

BGH13_B05a2-pET16-DEST

5' ttcttgaagacgaaagggcctcgtgatacgcctatTTTTATAGGTTAATGTCATGATAATAATGGTTTCT
 0 ++++++
 3' aagaacttctgctttcccggagcactatgCGGATAAAAATATCCAATTACAGTACTATTATTACCAAAGA
 0
 5' tagacgtcaggtggcacttttcggggaaatgtgCGCGGAACCCCTATTTGTTATTTTTCTAAATACATT
 0 ++++++
 3' atctgcagtccaccgtgaaaagccccctttacacgCGCCTGGGGATAAACAAATAAAAAGATTTATGTA
 0
 0
 5' caaatatgtatccgctcatgagacaataaccctgataaatgcttcaataatattgaaaaaggaagagtat
 0 ++++++
 3' gtttatacataggcgagtactctgttattgggactatttacgaagtattataactTTTTCTTCTCATA
 0

Sspl

70
140
210



5' gagtattcaacatttccgtgTCGCCCTTATCCCTTTTTGCGGCATTTTGCTTCTCTGTTTTGCTCAC
 0 ++++++
 3' ctcataagttgtaaaggcacagCGGGAATAAGGGAAAAACGCCGTA AACCGGAAGGACAAAACGAGTG
 0
 0
 5' ccagaaacgctggTGAAGTAAAAGATGCTGAAGATCAGTTGGGTGCACGAGTGGGTACATCGAAGTGG
 0 ++++++
 3' ggtctttgCGACCCTTTCATTTCTACGACTTCTAGTCAACCCACGTGCTCACCCAATGTAGCTTGACC
 0
 0

Ap

Ap

280
350

5' atctcaacagCGGTAAGATCCTTGAGAGTTTTCGCCCCGAAGAACGTTTTCCAATGATGAGCACTTTTAA
 0 ++++++
 3' TAGAGTTGTCGCCATTCTAGGAACCTCTCAAAAGCGGGGCTTCTTGCAAAAGGTACTACTCGTGAAAAT
 0
 0
 5' agttctgctatgtggCGCGGTATTATCCCGTGTGACGCGGGCAAGAGCAACTCGGTGCGCGCATACAC
 0 ++++++
 3' tcaagacgatacaccgCGCCATAATAGGGCACAACCTGCGGCCGTTCTCGTTGAGCCAGCGGCATATGTG
 0
 0

Ap

Ap

420
490

5' tattctcagaatgacttggTGTGAGTACTCACCAGTCACAGAAAAGCATCTTACGGATGGCATGACAGTAA
 0 ++++++
 3' ATAAGAGTCTTACTGAACCAACTCATGAGTGGTCAGTGTCTTTTCGTAGAATGCCTACCCTACTGTCAAT
 0
 0

Scal

Ap

560

5' gagaattatgcagTGTGCCATAACCATGAGTGATAACACTGCGGCCAACTTACTTCTGACAACGATCGG
 0 ++++++
 3' CTCTTAATACGTCACGACGGTATTGGTACTCACTATTGTGACGCGGTTGAATGAAGACTGTTGCTAGCC
 0
 0

PvuI

Ap

630

BGH13_B05a2-pET16-DEST

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3' tectggcttctcgattggcgaaaaaacgtggtgtacccttagtacattgagcggaactagcaaccctt
Ap

700

PstI

5' ccggagctgaatgaagccataccaaacgacgagcgtgacaccacgatgcctgcagcaatggcaacaacgt
0
3' ggccctcgacttacttccggtatggtttgctgctcgcaactgtgggtgctacggacgtcgttaccgttgttgca
Ap

770

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0
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Ap

840

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0
3' cctatttcaacgtcctggtgaagacgacgagccgggaaggccgaccgaccaataacgactattagacct
Ap

910

BsaI

5' gccggtgagcgtgggtctcgcggtatcattgcagcactggggccagatggtaagccctcccgatcgtag
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Ap

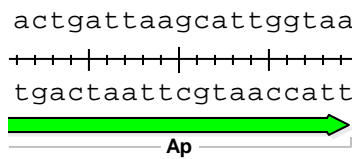
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AhdI

