For Research Use Only. Not For Use In Diagnostic Procedures

E. coli Formamidopyrimidine-DNA Glycosylase (Fpg)

Catalog #: 4040-500-EB

			Size:
Contents:	4040-100-01	Fpg	5 x 500 Units
	3900-500-10	10X REC [™] Buffer 10	1 ml

Description: Fpg releases damaged bases preferentially from duplex DNA. It has an associated class I AP lyase activity, leaving both 3' and 5' phosphoryl groups. This results from a β , δ elimination reaction at the AP sites, producing a single nucleotide gap in the DNA. The enzyme consists of 269 amino acids with a molecular weight of 30.2 kDa.

- - -

Source: Purified from *E. coli* containing a recombinant plasmid harboring the *E. coli* fpg gene.

Unit Definition: One Unit cleaves 1 pmole of a labeled oligonucleotide probe containing 8-oxoguanine, within an oligonucleotide duplex in one hour at 37° C.

Specificity: Fpg catalyzes the excision of the following forms of DNA damage:

- Open ring forms of 7-methylguanine, including 2,6-diamino-4-hydroxy-5-N-methylformamidopyrimidine and 4,6-diamino-5-amidopyrimidine, a lethal lesion.
- 2. 8-oxoguanine, a highly mutagenic lesion and probably the most important biological substrate of Fpg.
- 3. 5-hydroxycytosine
- 4. 5-hydroxyuracil
- 5. Aflatoxin-bound imidazole-ring-opened guanine
- 6. Imidazole ring opened N-2-aminofluorene-C8-guanine

Assay Conditions & Analysis: 1X REC Buffer 10 (10 mM HEPES-KOH (pH 7.4), 100 mM KCl, 10 mM EDTA, and 0.1 mg/ml BSA), 4 pmoles of labeled 8-oxo-dG oligonucleotide annealed to the compliment oligonucleotide, and serial dilutions of enzyme in a reaction volume of 20 μ l are incubated for 1 hour at 37 °C. For analysis, 20 μ l of 2X Loading Buffer (20 mM EDTA, 95% formamide, and 0.13% bromophenol blue) are added, the samples are heated to 95 °C for 10 min then fast cooled to 4 °C, and 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-thawings. May be diluted in 10 mM HEPES-KOH (pH 7.4), 100 mM KCl, 1 mM EDTA, 1 mM DTT, 0.1 mg/ml BSA, 50% glycerol and store at -20 °C for up to 1 week. Otherwise, dilute enzyme in 1X REC Buffer 10 and use immediately. It is stable for up to 8 hours at 37 °C without any loss in activity.

TREVIGEN® Product Data

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References:

- Tchou, J., V. Bodepudi, S. Shibutani, I. Antoshechkin, J. Miller, A.P. Grollman, and F. Johnson. 1994. Substrate specificity of Fpg protein: recognition and cleavage of oxidatively damaged DNA. J Biol Chem 269: 15318-15324.
- 2. Friedberg, E.C., G.C. Walker, and W. Siede. 1995. DNA Repair and Mutagenesis. American Society of Microbiology, Washington, D.C.: ASM Press.
- Boiteux, S., T.R. O'Connor, and J. Laval. 1987. Formamidopyrimidine-DNA glycosylase of Escherichia coli: cloning and sequencing of the Fpg structural gene and overproduction of the protein. EMBO J 6: 3177-3183.

DNA Repair Enzymes:

Catalog #	Description	Size
4150-010-EB	Sulfolobus silfataricus DNA Polymerase IV (Dpo4)	10 µg
4045-01K-EB	E. coli Endonuclease III	1000 U
4050-100-EB	<i>E. coli</i> Endonuclease IV	100 U
4065-100-EB	Chorella Virus Pyrimidine Dimer Glycosylase	1000 U
4130-100-EB	Human 8-oxo-G DNA Glycosylase (hOGG1)	100 U
4100-100-EB	S. pombe UVDE	100 µl
4025-100-EB	E. coli Uracil-N-Glycosylase	100 U
4000-500-EB	E. coli 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	E. coli 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
4055-100-EB	T4 Endonuclease V	100,000 U

References:

- Tchou, J., V. Bodepudi, S. Shibutani, I. Antoshechkin, J. Miller, A.P. Grollman, and F. Johnson. 1994. Substrate specificity of Fpg protein: recognition and cleavage of oxidatively damaged DNA. J Biol Chem 269: 15318-15324.
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4020-01K-EB	Human β Polymerase	1000 U
4120-100-EB	Human FEN-1	100 U
4125-100-EB	E. coli 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
4055-100-EB	T4 Endonuclease V	100,000 U

Unit Conc.: 4 U/µL Prot. Conc.: 4.65 mg/mL Spec. Activity: 860 units/mg *E. coli* Formamidopyrimidine DNA Glycosylase (Fpg) Catalog #: 4040-500-EB Lot #: 20935G10 TREVIGEN® 1-800-873-8443

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