Endonuclease V (*E. coli*)

Catalogue number: MB21301, 250 U

Description

Endonuclease V is a DNA repair enzyme from *Escherichia coli*, expressed in the recombinant form in the same host, which recognizes deaminated bases. Specifically, the enzyme recognizes deoxyinosines in DNA strands (double or single-stranded), obtained by deamination of deoxyadenosines. Endonuclease V also recognizes DNA containing abasic sites or urea sites, base mismatches, harpins or loops. Endonuclease V, also termed deoxyinosine 3´ endonuclease, catalyses the cleavage of the second phosphodiester bond 3´ to the mismatch of deoxyinosine, leaving a nick with 3´-OH and 5´-phosphate.
The enzyme has a variety of applications, including: cleavage of oligonucleotides containing deoxyinosines and base mismatches cleavage. Endonuclease V has also affinity for oligonucleotides containing base mismatches.

Storage conditions

Endonuclease V should be stored at -20 °C in a constant temperature freezer. The protein will remain stable till the expiry date if stored as specified.

Unit definition

One unit is defined as the amount of enzyme required to cleave 1 pmol of a 34-mer oligonucleotide duplex containing a single deoxyinosine site in a reaction volume of 10 µL in 1 hour at 37 °C.

Enzyme concentration: 10 U/µL

Inactivation

Endonuclease V is heat inactivated at 65 °C for 20 min.

System components and Reaction conditions

Endonuclease V is provided with a dedicated and highly optimized NZYtech reaction buffer and displays an optimum temperature of 37 °C.

Standard Protocol

The following standard protocol serves as a general guideline for the cleavage of DNA using Endonuclease V.

1. Combine 1 µg of the nucleic acid with 10–15 units of Endonuclease V in 1x reaction buffer provided.

2. Incubate at 37 °C for 4 hours.

We recommend incubating 1 µg of the nucleic acid with 10–15 units of the enzyme in 1× reaction buffer for 4 h at 37 °C.

Quality control assays

Purity

Recombinant Endonuclease V is >95% pure as judged by SDS polyacrylamide gel electrophoresis followed by BlueSafe staining (MB15201).

Functional assay

Endonuclease V is tested for activity in a nicking reaction using 0.2-0.3 µg of pNZY28 plasmid DNA. The reaction performs in 1× Reaction buffer for 1 hour at 37 °C. Following incubation, the DNA is visualized on a GreenSafe Premium (MB132)-stained agarose gel. Nicking DNA must be visible.

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For research use only.

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