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Ap

1050

Ap



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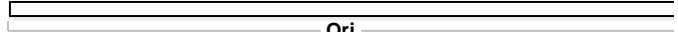
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1190

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1260

Ori



BGH13_B05a2-pET16-DEST

5' cgtaatctgctgcttgcaaacaaaaaaaccaccgctaccagcgggtggtttggttgccggatcaagagcta
 1330
 3' gcattagacgacgaacgtttgttttttggggcgatggtcgccaccaaacaaacggcctagttctcgat
 Ori

5' ccaactcttttccgaaggtaactggcttcagcagagcgcagataccaaatactgtccttctagtgtagc
 1400
 3' ggttgagaaaaaggcttccattgaccgaagtctgtctcgctctatggtttatgacaggaagatcacatcg
 Ori

5' cgtagttaggccaccacttcaagaactctgtagcaccgcctacatacctcgctctgctaactctgttacc
 1470
 3' gcatcaatccgggtggtgaagtcttgagacatcgtggcggatgtatggagcgcgagacgattaggacaatgg
 Ori

5' agtggctgctgccagtggcgataagtctgtgcttaccgggttgactcaagacgatagttaccggataag
 1540
 3' tcaccgacgacggtcaccgctattcagcacagaatggcccaacctgagttctgctatcaatggcctattc
 Ori

5' ggcgagcggtcgggctgaacggggggttcgtgcacacagcccagcttgagcgaacgacctacaccgaac
 1610
 3' cgcgctgccagcccgacttgcccccaagcacgtgtgtcgggtcgaacctcgcttgctggatgtggcttg
 Ori

5' tgagatacctacagcgtgagctatgagaaagcgccacgcttcccgaagggagaaaggcggacaggtatcc
 1680
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 Ori

5' ggtaagcggcagggtcggaacaggagagcgcacgaggagcttccaggggaaacgcctggatatctttat
 1750
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 Ori

5' agtcctgtcgggttccgccacctctgacttgagcgtcgatTTTTGTGATGCTCGTCAGGGGGGCGGAGCC
 1820
 3' tcaggacagcccaaagcgggtggagactgaactcgcagctaaaaacactacgagcagtcccccgcctcgg
 Ori

5' tatggaaaaacgccagcaacgcggcctttttacgggttctggccttttgetggccttttgetcacatggt
 1890
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 Ori

5' ctttcctgcggttatcccctgattctgtggataaccgtattaccgcctttgagtgagctgataaccgctcgc
 1960
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Pcil

BGH13_B05a2-pET16-DEST

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3'   cgtcgtcagcgaagtgcgaagcgagcgcatagccactaagtaagacgattgggtcattccggtggggcggtc
o
o
o
                    FspAI
                    |
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o   ++++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|
3'   ggatcggcccaggagttgctgtcctcgctgctagtagcgcgtgggcaccggctcctgggttgcgacgggctct
o
o
o
                    Aval
                    |
                    BmeT110I
                    ||
5'   tgcgcccgctgcggtgctggagatggcggacgcgatggatatgttctgccaagggttggtttgcgcatt
o   ++++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|
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o
o
o
5'   cacagttctccgcaagaattgattgggtccaattcttggagtgggtgaatccggttagcggaggtgccgccgg
o   ++++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|
3'   gtgtcaagagggcgttcttaactaaccgaggttaagaacctcaccacttaggcaatcgctccacggcggcc
o
o
o
5'   ctccattcaggtcgaggtggccccggctccatgcaccgcgacgcaacgcggggaggcagacaaggtatag
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o
o
o
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o   ++++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|
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o
o
o
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o   ++++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|+++++|
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o
                    BspMI
                    |
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o
o
o
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                    |
                    EagI
                    |
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5'   gaaaatgaccagagcgcgtgccggcacctgtcctacgagttgcatgataaagaagacagtcataagtgcg
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o
o

