



Bsp120I (7)
PstI (6)
SdaI (6) **SpeI (13)** **BstEII (90)**
1 CCTGCAGGGCCACTAGTCTATTCTTACCTCCCTCCCTCTCCACCTGCTACTGGGTGCATCTCTGCTCCCCCTCCCCAGCAGATGGTTACCTTTG
101 GGCTGTTGCTTTCTTGTACCACCTGAGTTCTCAGACGCTGGAAAGCCATGTTCTCGGCTCTGTGAATGACAATGCTGACTGGAGTGTCCCTCTGTA
201 AAGGGCTGGGTGTGGATGGTCAACAAGCCCTCACATGCCTCAGCCAAGAGGAAGTAGTACAGGGGTCAGCCAGAGGTCCAGGGGAAAGGAGTGGAAACC
BamHI (392)
301 GATTTCCCAACCAAGGGAGGGCCTGTACCTCAGCTGTTCCCATAGCTACTTGCACAACCTGCCAAGCAAGTTTCGCTGAGTTTGACACATGGATCCCTG
AvrII (413)
401 TGGATCAACTGCCCTAGGACTCCGTTTGCACCCATGTGACTGTTGACTTTGCCCTGATGAAGCAGGGCCAACAGTCCCTAACTTAATACAAAAACT
EcoNI (539)
501 AATGACTAAGAGAGAGGTGGCTAGAGCTGAGGCCCTGAGTCAAGCTGTGGGTGGGATCATCTCCAGTACAGGAAGTGAAGCTTTCACTTTCTCTTTCC
NcoI (675)
601 AAGAGAGGGCTGAGGGAGCAGGGTTGAGCAACTGGTGCAGACAGCTAGCTGGACTTTGGGTGAGGCGTTTCAGCCATGGGGGTTCTCATCATCATC
▶ Me tG lYg lYse rH isH isH isH is
NheI (713) **Bsu36I (774)** **Acc65I (769)**
701 CATCATGGTATGGCTAGCATGACTGGTGGACAGCAAATGGGTGGGATCTGTACGACGATGACGATAAGGTACCTAAGGATCAGCTTGGAGTTGATCCCG
9▶ H isH isG lYme tA laSe rMe tTh rG lYg lYg lN lNme tG lYrArgAspLeuTy rAspAspAspLysVa lP roLysAspG lNLeuG lYVa lAsp rOv
801 TCGTTTTACAACGCTGTGACTGGGAAAACCTGGCGTTACCAACTTAATCGCTTGCAGCACATCCCTTTCCAGCTGGCGTAATAGCGAAGAGGC
42▶ a Ma lLeuG lNArgArgAspT rP g lUAsn rOg lYVa lTh rG lNLeuAsn rglLeuA la laH isP roP roPheA laSe rT rP rArgAsnSe rG lUg lUa l
901 CCGCACCGATCGCCCTCCCAACAGTTCGCGAGCCTGAATGGCGAATGGCGTTTGCCTGGTTCCGGCACCAGAAGCGGTGCCGAAAGCTGGCTGGAG
75▶ aArgTh rAspArgP roSe rG lN g lNLeuArgSe rLeuAsnG lYg lU t rP rArgPheA laT rP rPheP roA laP roG lUa laVa lP roG lUSe rT rPLeuG lU
Bsu36I (1011)
1001 TGCATCTTCTGAGGCCGATACTGTCTGCTGCTCCCTCAAACTGGCAGATGCACGGTTACGATGCGCCCATCTACACCAACGTAACCTATCCCACTACGG
109▶ CysAspLeuP roG lUa laAspTh rVa lVa lVa lP roSe rAsnT rP g lNme tH isG lYTy rAspA laP ro l leTy rTh rAsnVa lTh rTy rP ro l leTh rV
1101 TCAATCCCGCGTTTGTCCACGGAGAATCCGACGGGTTGTACTCGCTCACATTTAATGTTGATGAAAGCTGGCTACAGGAAGCCAGACCGCAATTAT
142▶ a lAsnP roP roPheVa lP roTh rG lUAsnP roTh rG lYCysTy rSe rLeuTh rPheAsnVa lAspG lUSe rT rPLeuG lN g lUg lYg lNTh rArg l le l l
1201 TTTTGATGGCGTTAACTCGCGTTTCATCTGTGGTGAACGGGCGTGGTGGTTCAGGCCAGGACAGTTCGTTTCCGCTGAATTTGACTGAGCGCA
175▶ ePheAspG lYVa lAsnSe rA laPheH isLeuT rP CysAsnG lY rG t rP Va lG lYTy rG lYg lNAspSe rArgLeuP roSe rG lU lPheAspLeuSe rA la
1301 TTTTACCGCGCGGAGAAAACCGCCTCGCGGTGATGGTGTGCTGCTGGAGTGACGGCAGTTATCTGGAAGATCAGGATATGTGGCGGATGAGCGGCATTT
209▶ PheLeuArgA laG lYg lUAsn rglLeuA laVa Me tVa lLeuArgT rPSe rAspG lYSe rTy rLeuG lUAspG lNAspMe tT rP rArgMe tSe rG lY l leP
AatII (1410)
1401 TCCGTGACGCTCGTGTGTCATAAACCGACTACACAAATCAGCGATTTCCATGTTGCCACTCGCTTAAATGATGATTTACGCCGCGTGTACTGGAGGC
242▶ heArgAspVa lSe rLeuLeuH isLysP roTh rTh rG lN l leSe rAspPheH isVa lA laTh rArgPheAsnAspAspPheSe rArgA laVa lLeuG lUa l
1501 TGAAGTTGATGTGCGGCGAGTTGCGTGACTACCTACGGGTAACAGTTTCTTTATGGCAGGGTGAACGCAGGTCCGCGAGCCGCGCCTTTCCGGC
275▶ aG lUa lG lNme tCysG lYg lULeuArgAspTy rLeuArgVa lTh rVa lSe rLeuT rP g lN g lYg lUTh rG lN Va lA laSe rG lYTh rA laP roPheG lY
Clal (1611)
1601 GGTGAAATTCGATGAGCGTGGTGGTTATGCCGATCGCTCACACTACGCTGAACGTCGAAAACCCGAACTGTGGAGCGCCGAAATCCCGAATCTCT
309▶ G lYg lU l le l leAspG lU rG lYg lYTy rA laAspArgVa lTh rLeuArgLeuAsnVa lG lUAsnP roLysLeuT rPSe rA laG lU l leP roAsnLeuT
