

BGT41_T01a3b-pET32-DEST

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 3' agcggggcttcttgcaaaagggttactactcgtgaaaatttcaagacgatacacccgcgcataataggga
 0
 0
 0

840

Ap

Scal

5' attgacgccgggcaagagcaactcggtcgccgcatacactattctcagaatgacttgggtgagtactcac
 0
 3' taactgcgggcccgttctcgttgagccagcggcgtatgtgataagagtcttactgaaccaactcatgagtg
 0
 0
 0

910

Ap

5' cagtcaagaaaagcatcttacggatggcatgacagtaagagaattatgcagtgctgccataacatgag
 0
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 0
 0
 0

980

Ap

PvuI

5' tgataaactgcgccaacttacttctgacaacgatcggaggaccgaaggagctaaccgcttttttgca
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 3' actattgtgacgccggttgaatgaagactgttgctagcctcctggcttctcgcattggcgaaaaaacgtg
 0
 0
 0

1050

Ap

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 0
 0
 0

1120

Ap

PstI

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 0
 3' tcgcactgtgggtgctacggacgtcgttaccgcttggtgcaacgcgcttgataattgaccgcttgatgaatg
 0
 0
 0

1190

Ap

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 0
 0

1260

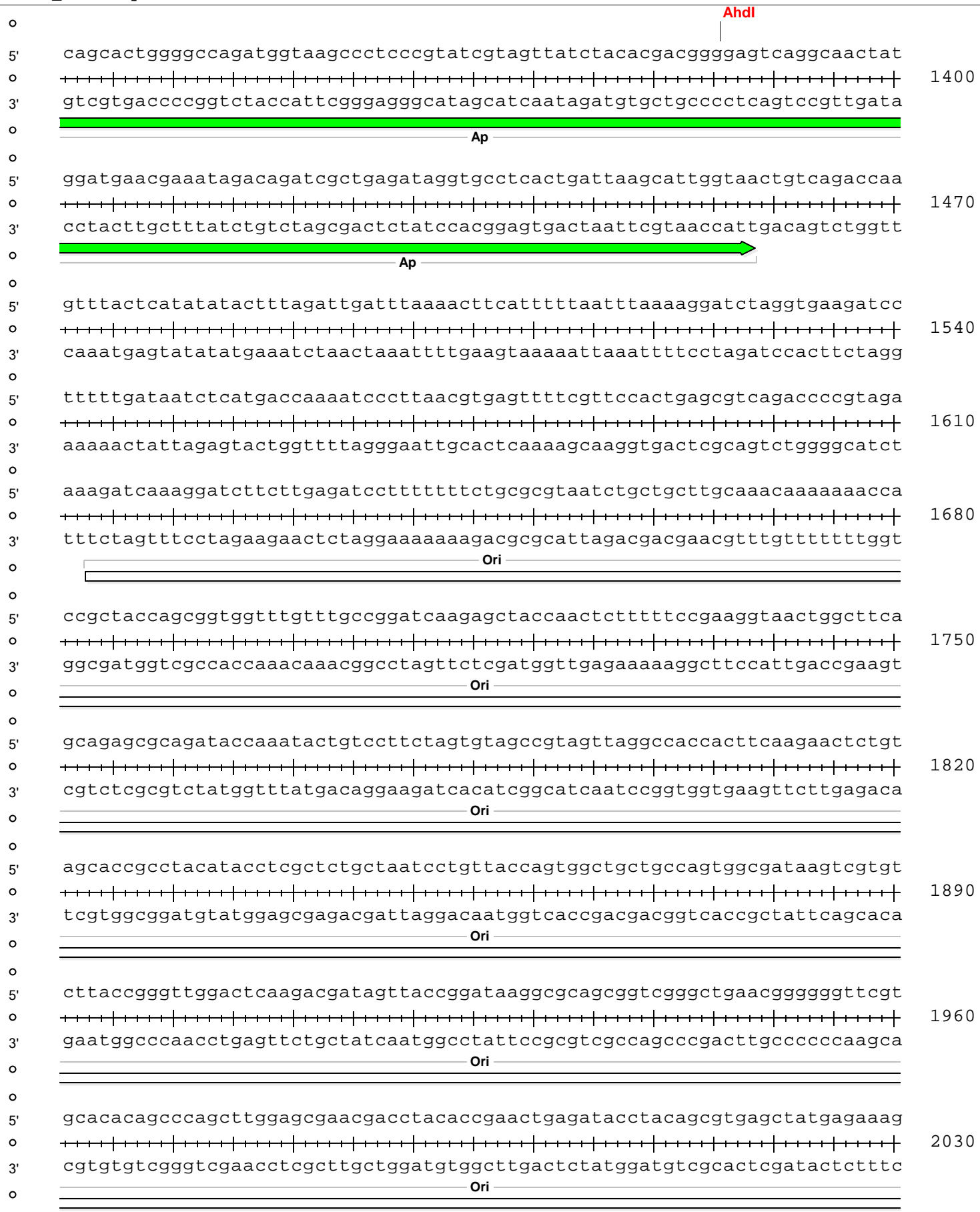
Ap

Bsal

5' gcccttccggctggctggtttattgctgataaatctggagccggtagcgtgggtctcgcggtatcattg
 0
 3' cgggaaggccgaccgaccaataacgactatthagacctcggccactcgcaccagagcgcctatagtaac
 0
 0
 0

1330

Ap



BGT41_T01a3b-pET32-DEST

5' cgccacgcttcccgaaggggagaagggcggacaggtatccggtaagcggcagggctcggaacaggagagcgc
 2100
 3' gcggtgcaagggcttccctctttccgcctgtccataggccattcgccgtccacgccttgtcctctcgcg
 Ori

