Restriction Endonuclease BamH I

From Bacillus amyloliquefaciens H

Cat. No. 10 220 612 001 1000 units (10 U/μl)
Cat. No. 10 567 604 001 2500 units (10 U/μl)
Cat. No. 10 656 275 001 10 000 units (10 U/μl)
Cat. No. 10 798 975 001 10 000 units, high concentration (40 U/μl)
Cat. No. 11 274 031 001 50 000 units, high concentration (40 U/μl)

Stability/Storage

The undiluted enzyme solution is stable when stored at −15 to −26°C until the control date printed on the label. Do not store below -25°C to avoid freezing.

Sequence specificity

BamH I recognizes the sequence G/GATCC and generates fragments with 5'-cohesive termini (1), which contain the same tetra-nucleotide sequence GATC as the cohesive termini of the fragments of Bcl I, Bgl II, Mbo I, Sau 3A or Xho II (2). BamH I exhibits star activity under non-optimal conditions.

Compatible ends

BamHI generates compatible ends to Bcl I, Bgl II, Nde II (Mbo I), Sau 3A and Xho II.

Enzyme that can cut this new sequence

BamH I - Enzyme

BamH I + Bat I

Bcl I

T/GATCA

G/GATCC

T/GATCC

Bgl II

A/GATCT

G/GATCC

A/GATCC

Nde II

/G*AT°C

G/GATCC

/G*AT°C

Sau 3A

/G*AT°C

G/GATCC

/G*AT°C

Xho II

Pu/GATCPy

G/GATCC

Pu/GATCC

Isoschizomers

The BamH I is an isoschizomer of Bat I.

Methylation sensitivity

BamH I is not inhibited by overlapping dam-methylation but is inhibited by the presence of 5- or 4-methylcytosine at the internal C residue as indicated (*).

Storage buffer

10 mM Tris-HCl, 300 mM KCl, 1 mM EDTA, 1 mM DTE, 0.01% (v/v) Polydocanol, 50% Glycerol (v/v), pH approx. 7.4 (4°C).

Supplied Incubation buffer (10x)

100 mM Tris-HCl, 1 M NaCl, 50 mM MgCl₂, 10 mM 2-Mercaptoethanol; pH 8.0 (at 37°C), (5 × SuRE/Cut Buffer B).

Activity in SuRE/Cut Buffer System

Bold face printed buffer indicates the recommended buffer for optimal activity:

A

100%

B

100%

L

75-100%

M

100%

H

25-50%

Incubation temp.

37°C

Unit definition

One unit is the enzyme activity that completely cleaves 1 μg λDNA in 1 h at 37°C in a total volume of 50 μl SuRE/Cut buffer B. 1 μg pBR322 DNA is digested completely by ca. 2 units of Bam HI on account of the larger number of cleavage sites per μg pBR322 DNA as compared to λDNA.

Typical experiment

<table>
<thead>
<tr>
<th>Component</th>
<th>Final concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>DNA</td>
<td>1 μg</td>
</tr>
<tr>
<td>10 × SuRE/Cut Buffer B</td>
<td>5.0 μl</td>
</tr>
<tr>
<td>Nuclease-free water</td>
<td>Up to total volume of 50 μl</td>
</tr>
<tr>
<td>Restriction enzyme</td>
<td>1 unit</td>
</tr>
</tbody>
</table>

Incubate at 37°C for 1 h.

Heat inactivation

Up to 10 Units BamH I / μg DNA can be heat-inactivated by 15 min incubation at 65°C, higher enzyme concentrations can no more be completely inactivated under these conditions.

Number of cleavage sites on different DNAs (2):

Activity in PCR buffer

Relative activity in PCR mix (Taq DNA Polymerase buffer) is 100%. The PCR mix contained a target DNA, 10 mM Tris-HCl (pH 8.3, 20°C), 50 mM KCl, 1.5 mM MgCl₂, 200 μM dNTPs, 2.5 U Taq DNA polymerase. The mix was subjected to 25 amplification cycles.

Ligation and recutting assay

BamH I fragments obtained by complete digestion of 1 μg λDNA are ligated with 1 unit T4-DNA ligase in a volume of 10 μl by incubation for 16 h at 4°C in 66 mM Tris-HCl, 5 mM MgCl₂, 5 mM Dithioerythritol, 1 mM ATP, pH 7.5 (20°C) resulting in >95% recovery of 1 μg λDNA fragments. Subsequent re-cutting with BamH I yields >95% of the typical pattern of λDNA + BamH I fragments.

