

BamHI Methyltransferase



1-800-632-7799
info@neb.com
www.neb.com



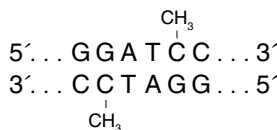
M0223S 008151016101

M0223S



100 units **4,000 U/ml** **Lot: 0081510**
RECOMBINANT Store at **-20°C** Exp: **10/16**

Methylation Site:



Description: BamHI Methyltransferase modifies the internal cytosine residue (N⁴) in the sequence above.

Source: An *E. coli* strain that carries the cloned BamHI modification gene from *Bacillus amyloliquefaciens* H (ATCC 49763)

Supplied in: 50 mM Tris-HCl (pH 7.5), 10 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:

10X BamHI Methyltransferase Reaction Buffer,
400X S-adenosylmethionine (32 mM).

Reaction Conditions: 1X BamHI Methyltransferase Reaction Buffer, 80 µM S-adenosylmethionine. Incubate at 37°C.

1X BamHI Methyltransferase Reaction Buffer:

50 mM Tris-HCl
10 mM EDTA
5 mM DTT
pH 7.5 @ 25°C

Protection Assay Conditions: BamHI Methyltransferase is incubated with 1 µg λ DNA in 10 µl 1X BamHI Methyltransferase Reaction Buffer, supplemented with 80 µM S-adenosylmethionine, for one hour at 37°C

Source: An *E. coli* strain that carries the cloned BamHI modification gene from *Bacillus amyloliquefaciens* H (ATCC 49763)

Supplied in: 50 mM Tris-HCl (pH 7.5), 10 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:

10X BamHI Methyltransferase Reaction Buffer,
400X S-adenosylmethionine (32 mM).

Reaction Conditions: 1X BamHI Methyltransferase Reaction Buffer, 80 µM S-adenosylmethionine. Incubate at 37°C.

1X BamHI Methyltransferase Reaction Buffer:

50 mM Tris-HCl
10 mM EDTA
5 mM DTT
pH 7.5 @ 25°C

Protection Assay Conditions: BamHI Methyltransferase is incubated with 1 µg λ DNA in 10 µl 1X BamHI Methyltransferase Reaction Buffer, supplemented with 80 µM S-adenosylmethionine, for one hour at 37°C

followed by 15 minutes at 65°C. The extent of protection by BamHI Methyltransferase is determined by the addition of 40 µl NEBuffer 1 supplemented with 10 mM MgCl₂ and 10 units of BamHI restriction endonuclease. Incubation at 37°C for 30 minutes is followed by analysis on agarose gels.

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 BamHI restriction endonuclease.

Quality Control Assays

Exonuclease Activity: Incubation of 30 units of BamHI Methyltransferase with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl NEBuffer 2 [50 mM NaCl, 10 mM Tris-HCl (pH 7.9 @ 25°C), 10 mM MgCl₂, 1 mM DTT] released < 0.3% of the total radioactivity.

Endonuclease Activity: Incubation of 15 units with 1 µg of λ HindIII DNA fragments in a 50 µl NEBuffer 2 for 16 hours at 37°C resulted in no degradation of the DNA as determined by

visualization of clear and sharp bands following gel electrophoresis.

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 BamHI restriction endonuclease.

Quality Control Assays

Exonuclease Activity: Incubation of 30 units of BamHI Methyltransferase with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl NEBuffer 2 [50 mM NaCl, 10 mM Tris-HCl (pH 7.9 @ 25°C), 10 mM MgCl₂, 1 mM DTT] released < 0.3% of the total radioactivity.

Endonuclease Activity: Incubation of 15 units with 1 µg of λ HindIII DNA fragments in a 50 µl NEBuffer 2 for 16 hours at 37°C resulted in no degradation of the DNA as determined by

visualization of clear and sharp bands following gel electrophoresis.

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:

- Hoffman, J. L. (1986) *Biochemistry* 25, 4444-4449.

Companion Product:

S-adenosylmethionine (SAM)
#B9003S 0.5 ml

CERTIFICATE OF ANALYSIS

BamHI Methyltransferase



1-800-632-7799
info@neb.com
www.neb.com



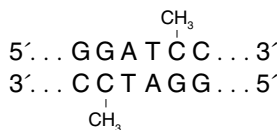
M0223S 008151016101

M0223S



100 units **4,000 U/ml** **Lot: 0081510**
RECOMBINANT Store at **-20°C** Exp: **10/16**

Methylation Site:



Description: BamHI Methyltransferase modifies the internal cytosine residue (N⁴) in the sequence above.

Source: An *E. coli* strain that carries the cloned BamHI modification gene from *Bacillus amyloliquefaciens* H (ATCC 49763)

Supplied in: 50 mM Tris-HCl (pH 7.5), 10 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:

10X BamHI Methyltransferase Reaction Buffer,
400X S-adenosylmethionine (32 mM).

Reaction Conditions: 1X BamHI Methyltransferase Reaction Buffer, 80 µM S-adenosylmethionine. Incubate at 37°C.

1X BamHI Methyltransferase Reaction Buffer:

50 mM Tris-HCl
10 mM EDTA
5 mM DTT
pH 7.5 @ 25°C

Protection Assay Conditions: BamHI Methyltransferase is incubated with 1 µg λ DNA in 10 µl 1X BamHI Methyltransferase Reaction Buffer, supplemented with 80 µM S-adenosylmethionine, for one hour at 37°C

visualization of clear and sharp bands following gel electrophoresis.

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 BamHI restriction endonuclease.

Quality Control Assays

Exonuclease Activity: Incubation of 30 units of BamHI Methyltransferase with 1 µg sonicated ³H DNA (10⁵ cpm/µg) for 4 hours at 37°C in 50 µl NEBuffer 2 [50 mM NaCl, 10 mM Tris-HCl (pH 7.9 @ 25°C), 10 mM MgCl₂, 1 mM DTT] released < 0.3% of the total radioactivity.

Endonuclease Activity: Incubation of 15 units with 1 µg of λ HindIII DNA fragments in a 50 µl NEBuffer 2 for 16 hours at 37°C resulted in no degradation of the DNA as determined by

visualization of clear and sharp bands following gel electrophoresis.

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:

- Hoffman, J. L. (1986) *Biochemistry* 25, 4444-4449.

Companion Product:

S-adenosylmethionine (SAM)
#B9003S 0.5 ml

CERTIFICATE OF ANALYSIS



NEW ENGLAND BIOLABS® is a registered trademark of New England Biolabs, Inc.

This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.



NEW ENGLAND BIOLABS® is a registered trademark of New England Biolabs, Inc.

This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.