5' acgagggagcttccaggggaaacgcctggtatctttatagtcctgtcgggttcgccacctctgacttg
 2170
 3' tgctccctcgaaggtccccctttgcgaccatagaaatatcaggacagccaaagcgggtggagactgaac
 Ori

5' agcgtcgatTTTTgtgatgctcgtcagggggcggagcctatggaaaaacgccagcaacgcggcctttt
 2240
 3' tcgcagctaaaaaactacgagcagtcccccgcctcggatacctttttgcggtcgttgcgccggaaaaa
 Ori

5' acggttctctggccttttggctggccttttggctcacatgttctttcctgcttatccctgattctgtggat
 2310
 3' tgccaaggaccggaaaacgaccggaaaacgagtgtacaagaaaggacgcaataggggactaagacaccta
 PciI

5' aaccgtattaccgcttttgagtgagctgataccgctcgccgcagccgaacgaccgagcgcagcagtcag
 2380
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 SapI

5' tgagcgaggaagcgaagagcgcctgatgcggtatTTTctccttacgcatctgtgcggtatttcacaccg
 2450
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 BstZ17I

5' catatatggtgcactctcagtacaatctgctctgatgccgcatagttaagccagtatacactccgctatc
 2520
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 Tth111I

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 2660
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BGT41_T01a3b-pET32-DEST

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 0 ++++++
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SgrAI

5' accgcacctgtggcgccgggtgatgccggccacgatgcgctccggcgtagaggatcgagatcgatctcgatc 5110
 0 ++++++
 3' tggcgtggacaccgcggccactacggccgggtgctacgcaggccgcattctcctagctctagctagagctag

Xbal

5' ccgcgaaattaatacgactcactataggggaattgtgagcggataacaattcccctctagaataatttt 5180
 0 ++++++
 3' ggcgctttaattatgctgagtgatatccccttaacactcgctattgttaaggggagatctttattaa

