

TREVIGEN® Product Data

For Research Use Only. Not For Use In Diagnostic Procedures.

E. coli Endonuclease IV (nfo protein)

Catalog #: 4050-100-EB

Contents: 4050-100-01 Endonuclease IV
3900-500-01 10X REC™ Buffer 1

Size: 100 Units
1 ml

Description: Endonuclease IV is a class II AP endonuclease with no associated N-glycosylase activity (see reverse).

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

Unit Definition: One Unit is the amount of enzyme required to cleave an AP-site oligonucleotide within an oligonucleotide duplex at the rate of 1 pmol/hour at 37°C.

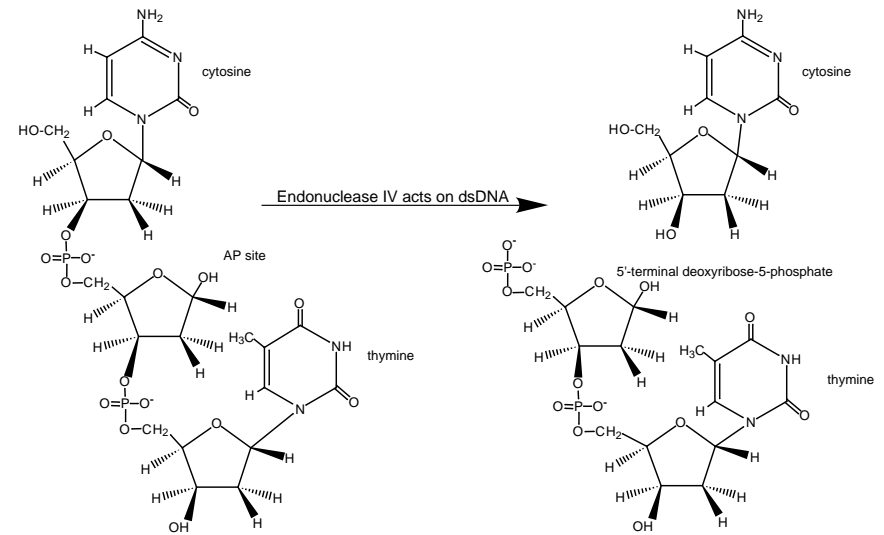
Specificity: Endonuclease IV removes phosphoglycoaldehyde, deoxyribose-5-phosphate, 4-hydroxy-2-pentanal, and phosphate groups from the 3' ends of DNA. It is not stimulated by cofactors such as Mg²⁺ or Ca²⁺, but is inhibited by EDTA (suggesting a metal ion cofactor). The preparation is free of exonuclease activity.

Assay Conditions: 1X REC Buffer 1 (10 mM HEPES-KOH, pH 7.4, 100 mM KCl), 4 pmole AP-Site Oligonucleotide (Cat# 3851-100-01) labeled with ³²P, 4 pmole Oligo Complement B (Cat# 3849-100-02), and serial dilutions of enzyme in a 20 µl reaction volume are incubated for 1 hour at 37°C. For analysis, 5 µl of 5X REC Loading Buffer (Cat# 4018-250:20 mM EDTA, 20% Ficoll, and 0.2% bromophenol blue) are added, and the cleavage products are resolved by 20% denaturing polyacrylamide gel electrophoresis. The bands are cut out and radioactivity counted to quantify cleavage products.

Storage Buffer: 10 mM HEPES-KOH, pH 7.4, 100 mM KCl, 0.1 mg/ml BSA, and 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.

- References:**
1. Doetsch, P.W. and R.P. Cunningham. 1990. The enzymology of apurinic/aprimidinic endonucleases. *Mutat Res* **236**:173-201.
 2. Cunningham, R.P., S.M. Saporito, S.G. Spitzer, and B. Weiss. 1986. Endonuclease IV (nfo) mutant of *Escherichia coli*. *J Bacteriol.* **168**:1120-27.
 3. Friedberg, E.C., G.C. Walker, and W. Siede. 1995 in *DNA Repair and Mutagenesis*, American Society of Microbiology, Washington, D.C.: ASM Press.
 4. Levin, J.D., R. Shapiro, and B. Demple. 1991. Metalloenzymes in DNA repair: *Escherichia coli* endonuclease IV and *Saccharomyces cerevisiae*. *J Biol Chem* **266**:22893-898



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