**M0217S**

1,000 units 25,000 U/ml Lot: 0061311

Methyltransferase

**Description:** HhaI Methyltransferase modifies the internal cytosine residue (C') in the sequence above.

**Source:** An *E. coli* strain that carries the cloned HhaI modification gene from *Haemophilus haemolyticus* (ATCC 10014)

Supplied in: 150 mM NaCl, 50 mM Tris-Cl (pH 7.5), 10 mM EDTA, 5 mM 2-mercaptoethanol, 200 µg/ml BSA and 50% glycerol.

**Reagents Supplied with Enzyme:**
- 10X HhaI Methyltransferase Reaction Buffer, 400X S-adenosylmethionine (32 mM).
- 10X HhaI Methyltransferase Reaction Buffer, 80 µM S-adenosylmethionine. Incubate at 37°C.

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The extent of protection by HhaI Methyltransferase is determined by the addition of 40 µl NEBuffer 4 supplemented with 100 µg/ml BSA, 10 mM MgCl₂ and 10 units of HhaI restriction endonuclease. Incubation at 37°C for 30 minutes is followed by analysis on an agarose gel.

**Unit Definition:** One unit is defined as the amount of enzyme required to protect 1 µg λ DNA in 1 hour at 37°C in a total reaction volume of 10 µl against cleavage by HhaI restriction endonuclease.

**Quality Control Assays**

**16-Hour Incubation:** Incubation of 125 units of HhaI Methyltransferase with 1 µg of HindIII-digested DNA in 50 µl 1X NEBuffer 2 for 16 hours at 37°C resulted in no detectable endonuclease contamination.

**Exonuclease Activity:** Incubation of 250 units of HhaI Methyltransferase with 1 µg sonicated λ DNA (10^5 cpm/µg) for 4 hours at 37°C in 50 µl NEBuffer 2 [50 mM NaCl, 10 mM Tris-Cl (pH 7.9 @ 25°C), 10 mM MgCl₂, 1 mM DTT] released 0.39% of the total radioactivity.

**Storage of SAM:** S-adenosylmethionine or SAM is stored at –20°C as a 32 mM solution dissolved in 0.005 M sulfuric acid and 10% ethanol. Under these conditions SAM is stable for up to 6 months. SAM is unstable at (pH 7.5), 37°C, (1) and should be replenished in reactions incubated longer than 4 hours.

Methylation can be optimized by using fresh SAM.

**Reference:**

**Companion Product:**
S-adenosylmethionine (SAM) #90003S 0.5 ml