Hpy188I

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

R0617S 003121114111

R0617S

NEB4 37° dam Yes

1,000 units **10,000 U/ml** **Lot: 0031211**
RECOMBINANT **Store at –20°C** **Exp: 11/14**

Recognition Site:

5′... TCN▼GA ...3′
3′... AG▲NCT ...5′

Source: An *E. coli* strain that carries the cloned Hpy188I gene from *Helicobacter pylori* 188 (S.A. Thompson)

New Storage Conditions

Supplied in: 100 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:
10X NEBuffer 4.

Reaction Conditions: 1X NEBuffer 4.
Incubate at 37°C.

1X NEBuffer 4:
50 mM potassium acetate
20 mM Tris-acetate
10 mM magnesium acetate
1 mM dithiothreitol
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of pBR322 in 1 hour at 37°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer A
50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 5-fold overdigestion with Hpy188I, approximately 50% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5′ termini concentration of 1–2 µM) at 16°C. Of these ligated fragments, approximately 75% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 10 units of enzyme incubated for 16 hours resulted in the same pattern of DNA bands as a reaction incubated for 1 hour with 1 unit of enzyme. Incubations of more than 10 units for 16 hours may result in star activity.

Exonuclease Activity: Incubation of 30 units of enzyme with 1 µg sonicated ^{3}H DNA (10^{5} cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	50%
NEBuffer 2	25%
NEBuffer 3	10%
NEBuffer 4	**100%**

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

Survival in a Reaction: A minimum of 1.0 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 80 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Notes: Hpy188I produces DNA fragments that have a single-base 3′ extension which are difficult to ligate.

Blocked by overlapping *dam* methylation.

Companion Products:
dam^{-}/dcm^{-} Competent *E. coli*

#C2925H	20 transformation reactions
#C2925I	24 transformation reactions

= Time-Saver™ Qualified (See www.neb.com for details).

U.S. Patent No. 6,258,583

CERTIFICATE OF ANALYSIS

Hpy188I

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

R0617S 003121114111

R0617S

NEB4 37° dam Yes

1,000 units 10,000 U/ml Lot: 0031211
RECOMBINANT Store at –20°C Exp: 11/14

Recognition Site:

5′... TCN▼GA ...3′
3′... AG▲NCT ...5′

Source: An *E. coli* strain that carries the cloned Hpy188I gene from *Helicobacter pylori* 188 (S.A. Thompson)

New Storage Conditions

Supplied in: 100 mM NaCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme:
10X NEBuffer 4.

Reaction Conditions: 1X NEBuffer 4.
Incubate at 37°C.

1X NEBuffer 4:
50 mM potassium acetate
20 mM Tris-acetate
10 mM magnesium acetate
1 mM dithiothreitol
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of pBR322 in 1 hour at 37°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer A
50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

Ligation: After 5-fold overdigestion with Hpy188I, approximately 50% of the DNA fragments can be ligated with T4 DNA Ligase (at a 5′ termini concentration of 1–2 µM) at 16°C. Of these ligated fragments, approximately 75% can be recut.

16-Hour Incubation: A 50 µl reaction containing 1 µg of DNA and 10 units of enzyme incubated for 16 hours resulted in the same pattern of DNA bands as a reaction incubated for 1 hour with 1 unit of enzyme. Incubations of more than 10 units for 16 hours may result in star activity.

Exonuclease Activity: Incubation of 30 units of enzyme with 1 µg sonicated ^{3}H DNA (10^{5} cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	50%
NEBuffer 2	25%
NEBuffer 3	10%
NEBuffer 4	100%

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

Survival in a Reaction: A minimum of 1.0 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 80 units of enzyme were inactivated by incubation at 65°C for 20 minutes.

Notes: Hpy188I produces DNA fragments that have a single-base 3′ extension which are difficult to ligate.

Blocked by overlapping *dam* methylation.

Companion Products:
dam^{-}/dcm^{-} Competent *E. coli*

#C2925H	20 transformation reactions
#C2925I	24 transformation reactions

= Time-Saver™ Qualified (See www.neb.com for details).

U.S. Patent No. 6,258,583

CERTIFICATE OF ANALYSIS