1701 ATCGTGGCGTGGTTGAACTGCACACCGCCGACGCGCTGATTGAAGCAGAAGCCTCGATGTGCGTTTCCGCGAGGTGCGGATTGAAATGGTCTGCT
342▶ y rArgA laVa Ma lG lULeuH isTh rA laAspG lYTh rLeu l leG lUa laG lUa laCysAspVa lG lY lPheArgG lUa lArg l leG lUAsnG lYLeuLe
EcoRV (1900)
1801 GCTGCTGAACGGCAAGCGTGTGCTGATTGAGGCGTTAACCGTACAGGATCATCTCTGATGTTGAGGTGATGGATGAGCAGACGATGGTGCAGGAT
375▶ uLeuLeuAsnG lY lYsP roLeuLeu l leArg lYVa lAsn rgh isG lUh isH isP roLeuH isG lYg lN Va Me tAspG lUg lNTh rMe tVa lG lNAsp
DraIII (1977)
1901 ATCCTGCTGATGAAGCAGAACAACCTTAAACCGCGTGGCTGTTCCGATTATCCGAACATCCGCTGTGGTACACGCTGTGCGACCGCTACGGCTGTATG
409▶ l leLeuLeuMe tLysG lNAsnAsnPheAsnA laVa lArgCysSe rH isTy rP roAsnH isP roLeuT rP Ty rTh rLeuCysAspArgTy rG lYLeuTy rV
SspI (2017)
2001 TGGTGGATGAAGCAATATTGAAACCCACGGCATGGTCCAATGAATCGTCTGACCGATGATCCGCGTGGCTACCGCGATGAGCGAACCGCTAACCGG
442▶ a Ma lAspG lUa laAsn l leG lUTh rH isG lYme tVa lP roMe tAsn rglLeuTh rAspAspP ro rG t rPLeuP roA laMe tSe rG lU rglVa lTh rAr
BsaBI (2113)
2101 AATGGTGCAGCGCATCGTAATCACCCGAGTGTGATCATCTGGTCCGCTGGGGAATGAATCAGGCCACGGCGTAATCAGCAGCGCTGTATCGCTGGATC
475▶ gMe tVa lG lN rArgAsp rAsnH isP roSe rVa l l le l leT rPSe rLeuG lYAsnG lUSe rG lYH isG lY lA laAsnH isAspA laLeuTy rArgT rP l le
BssHIII (2285) **BbsI (2304)**
2201 AAATCTGTCGATCCTTCCCGCCGGTGCAGTATGAAGCGCGGAGCCGACACCAGCCACCGATATTATTTGCCGATGTACGCGCGTGGATGAAG
509▶ LysSe rVa lAspP roSe rArgP roVa lG lN Ty rG lUg lYg lYg lY lA laAspTh rTh rA laTh rAsp l le l leCysP roMe tTy rA la rglVa lAspG lUa
2301 ACCAGCCCTTCCCGGCTGTGCGGAAATGGTCCATCAAAAATGGCTTTTCGCTACCTGGAGAGACGCGCCGCTGATCCTTTGCGAATACGCCACCGCAT
542▶ spG lN rP rPheP roA laVa lP roLysT rPSe r l leLysLysT rPLeuSe rLeuP roG lYg lUTh rArgP roLeu l leLeuCysG lU Ty rA laH isA laMe
2401 GGGTAACAGTCTGGCGGTTTCGCTAAACTGCGAGCTGGCAGTACCCCGTTTACAGGGCGGCTTCGCTGGACTGGGTGGATCAGTCCGCTG
575▶ tG lYAsnSe rLeuG lYg lY lPheA laLysTy rT rP g lN la PheA rglN Ty rP ro rglLeuG lN g lYg lY lPheVa lT rPAspT rP Va lAspG lNSe rLeu
2501 ATTAATATGATGAAAACCGCAACCCGCTGGTGGCTACGGCGGTGATTTGGCGATACGCCAACGATCGCCAGTTCTGTATGAACGGTCTGGTCTTTG
609▶ l leLysTy rAspG lUAsnG lYAsnP roT rPSe rA laTy rG lYg lYAspPheG lYAspTh rP roAsnAspArgG lN lPheCysMe tAsnG lYLeuVa lPheA
Eco47III (2622)
2601 CCGACCGCAGCCGATCCAGCGCTGACGGAAGCAAAACACCCAGCAGCAGTTTTTCCAGTCCGTTTATCCGGGCAAAACCTCGAAGTGACCAGCGAATA
642▶ laAspArgTh rP roH isP roA laLeuTh rG lUa laLysH isG lN g lN g lN lPhePheG lN lPheArgLeuSe rG lYg lNTh r l leG lUa lTh rSe rG lU Ty
SacI (2727)
2701 CCTGTTCCGTCATAGCGATAACGAGCTCTGCACTGGATGGTGGCGCTGGATGGTAAGCCGCTGGCAAGCGGTGAAGTGCCTCTGGATGTCGCTCCACAA
675▶ rLeuPheArgH isSe rAspAsnG lULeuLeuH isT rPMe tVa lA laLeuAspG lY lYsP roLeuA laSe rG lYg lUa lP roLeuAspVa lA laP roG lN
2801 GGTAACAGTGTGATTGAACTGCCTGAACTACCGAGCCGAGAGCGCCGGCAACTCGCTCACAGTACCGGTAGTGAACCGAACCGACCGCATGGT
709▶ G lY lYsG lNLeu l leG lULeuP roG lULeuP roG lN rOg lUSe rA laG lYg lNLeuT rPLeuTh rVa lA rglVa Ma lG lN rP roAsnA laTh rA laT rP s
2901 CAGAAGCCGGGCACATCAGCGCTGGCAGCAGTGGCGTCTGGCGGAAAACCTCAGTGTGACGCTCCCGCCGCTCCACGCCATCCCGCATCTGACCAC
742▶ e rG lUa laG lYH is l leSe rA laT rP g lN g lN t rP rglLeuA laG lUAsnLeuSe rVa lTh rLeuP roA laA laSe rH isA la l leP roH isLeuTh rTh

