

Methyl-directed DNA
Endonuclease



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Mte I

E554



2500 u

10 000 u/ml

Lot: 1

Store at -20°C

Recognition sequence

5'-G(5mC)G(5mC)^NG(5mC)G(5mC)-3'
3'-(5mC)G(5mC)GN^(5mC)G(5mC)G-5'

Source: *Microbacterium testaceum 17B*

Substrate specificity:

The enzyme cleaves C5-methylated DNA and doesn't cut unmodified DNA.

DNA pHspAI10/
Dril+M.Fsp4HI

Supplied in:

10 mM Tris-HCl (pH 7.5); 200 mM NaCl; 0,1 mM EDTA;
7 mM 2-mercaptoethanol; 200 µg/ml BSA; 50% glycerol.

Reaction Conditions:

1×SEBuffer W. Incubate at 55°C.

1×SEBuffer W (pH 8.5 @ 25°C)

Warranty period for the enzyme storage at-20°C is two years from the date of the last assay indicated on the enzyme vial.

10 mM Tris-HCl 150 mM NaCl
10 mM MgCl₂; 1 mM DTT

Unit Definition: One unit is defined as the amount of enzyme required to hydrolyze completely 1 µg of linearized plasmid pHspAI10/Dril+M.Fsp4HI in 1 hour at 55°C in a total reaction volume of 50 µl.

pHspAI10/Dril+M.Fsp4HI is a plasmid pHspAI10, which is linearized with Dril, and, additionally, modified with Fsp4HI DNA methyltransferase pHspAI10 carries a gene of HspAI DNA methyltransferase, that modifies the sequence . 5'-GCGC-3', producing 5'-G(5mC)GC-3'. M.Fsp4HI modifies the sequence 5'-GCNGC-3', producing 5'-G(5mC)NGC-3'. A substrate pHspAI10/Dril+M.Fsp4HI includes one site 5'-G(5mC)G(5mC)NG(5mC)G(5mC)-3'/3'-(5mC)G(5mC)GN(5mC)G(5mC)G-5', which is Mtel canonical site [1]. The enzyme activity depends on a number and positions of methylated nucleotides in the recognition sequence. For example, Mtel cuts the recognition site with six 5-methylcytosines, but the enzyme activity is reduced for more that one order [1].

Warranty period for the enzyme storage at-20°C is two years from the date of the last assay indicated on the enzyme vial.

Enzyme Properties

Activity in SEBuffers:

SEBuffer B 25-50%
SEBuffer G 75-100%
SEBuffer O 75-100%
SEBuffer W 100%
SEBuffer Y 50-75%
SEBuffer ROSE 100%

When using a buffer other than the optimal (supplied) SEBuffer, it may be necessary to add more enzyme to achieve a complete digestion.

Quality Control Assays

16-Hour Incubation:

No detectable degradation of 1µg of Lambda DNA was observed after incubation with 10 units of enzyme for 16 hours at 55°C in a total reaction volume of 50 µl.

Oligonucleotide Assay:

No detectable degradation of a single- and double-stranded oligonucleotide was observed after incubation with 10 units of enzyme for 3 hours.

Reagents Supplied with Enzyme: 10×SEBuffer W

Heat Inactivation: No (80°C for 20 minutes)

References:

1. *V.A. Chernukhin, E.V. Kileva, V.A. Sokolova., D.A. Gonchar, L.N. Golikova, V.S. Dedkov, N.A. Mikhnenkova, S.Kh. Degtyarev*
A new methyl-directed site-specific DNA endonuclease Mtel cleaves nine nucleotides sequence
5'-G(5mC)G(5mC)^NG(5mC)GC-3'/3'-CG(5mC)GN^(5mC)G(5mC)G-5'
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CERTIFICATE OF ANALYSIS