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o

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Lac I

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Lac I

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Lac I

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Lac I

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Lac I

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o

5' ttactggtttcacattcaccacctgaattgactctcttccgggcgctatcatgccataccgcgaaaggt 4900

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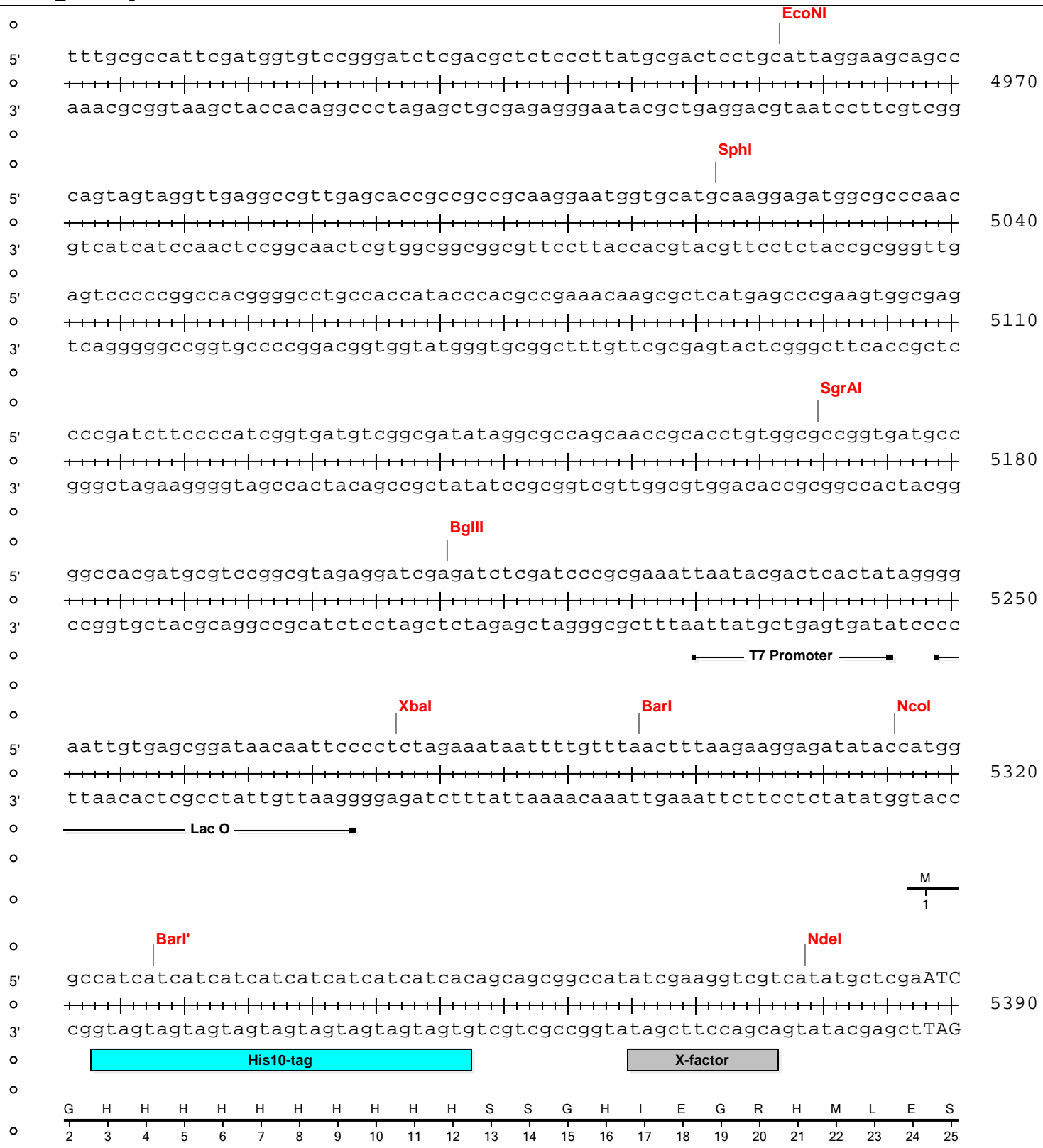
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PspOMI

