

## Uracil-DNA Glycosylase (UDG)



1-800-632-7799  
info@neb.com  
www.neb.com



M0280S 012140216021

# M0280S



1,000 units    5,000 U/ml    Lot: 0121402  
RECOMBINANT    Store at -20°C    Exp: 2/16

**Description:** *E. coli* Uracil-DNA Glycosylase (UDG) catalyzes the release of free uracil from uracil-containing DNA. UDG efficiently hydrolyzes uracil from single-stranded or double-stranded DNA, but not from oligomers (6 or fewer bases).

**Source:** An *E. coli* strain that carries the cloned UDG gene from *E. coli*.

Supplied in: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 0.1 mg/ml BSA and 50% glycerol.

[More Units](#)

**Application:** Treatment of 0.1 µg of uracil-containing DNA with 1 unit of UDG for 10 minutes at 37°C renders the DNA incapable of being copied by DNA polymerase. The enzyme can be 95% heat killed by incubation at 95°C for 10 minutes. Since UDG remains partially active following heat treatment at 95°C, it is recommended that uracil glycosylase inhibitor be added to prevent degradation of product DNA. Alternatively, reaction products can be immediately extracted with phenol/chloroform.

**Reagents Supplied with Enzyme:**  
10X UDG Reaction Buffer.

**Reaction Conditions:** 1X UDG Reaction Buffer. Incubate at 37°C.

**1X UDG Reaction Buffer:**  
20 mM Tris-HCl  
1 mM EDTA  
1 mM dithiothreitol  
(pH 8.0 @ 25°C).

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(pH 8.0 @ 25°C).

**Unit Definition:** One unit is defined as the amount of enzyme that catalyzes the release of 60 pmol of uracil per minute from double-stranded, uracil-containing DNA. Activity is measured by release of [<sup>3</sup>H]-uracil in a 50 µl reaction containing 0.2 µg DNA (10<sup>4</sup>-10<sup>5</sup> cpm/µg) in 30 minutes at 37°C.

**Unit Assay Conditions:** 1X UDG Reaction Buffer, 1 unit of uracil DNA Glycosylase, 0.2 µg <sup>3</sup>H-uracil DNA (10<sup>4</sup>-10<sup>5</sup> cpm/µg) for 30 minutes at 37°C in a total reaction volume of 50 µl.

### Quality Control Assays

**16-Hour Incubation:** A 50 µl reaction containing 1 µg of λ phage DNA in NEBuffer 1 and 50 units of UDG showed no degradation following overnight incubation at 37°C.

**Exonuclease Activity:** Incubation of 50 units for 4 hours at 37°C in 50 µl of NEBuffer 1 with 1 µg <sup>3</sup>H DNA (10<sup>5</sup> cpm/µg) released < 0.1% radioactivity.

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**Endonuclease Activity:** Incubation of 50 units of UDG with 1 µg of φX174 RF I DNA for 4 hours at 37°C in 50 µl reaction buffer resulted in < 5% conversion to RF II.

**Physical Purity:** Purified to > 95% homogeneity as determined by SDS-PAGE analysis using Coomassie Blue detection. BSA is added to the enzyme for stability.

**Heat Inactivation:** No

**Notes On Use:** UDG is active over a broad pH range with an optimum at pH 8.0, does not require divalent cation, and is inhibited by high ionic strength (> 200 mM).

### References:

1. Lindahl, T. et al. (1977) *J. Biol. Chem.* 252, 3286-3294.
2. Wang, Z. et al. (1991) *Gene* 99, 31-37.
3. Devchand, P.R. et al. (1993) *Nucl. Acids Res.* 21, 3437-3443.

CERTIFICATE OF ANALYSIS

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