3001 CAGCGAAATGGATTTTGCATCGAGCTGGGTAATAAGCGTTGGCAATTAACCGCCAGTCAGGCTTCTTTACAGATGTGGATTGGCGATAAAAAACAA
775 ▶ rSe rG luMe tAspPheCys I leG luLeuG lyAsnLysA rGt rpG lnPheAsnA rG InSe rG lyPheLeuSe rG lnMe tT rp I leG lyAspLysLysG ln
3101 CTGCTGACGCCGCTGCGCATCAGTTCACCCGTGCACCGCTGGATAACGACATTGGCGTAAGTGAAGCGACCCGATTGACCTAACGCCTGGGTGGAAC
809 ▶ LeuLeuTh rP roLeuArgAspG lnPheTh rArgA laP roLeuAspAsnAsp I leG lyVa lSe rG luA laTh rArg I leAspP roAsnA laT rpVa lG luA
3201 GCTGGAAGGCGCGGCCATTACCAGGCCGAAGCAGCGTTGTTGCACTGCACGGCAGATACACTTGTCTGATGCGGTGCTGATTACGACCGCTCACGCGTG
842 ▶ rgT rpLysA laA laG lyH isTy rG lnA laG luA laA laLeuLeuG lnCysTh rA laAspTh rLeuA laAspA laVa lLeu I leTh rTh rA laH isA laT r
3301 GCAGCATCAGGGGAAAACCTTATTTATCAGCCGAAAACCTACCGGATTGATGGTAGTGGTCAAATGGCGATTACCGTGTGATTGAAGTGGCGAGCGAT
875 ▶ pG lnH isG lnG lyLysTh rLeuPhe I leSe rArgLysTh rTy rArg I leAspG lySe rG lyG lnMe tA la I leTh rVa lAspVa lG luVa lA laSe rAsp
3401 ACACCGCATCCGGCGGATTGGCCTGAACTGCCAGCTGGCGCAGGTAGCAGAGCGGGTAAACTGGCTCGGATTAGGGCCGCAAGAAAACCTATCCCGACC
909 ▶ Th rP roH isP roA laArg I leG lyLeuAsnCysG lnLeuA laG lnVa lA laG luArgVa lAsnT rpLeuG lyLeuG lyP roG lnG luAsnTy rP roAspA

