in polymerase chain reactions, Gene, 93, 125-128, 1990.
3. Vaughan, P., McCarthy, T.V., A novel process for mutation detection using uracil DNA glycosylase, Nucleic Acids Res., 26, 810-815, 1998.
4. Kunkel, T.A., Rapid and efficient site-specific mutagenesis without phenotypic selection, Proc. Natl. Acad. Sci. USA, 82, 488-492, 1985.
5. Devchand, P.R., et al., Uracil-DNA glycosylase as a probe for protein-DNA interactions, Nucleic Acids Res, 21, 3437-3443, 1993.
6. Booth, P.M., et al., Assembly and cloning of coding sequences for neurotrophic factors directly from genomic DNA using polymerase chain reaction and uracil DNA glycosylase, Gene, 146, 303-308, 1994.
7. Serrano-Heras, G., et al., Protein p56 from the *Bacillus subtilis* phage phi29 inhibits DNA-binding ability of uracil-DNA glycosylase, Nucleic Acids Res, 13, 1-9, 2007.

PRODUCT USE LIMITATION

This product is developed, designed and sold exclusively *for research purposes and in vitro use only.* The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals.
Please refer to www.thermoscientific.com/onebio for Material Safety Data Sheet of the product.