

TREVIGEN® Product Data

For Research Use Only. Not For Use In Diagnostic Procedures

T4 Endonuclease V (T4-Pyrimidine Dimer Glycosylase/T4-PDG)

Catalog #: 4055-100-EB
Contents: 4055-100-01 T4 Endonuclease V **Size:** 100,000 Units
3900-500-11 10X REC™ Buffer 11 1 ml

Description: T4 Endonuclease V is a DNA glycosylase of MW 16 kDa with an associated AP lyase activity that is specific for the cis-syn isomer of cyclobutane pyrimidine dimers induced by UV irradiation. This highly processive enzyme cleaves the glycosyl bond, 5' to the pyrimidine dimer, via nucleophilic attack of the alpha amino group of Thr-2, forming a series of protein-DNA imino intermediates.^{1,2} The associated AP lyase activity results in cleavage of the phosphodiester bond 3' to the AP site, thus generating a 3' unsaturated aldehyde and a 5' phosphate (Figure 1). This site is then incised on the 5' side by an AP endonuclease, generating an appropriate substrate for resynthesis and DNA repair.

Source: Purified from *E. coli* containing a recombinant plasmid harboring the T4 phage *denV* gene.

Unit Definition: One Unit is the amount of enzyme required to completely relax 250 ng of a UV-irradiated supercoiled plasmid in 30 minutes at 37°C.

Substrate Specificity: T4-PDG recognizes cis-syn cyclobutane pyrimidine dimers and AP sites. The enzyme recognizes trans-syn dimers at less than 1% the efficiency of the cis-syn isomer. T4-PDG also cleaves at 2,6-diamino-4-hydroxy-5-N-methylformamidoadenine (FAPYA) residues but at only 1-3% that of cis-syn cyclobutane pyrimidine dimers.

Assay Conditions & Analysis: 1X REC™ Buffer 11 (25 mM sodium phosphate pH 6.8, 1 mM EDTA, 100 mM NaCl, 1 mM DTT, 0.01% Triton X-100); 0.1 mg/ml BSA, supercoiled plasmid (250 ng) irradiated with 100 J/m² UV light, and serial dilutions of enzyme in a 20 µl reaction volume are incubated for 30 minutes at 37°C. For analysis, 5 µl of 5X REC Loading buffer (20 mM EDTA, 25% Ficoll, and 0.1% bromophenol blue) are added, and the supercoiled, linear, and open circle forms of the plasmid are resolved by 1% agarose gel electrophoresis. Bands are visualized by ethidium bromide staining.

Storage Buffer: 20 mM HEPES-NaOH pH 7.0, 0.5 M NaCl, 0.1 mM EDTA.

Storage Conditions: Store at 4°C. Do not freeze enzyme. May be diluted in 1X REC™ Buffer 11 and used immediately. To preclude loss of activity due to adsorption to plastic or glass surfaces, include BSA at 100 µg/ml in all buffers and assays.

References:

1. Vassilyev DG, Kashiwagi T, Mikami Y, Ariyoshi M, Iwai S, Ohtsuka E, Morikawa K (1995). Atomic model of a pyrimidine dimer excision repair enzyme complexed with a DNA substrate: structural basis for damaged DNA recognition. *Cell* **83**:773-82.
2. Golan G, Zharkow DO, Grollman AP, Dodson ML, McCullough AK, Lloyd RS, Shoham G (2006) Structure of T4 pyrimidine dimer glycosylase in a reduced imine covalent complex with abasic site-containing DNA. *J Mol Biol* **362**:241-58.

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TREVIGEN®

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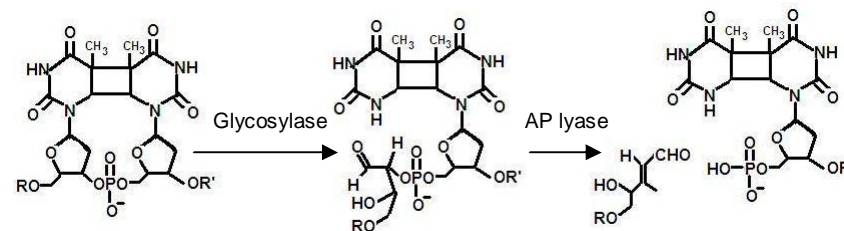


Figure 1. T4-PDG catalyzes the first step of DNA excision repair by employing two distinct activities, a pyrimidine dimer (PD)-specific glycosylase, which cleaves the glycosyl bond 5' of the PD, and an apyrimidinic (AP) lyase, which cleaves the phosphodiester bond 3' of the abasic site through β elimination to produce an α,β -unsaturated aldehyde and a 5' terminal phosphate.

Related Products:

DNA Repair Enzymes:

Catalog #	Description	Size
4150-010-EB	<i>Sulfolobus solfataricus</i> DNA Polymerase IV (Dpo4)	10 µg
4045-01K-EB	<i>E. coli</i> Endonuclease III	1000 U
4050-100-EB	<i>E. coli</i> Endonuclease I V	100 U
4035-500-EB	<i>E. coli</i> Endonuclease V	500 U
4060-01K-EB	<i>E. coli</i> Endonuclease VIII	1000 U
4030-05K-EB	<i>E. coli</i> Exonuclease III	5000 U
4130-100-EB	Human 8-oxo-G DNA Glycosylase (hOGG1)	100 U
4040-100-EB	<i>E. coli</i> Fpg	500 U
4025-100-EB	<i>E. coli</i> Uracil-N-Glycosylase	100 U
4000-500-EB	<i>E. coli</i> MutY DNA Glycosylase	500 U
4110-01K-EB	Human AP Endonuclease	1000 U
4020-01K-EB	Human β Polymerase	1000 U
4120-100-EB	Human FEN-1	100 U
4125-100-EB	<i>E. coli</i> Mismatch Uracil DNA Glycosylase (Mug)	100 U
4090-100-EB	Mouse 3-Methyladenine DNA Glycosylase (Aag)	100 U
4070-500-EB	Thermostable thymine mismatch DNA Glycosylase	500 U
4100-100-EB	<i>S. pombe</i> Ultraviolet DNA Endonuclease (UVDE)	100 µl
4065-100-EB	Chorella Virus Pyrimidine Dimer Glycosylase	1000 U
4145-100-EB	<i>E. coli</i> Photolyase	100 U

DNA damage characterization:

Catalog #	Description	Size
4055-100-FK	T4-PDG FLARE™ Kit	75 samples

Lot#:

Protein Concentration:

Activity:

T4 Endonuclease V

Catalog #:4055-100-EB

Storage: 4 °C

Do Not Freeze

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