BsiWI (3557)

Bst1107I (3549)

BspLU11I (3546) BbsI (3556)

3501 GCCTTACTGCCGCTGTTTTGACCGCTGGGATCTGCCATTGTGACACATGTATACCCCGTACGTCTTCCGAGCGAAAACGGTCTGCGCTGCGGGACGGC
942 ▶ rgLeuTh rA laA laCysPheAspA rGt rpAspLeuP roLeuSe rAspMe tTy rTh rP roTy rVa lPheP roSe rG luAsnG lyLeuA rgCysG lyTh rA r
3601 CGAATTGAATTATGGCCACACCAAGTGGCGGGGCACTTCCAGTTCAACATCAGCCGCTACAGTCAACAGCAACTGATGGAACCCAGCCATCGCCATCTG
975 ▶ gG luLeuAsnTy rG lyP roH isG lnT rpArgG lyAspPheG lnPheAsn I leSe rArgTy rSe rG lnG lnG lnLeuMe tG luTh rSe rH isArgH isLeu

NdeI (3744)

3701 CTGCACGCGGAAGAAGGCACATGGCTGAATATCGACGGTTTCCATATGGGATTGGTGGCGACGACTCTGGAGCCCGTCAGTATCGGCGGAATTACAGC
1009 ▶ LeuH isA laG luG luG lyTh rT rpLeuAsn I leAspG lyPheH isMe tG ly I leG lyG lyAspAspSe rT rpSe rP roSe rVa lSe rA laG luLeuG lnL

NheI (3866)

EcoRI (3860)