Troubleshooting

A critical component is the DNA substrate. Many compounds used in the isolation of DNA such as phenol, chloroform, ethanol, SDS, high levels of NaCl metal ions (e.g., Hg²⁺, Mn²⁺) inhibit or alter recognition specificity of many restriction enzymes. Such compounds should be removed by ethanol precipitation followed by drying, before the DNA is added to the restriction digest reaction. Appropriate mixing of the enzyme is recommended.

Star activity

BamH I exhibits star activity under non-optimal conditions. The relaxed specificities of BamH I are GGATCN or G(R)ATCC. The relaxed specificity of BamH I can be induced by lowering the ionic conditions of the RE buffer, by increasing the glycerol conc. to >5% or by using excess enzyme.

Quality control

Lot-specific certificates of analysis are available at www.roche-applied-science.com/certificates.

1 μg λDNA is incubated for 16 h in 50 μl incubation buffer with excess of BamH I. The number of enzyme units which do not change the enzyme-specific pattern is stated in the certificate of analysis.

www.roche-applied-science.com
## References

5. Rebase The Restriction Enzyme Database: [http://rebase.neb.com](http://rebase.neb.com)

## Printed Materials

You can view the following manuals on our website:

- Lab FAQs “Find a Quick Solution”
- Restriction Enzyme Ordering Guide
- Molecular Weight Markers for Nucleic Acids

## Changes to previous version

Update of quality control.

## Trademarks

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## Regulatory

Disclaimer

For life science research only. Not for use in diagnostic procedures.

## Commonly used bacterial strains

<table>
<thead>
<tr>
<th>Strain</th>
<th>Genotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>BL21</td>
<td>E. coli F' dcm ompT hsdS (rK- m- g~) lacIq Yf- proAB Tn7* (supE44 thi-1 lacY1) F'(lacIq lacZ58 Tn7*)</td>
</tr>
</tbody>
</table>
| C600r  | supE44 thi-1 lacY1 recA1 supF1 galK2 rpsL22 xyl-s5 mitA1 lacI 
| DH5x  | supE44 ΔlacU169 (rpsLZacZ5815) hsdR17 recA1 endA1 gyrA86 thi-1 relA1 |
| HB101  | recA1 supE44 endA1 hsdR17 gyrA86 relA1 thi-1 Δlac-proAB; (Yanisch- Perron, C. et al., (1985) Gene **33**, 103.) |
| JM110  | rpsL (55') thi leu thi-lac galK1galK1 araA1 tsoE dam cmc supE44 Δlac-proAB F'Tn3D3proAB{lacIq lacZ5815}; (Yanisch- Perron, C. et al., (1985) Gene **33**, 103.) |
| SURE2  | recB recJ sbc C201 urwCmumuC-Tn5(karF) lacI ΔΔ(hsdRMS) endA1 gyrA86 thi-1lacI supE44 F'proAB+ lacIq lacZ5815 Trn10 (tet') | (Greener, A. (1990) Strategies, **3**, 5. |
| TG1  | supE hsdR lacI ΔΔ(lac-proAB) F'Tn3D3proAB+ lacIq lacZ5815; (Gibson, T.J. (1984) Phd Theses, Cambridge University, U.K.) |
| XL1-Blue  | supE44 hsdR17 recA1 endA1 gyrA86 thi-1relA1 lacI+ F'proAB+ lacIq lacZ5815 Trn10 (tet') | (Bullock et al., (1987) BioTechniques, **5**, 376.) |

## Contact and Support

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support Site** at:

[www.roche-applied-science.com/support](http://www.roche-applied-science.com/support)

To call, write, fax, or email us, visit the Roche Applied Science home page, [www.roche-applied-science.com](http://www.roche-applied-science.com), and select your home country. Country-specific contact information will be displayed. Use the Product Search function to find Pack Inserts and Material Safety Data Sheets.

## Ordering Information

Roche Applied Science offers a large selection of reagents and systems for life science research. For a complete overview of related products and information, as well as links to detailed information (e.g. package inserts) of the selected restriction enzyme.