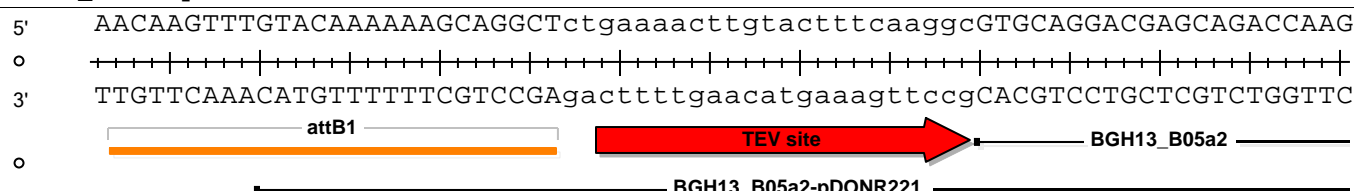
Apal

BstEII

MluI



BGH13_B05a2-pET16-DEST



5' AACAAAGTTTGTACAAAAAGCAGGCTctgaaaacttgtactttcaaggcGTGCAGGACGAGCAGACCAAG
 3' TTGTTCAAACATGTTTTTTCGTCCGAgacttttgaacatgaaagtccgcCACGTCCTGCTCGTCTGGTTC

5460

T S L Y K K A G S E N L Y F Q G V Q D E Q T K
 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48

5' ATATTCAGCGCACAAAGTCTGGGCACCGGAAAAGCCATATAAATTCAAGAAAAGACATTTAAGCCTACTA
 3' TATAAGTCGCGTGTTCAGACCCGTGGCCTTTTCGGTATATTTAAGTTCTTTTTCTGTAAATTCGGATGAT

5530

I F S A Q V W A P E K P Y K F K K K T F K P T
 49 50 51 52 53 54 55 56 57 58 59 60 61 62 63 64 65 66 67 68 69 70 71

Ajul

Ajul'

5' CGAATCCTTTGTTGATCTATGAATGTCATATTGGAATGGCACAAACAGGAGGAAAAAGTAGGCACATACAA
 3' GCTTAGGAAACAACACTAGATACTTACAGTATAACCTTACCGTGTGTCTCCTTTTTTCATCCGTGTATGTT

5600

T N P L L I Y E C H I G M A Q Q E E K V G T Y N
 72 73 74 75 76 77 78 79 80 81 82 83 84 85 86 87 88 89 90 91 92 93 94 95

5' TGAATCCGCGAAAAAACAACCTTCCACGCATAGCACAGGAAGGCTATAACTGTATTCAGATTATGGCCATT
 3' ACTTAAGGCGCTTTTTTGTGAAGGTGCGTATCGTGTCTTCCGATATTGACATAAGTCTAATACCGGTAA

5670

E F R E K T L P R I A Q E G Y N C I Q I M A I
 96 97 98 99 100 101 102 103 104 105 106 107 108 109 110 111 112 113 114 115 116 117 118

5' CAGGAGCATCCTTATTATGGTAGTTTTGGCTATCATGTATCCAGTTTCTTTGCTGCTTCCTCCCGCTTTG
 3' GTCCTCGTAGGAATAATACCATCAAACCGATAGTACATAGGTCAAAGAAACGACGAAGGAGGGCGAAAC

5740

Q E H P Y Y G S F G Y H V S S F F A A S S R F
 119 120 121 122 123 124 125 126 127 128 129 130 131 132 133 134 135 136 137 138 139 140 141

BGH13_B05a2-pET16-DEST

5' GTACGCCTGACGAAGCTGAAAGCGCTGATTGATGCTGCTCATGAGATGGGGATTGCCGTTATTATGGATAT
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5810

BGH13_B05a2

BGH13_B05a2-pDONR221

G T P D E L K A L I D A A H E M G I A V I M D I
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5880

BGH13_B05a2

BGH13_B05a2-pDONR221

V H S H A V K N E V E G L G N F A G D P N Q Y
 166 167 168 169 170 171 172 173 174 175 176 177 178 179 180 181 182 183 184 185 186 187 188

