DNA Methyltransferase M.BseCI

5’ ···ATCGAT···3’

Description: M.BseCI modifies the N6 atom of the 3′ adenine residue in the sequence 5′-ATCGAT-3′.

Catalogue No	*204-1, 1000 U* *204-2, 3x1000 U*
Concentration	*5u/µl*

Reagents supplied: 10x M.BseCI buffer

Source: M.BseCI is a methyltransferase purified from an *E. coli* strain that carries the BseCI methyltransferase gene (bseCIM) from Bacillus stearothermophilus, cloned in plasmid pBseCIM8 (1,2).

Reaction Buffer: 10 mM Tris-HCl (pH 7.4), 10 mM EDTA, 5 mM 2-mercaptoethanol, 0.02% Triton-X-100.

Unit definition: One unit is defined as the amount of enzyme required to protect 1µg of λ DNA in 1 hour at 55^{o}C in a total reaction volume of 10µl against cleavage by BseCI restriction endonuclease.

Protection Assay Conditions: M.BseCI is incubated with 1µg of λ DNA in 10µl 1x M.BseCI buffer, supplemented with 80µM S-adenosylmethionine (SAM), for 1 hour at 55^{o}C followed by 15 minutes at 70^{o}C. The extent of protection by M.BseCI is determined by the addition of 40µl BseCI Reaction Buffer and 10 units of BseCI restriction endonuclease. Incubation at 55^{o}C for 30 minutes is followed by analysis on agarose gel.

Note: M.BseCI exhibits 35% activity at 37^{o}C.

Storage buffer: 50 mM Tris-HCl (pH 7.4), 10 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol. Store at -20^{o}C.

Quality Control: Tested for the absence of endo- and exodeoxyribonucleases.

References: 1. Rina, M. and Bouriotis, V. (1993) Cloning, purification and characterization of the BseCI DNA methyltransferase from Bacillus stearothermophilus. Gene 133, 91-94.

2. Rina, M., Markaki, M. and Bouriotis, V. (1994) Sequence of the cloned bseCIM gene: M.BseCI reveals high homology to M.BanIII Gene 150, 71-73.