3801 TGAGCGCCGGTCTGCTACCATTACCAGTTGGTCTGGTGTCAAAAATAATAA TCTAGTCGAGAATTCGCTAGCTCGACATGATAAGATACATTGATGAGTTT
1042 ▶ euSe rA laG lyArgTy rH isTy rG lnLeuVa lT rpCysG lnLys●●●
3901 GGACAAACCACAAC TAGAATGCAGTGAAAAAATGCTTTATTTGTGAAATTTGTGATGCTATTGCTTTATTTGTGAAATTTGTGATGCTATTGCTTTATT

MfeI (4040)

DraI (4089)

4001 TGTAACCATTATAAGCTGCAATAAACAAAGTTAACAAACAATTCATTCTTTATGTTTCAGGTTCCAGGGGAGGTGTGGGAGGTTTTTAAAGCAAG

DraI (4128)

SwaI (4131)

4101 TAAAACTCTACAAATGTGGTAGATCCATTTAAATGTTAATTAAGTCCATGACCAAAATCCCTTAACGTGAGTTTTCTGTTCCACTGAGCGTCAGACCC

4201 CGTAGAAAAGATCAAAGGATCTTCTTGAGATCCTTTTTTCTGCGCGTAATCTGCTGCTTGC AAAACAAAAAACCCGCTACCAGCGGTGTTTGTTTG

4301 CCGGATCAAGAGCTACCAACTCTTTTTCCGAAGGTAAGTGGCTTCCAGCAGAGCCAGATACCAAACTACTGTTCTTAGTGTAGCCGTAGTTAGGCCACC

4401 ACTTCAAGAACTCTGTAGCACCGCTACATACCTGCTCTGTAATCTGTTACAGTGGCTGCTGCCAGTGGCGATAAGTCGTGTCTTACCGGGTTGGA

4501 CTC AAGACGATGTTACCGATAAAGCGCAGCGGTGGGCTGAACGGGGGTTCTGTCACACAGCCAGCTTGAGCGAACGACCTACACCGAACTGAGA

4601 TACCTACAGCGTGAGCTATGAGAAAGCCACGCTTCCCGAAGGGAGAAAGCGGACAGGTATCCGGTAAGCGGCAGGGTCGGAACAGGAGAGCCACGA

4701 GGGAGCTTCCAGGGGAAACGCCTGATCTTTATAGTCTGCTGGGTTTCCGCACCTCTGACTTGAGCGTCGATTTTTGTGATGCTGTCAGGGGGGGC

BspLU11I (4869)

4801 GAGCCTATGAAAAACGCCAGCAACCGCGCTTTTTACGGTTCCTGGCCTTTTGTGGCCTTTTGTCTACATGTTCTTAATTAATTTTTCAAAGTAGT

AseI (4907)

SfiI (4958) MscI (4969)

4901 TGACAATTAATCATCGGCATAGTATATCGGCATAGTATAATACGACTCACTATAGGAGGGCCATCATGGCCAAGTTGACCACTGCTGTCCAGTGTCTCAC

5001 AGCCAGGATGTGGCTGGAGCTGTTGAGTTCTGGACTGACAGGTTGGGTTCTCCAGAGATTTTGTGGAGGATGACTTGCAGTGTGGTCCAGAGATGAT

5101 GTCACCCCTGTTCTCAGCAGTCCAGGACAGGTGGTGCCTGACAACCCCTGGCTGGGTGGGTGAGAGGACTGGATGAGCTGTATGCTGAGTGGAG

5201 GTGAGGTGGTCTCCACCAACTTCAGGGATGCCAGTGGCCCTGCCATGACAGAGATTGGAGAGCAGCCCTGGGGGAGAGAGTTGCCCTGAGAGACCCAGC

5301 AGGCAACTGTGCTGCTTTGTGGCAGAGGAGCAGGACTGAGGATAAGAATTGAGTTTCAGAAAAGGGGGCCTGAGTGGCCCTTTTTTCAACTTAATTAA

DraIII (5319)

SfiI (5367)

112 ▶ aG lyAsnCysVa lH isPheVa lA laG luG luG lnAsp●●●