BseRI

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 3' AAGATAGGCCGAGGAGCAGCACTCGTAGGCCGTACCCTAAGTGATACGAAACTAATACCATTTTTACTTC

5950

BGH13_B05a2

BGH13_B05a2-pDONR221

F Y P A P R R E H P A W D S L C F D Y G K N E
 189 190 191 192 193 194 195 196 197 198 199 200 201 202 203 204 205 206 207 208 209 210 211

5' TGATCCATTTCTTATTGTCCAACCTGTAAATACTGGCTGGAAGAATATAAGTTTGACGGATTCCGTTTTGA
 3' ACTAGGTAAAGAATAACAGGTTGACATTTATGACCGACCTTCTTATATTCAAACCTGCCTAAGGCAAAACT

6020

BGH13_B05a2

BGH13_B05a2-pDONR221

V I H F L L S N C K Y W L E E Y K F D G F R F D
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5' CGGTGTGACGTCTATGTTGTATTATAGCCACGGGTTGGGAGAAGCATTCTGTAATTATGGAGATTACTTC
 3' GCCACACTGCAGATAACAATAATATCGGTGCCCAACCCTCTTCGTAAGACATTAATACCTCTAATGAAG

6090

BGH13_B05a2

BGH13_B05a2-pDONR221

G V T S M L Y Y S H G L G E A F C N Y G D Y F
 236 237 238 239 240 241 242 243 244 245 246 247 248 249 250 251 252 253 254 255 256 257 258

BGH13_B05a2-pET16-DEST

```
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+
3' TTACCAGTAGTCTTGCCGTTACGGTAAACGATGGACTGTAACCGCTTACTCAACTAAGTAGTCCAGTTAG
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6160

BGH13_B05a2

BGH13_B05a2-pDONR221

```
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259 260 261 262 263 264 265 266 267 268 269 270 271 272 273 274 275 276 277 278 279 280 281
```

```
5' CGAAGGCTATCTCTATTGCTGAAGAAGTTTCCGGTATGCCGGGACTTGCCGCCAAAGTAGAAGATGGCGG
+
3' GCTTCCGATAGAGATAACGACTTCTTCAAAGGCCATACGGCCCTGAACGGCGGTTTCATCTTCTACCGCC
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6230

BGH13_B05a2

BGH13_B05a2-pDONR221

```
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282 283 284 285 286 287 288 289 290 291 292 293 294 295 296 297 298 299 300 301 302 303 304 305
```

```
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+
3' AATGCCTAAACTAATGGCATAACCGATACTTGTAGGGACTAATGACCTAGTTCTGATAGTTCTTTTCTAA
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6300

BGH13_B05a2

BGH13_B05a2-pDONR221

```
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306 307 308 309 310 311 312 313 314 315 316 317 318 319 320 321 322 323 324 325 326 327 328
```

```
5' GATGAGGACTGGAAACCTTCCAGTATGTTCTGGGAAGTAACCAACCGTCGTCAGGATGAAAAGACGATCT
+
3' CTACTCCTGACCTTTGGAAGGTCATACAAGACCCTTCATTGGTTGGCAGCAGTCCTACTTTTCTGCTAGA
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6370

BGH13_B05a2

BGH13_B05a2-pDONR221

```
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329 330 331 332 333 334 335 336 337 338 339 340 341 342 343 344 345 346 347 348 349 350 351
```

```
5' CTTATGCGGAAAGTCATGATCAGGCATTGGTAGGAGACAAGACTATTATTTTCCGTTAGGACCCAGCTTT
+
3' GAATACGCCTTTCAGTACTAGTCCGTAACCATCCTCTGTTCTGATAATAAAAGGCAATCCTGGGTCGAAA
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6440

BGH13_B05a2

attB2

BGH13_B05a2-pDONR221

```
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352 353 354 355 356 357 358 359 360 361 362 363 364 365 366 367 368 369 370 371
```

BamHI

```
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+
3' GAACATGTTTACCAACTAagctcctaggccgacgattgtttcgggctttccttcgactcaaccgacgac
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6510

attB2

