

# TREVIGEN® Product Data

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## Thermostable TDG Protein (Thymine DNA Glycosylase)

**Catalog #:** 4070-500-EB  
**Contents:** 4070-500-01 Thermostable TDG Protein Size: 500 Units  
3900-500-4 10X REC™ Buffer 4 1 ml

**Description:** TDG is a thermostable thymine DNA glycosylase from *Methanobacterium thermoautotrophicum*. The optimal temperature for the enzyme is 65°C. The enzyme lacks significant AP lyase or endonuclease activity. TDG works effectively in heteroduplex analysis to detect C to T transitions.

**Source:** Thermostable TDG is purified from *E. coli* containing a recombinant plasmid harboring the *Methanobacterium thermoautotrophicum* TDG gene.

**Unit Definition:** One Unit is the amount of enzyme required to cleave 1 pmole of an oligonucleotide duplex containing a T/G mismatch in 1 hour at 65°C. Only the strand containing the T is cleaved.

**Substrate Specificity:** TDG enzyme recognizes T/G mismatches in duplex DNA and cleaves the strand with the T. The opposite strand is not cleaved. The enzyme also recognizes G/G mismatches if at least one nearest neighbor is an A or T and nicks one strand or the other. The enzyme exhibits poor AP lyase activity.

**Assay Conditions & Analysis:** 4 pmoles of T/A mismatch oligonucleotide set with the T oligo end-labeled, 1X REC™ Buffer 4 (10 mM HEPES-KOH (pH 7.4), 100 mM KCl, and 10 mM EDTA), and serial dilutions of enzyme in a 20 µl reaction volume are incubated for 1 hour at 65°C. To complete cleavage of abasic site, fresh 1N NaOH is added to final concentration of 166 mM then heated for 15 minutes at 95°C. For analysis, 24 µl of 2X Loading Buffer (20 mM EDTA, 95% formamide, and 0.13% bromophenol blue) are added, and the samples heated at 95°C for 10 min then fast cooled to 4°C. The cleavage products are resolved by 20% denaturing polyacrylamide gel electrophoresis, and percent cleavage quantified.

**Storage Buffer:** 20 mM Tris-Cl (pH 7.8), 100 mM NaCl, 1 mM EDTA, 1 mM DTT, 50% (v/v) glycerol.

**Storage Conditions:** Store at -20°C in a manual defrost freezer. For long term storage, freeze in working aliquots at -80°C. Avoid repeated freeze-thawing. Enzyme may be diluted in 1X REC Buffer 4 for immediate use. TDG protein in storage buffer can survive for up to 24 hours at 37°C with less than 10% loss in activity.

**References:** 1. Horst, J.P. and H.J. Fritz. 1996. Counteracting the mutagenic effect of hydrolytic deamination of DNA 5-methylcytosine residues at high temperature: DNA mismatch N-glycosylase Mig. Myth of the thermophilic archaeon *Methanobacterium thermoautotrophicum*. EMBO J 15:5459-5469.  
2. Begley, T.J., and R.P.C. Cunningham. 1999. *Methanobacterium thermoformicum* thymine DNA mismatch glycosylase; conversion of an N-glycosylase to an AP lyase. Protein Engineering 12:333-340.

## Related Products:

Catalog#	Description	Size
4020-100-EB	Human DNA Polymerase β	100 U
4025-100-EB	<i>E. coli</i> Uracil-N-Glycosylase (UNGase)	100 U
4040-100-EB	<i>E. coli</i> Formamidopyrimidine-DNA Glycosylase (Fpg)	500 U
4045-01K-EB	<i>E. coli</i> Endonuclease III (Thymine Glycol-DNA Glycosylase)	1000 U
4050-100-EB	<i>E. coli</i> Endonuclease IV (nfo protein)	100 U
4055-100-EB	T4 Endonuclease V (T4-Pyrimidine Dimer Glycosylase/T4-PDG)	10 <sup>5</sup> U
4060-01K-EB	<i>E. coli</i> Endonuclease VIII	1000 U
4065-100-EB	Chlorella Virus Pyrimidine Dimer Glycosylase (cv-PDG)	1000 U
4090-500-EB	Mouse 3-mA DNA Glycosylase (Aag protein)	500 U
4100-100-EB	<i>S. pombe</i> UVDE	100 µl
4110-01K-EB	Human Apurinic/Apyrimidinic Endonuclease (hAPE)	1000 U
4120-100-EB	Human FEN-1 (Flap Endonuclease)	100 U
4130-100-EB	Human 8-oxoGuanine DNA Glycosylase (hOGG1)	100 U
4135-250-01	Human Ku 70/80 Complex	250 U

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