<table>
<thead>
<tr>
<th>Product</th>
<th>Application</th>
<th>Packsize</th>
<th>Cat. No.</th>
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</thead>
<tbody>
<tr>
<td>Restriction Enzymes</td>
<td>DNA restriction digestion</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rapid DNA Ligation Kit</td>
<td>Ligation of sticky- or blunt-ended DNA fragments in 5 min at 15 - 25 °C</td>
<td>Kit (40 DNA ligations)</td>
<td>11 635 379 001</td>
</tr>
<tr>
<td>T4 DNA Ligase</td>
<td>Ligation of sticky- and blunt-ended DNA fragments</td>
<td>100 U 500 units (1 U/μg)</td>
<td>10 481 220 001</td>
</tr>
<tr>
<td>T4 ADP Phosphatase</td>
<td>Dephosphorylation of 5’-phosphate residues from nucleic acids</td>
<td>1000 U 5000 U</td>
<td>04 898 133 001</td>
</tr>
<tr>
<td>T4 ADP Dephospho and Ligation Kit</td>
<td>Dephosphorylation of nucleic acids</td>
<td>40 reactions 160 reactions</td>
<td>04 898 117 001</td>
</tr>
<tr>
<td>Alkaline Phosphatase (AP), special quality for molecular biology</td>
<td>Dephosphorylation of 5’-phosphate residues from nucleic acids</td>
<td>1000 U (20 U/μg)</td>
<td>11 097 075 001</td>
</tr>
<tr>
<td>Agarose MP</td>
<td>Multipurpose agarose for analytical and preparative electrophoresis of nucleic acids</td>
<td>100 g 500 g</td>
<td>11 388 963 001</td>
</tr>
<tr>
<td>Agarose LE</td>
<td>Separation of nucleic acids in the range 0.2 - 1.5 kbp</td>
<td>100 g 500 g</td>
<td>11 388 691 001</td>
</tr>
<tr>
<td>Agarose Gel DNA Extraction Kit</td>
<td>For the elution of DNA fragments from agarose gels</td>
<td>1 Kit (max. 100 reactions)</td>
<td>11 696 505 001</td>
</tr>
<tr>
<td>High Pure PCR Product Purification Kit</td>
<td>Purification of PCR or enzymatic modification reaction (e.g. restriction digestion)</td>
<td>50 purifications 250 purifications</td>
<td>11 732 668 001</td>
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<tr>
<td>Sure/Cut Buffer Set for Restriction Enzymes</td>
<td>Incubation buffers A, B, L, M and H for restriction enzymes</td>
<td>1 ml each (10× conc. solutions)</td>
<td>11 082 035 001</td>
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<tr>
<td>Sure/Cut Buffer A</td>
<td>Restriction enzyme incubation</td>
<td>5 × 1 ml (10× conc. solution)</td>
<td>11 417 959 001</td>
</tr>
<tr>
<td>Sure/Cut Buffer B</td>
<td>Restriction enzyme incubation</td>
<td>5 × 1 ml (10× conc. solution)</td>
<td>11 417 967 001</td>
</tr>
<tr>
<td>Sure/Cut Buffer H</td>
<td>Restriction enzyme incubation</td>
<td>5 × 1 ml (10× conc. solution)</td>
<td>11 417 981 001</td>
</tr>
<tr>
<td>Sure/Cut Buffer L</td>
<td>Restriction enzyme incubation</td>
<td>5 × 1 ml (10× conc. solution)</td>
<td>11 417 975 001</td>
</tr>
<tr>
<td>Sure/Cut Buffer M</td>
<td>Restriction enzyme incubation</td>
<td>5 × 1 ml (10× conc. solution)</td>
<td>11 417 983 001</td>
</tr>
<tr>
<td>Water, PCR Grade</td>
<td>Specially purified, double-distilled, deionized, and autoclaved</td>
<td>100 ml (4 vials of 25 ml) 25 ml (25 vials of 1 ml)</td>
<td>03 315 843 001</td>
</tr>
<tr>
<td>BSA, special quality for molecular biology</td>
<td>Maintaining enzyme stability</td>
<td>20 mg (1 ml)</td>
<td>10 711 454 001</td>
</tr>
</tbody>
</table>