T7 Promoter Lac Operator

5' gtttaactttaagaaggagatatacatatgagcgataaaattattcacctgactgacgacagttttgaca 5250
 0 ++++++
 3' caaattgaaattcttccctctatatgtatactcgctattttaataagtgactgactgctgtcaaaactgt

Trx Tag

M S D K I I H L T D D S F D
 1 2 3 4 5 6 7 8 9 10 11 12 13 14

5' cggatgtactcaaagcggacggggcgatcctcgtcgatttctgggcagagtgggtgcggtccgtgcaaaat 5320
 0 ++++++
 3' gcctacatgagtttcgcctgccccgctaggagcagctaaagaccctctcaccacgccaggcacgtttta

Trx Tag

T D V L K A D G A I L V D F W A E W C G P C K M
 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38

5' gatcggcccgattctggatgaaatcgctgacgaatatcagggcaaaactgaccgttgcaaaactgaacatc 5390
 0 ++++++
 3' cttagcggggctaagacctactttagcgactgcttataagctcccgtttgactggcaacgttttgactttag

Trx Tag

I A P I L D E I A D E Y Q G K L T V A K L N I
 39 40 41 42 43 44 45 46 47 48 49 50 51 52 53 54 55 56 57 58 59 60 61

5' gatcaaaaccctggcactgcgccgaaatatggcatccgtggtatcccgactctgctgctgttcaaaaacg 5460
 0 ++++++
 3' ctagttttgggaccgtgacgcggctttataccgtaggcaccatagggctgagacgacgacaagtttttgc

Trx Tag

D Q N P G T A P K Y G I R G I P T L L L F K N
 62 63 64 65 66 67 68 69 70 71 72 73 74 75 76 77 78 79 80 81 82 83 84

BGT41_T01a3b-pET32-DEST

5' gtgaagtggcggcaaccaaagtgggtgcaactgtctaaaggtcagttgaaagagttcctcgacgctaacct
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 3' cacttcaccgcccgttggtttcaccacgtgacagatttccagtcactttctcaaggagctgcgattgga
 0
 0
 Trx Tag

5530

G E V A A T K V G A L S K G Q L K E F L D A N L
 85 86 87 88 89 90 91 92 93 94 95 96 97 98 99 100 101 102 103 104 105 106 107 108

5' ggccggttctggttctggccatatgcaccatcatcatcattcttctggtctggtgccacgcggttct
 0 ++++++
 3' ccggccaagaccaagaccggtatacgtggtagtagtagtaagaagaccagaccacggtgcgccaaga
 0
 0
 T...g His6-tag Thrombin

5600

A G S G S G H M H H H H H S S G L V P R G S
 109 110 111 112 113 114 115 116 117 118 119 120 121 122 123 124 125 126 127 128 129 130 131

5' ggtatgaaagaaaccgctgctgctaaattcgaacgccagcacatggacagcccagatctgggtaccgacg
 0 ++++++
 3' ccatacttcttggcgacgacgatttaagcttgcggtcgtgtacctgtcgggtctagacctatggctgc
 0
 0
 BstBI Acc65I KpnI En...e

5670

G M K E T A A A K F E R Q H M D S P D L G T D
 132 133 134 135 136 137 138 139 140 141 142 143 144 145 146 147 148 149 150 151 152 153 154

5' acgacgacaaggccatggcgatATCAACAAGTTTGTACAAAAAAGCAGGCTctgaaaacttgtactttca
 0 ++++++
 3' tgctgctgttccggtaccgctaTAGTTGTTCAAACATGTTTTTTCGTCCGAgacttttgaacatgaaagt
 0
 0
 Enterokinase attB1 TEV site

5740

D D D K A M A I S T S L Y K K A G S E N L Y F Q
 155 156 157 158 159 160 161 162 163 164 165 166 167 168 169 170 171 172 173 174 175 176 177 178

5' aggcAAAGCTATTAATCCATGGGGATTACTTTTCATTAGAAAATAATCCTTTAAATCATTAAAAAGATCT
 0 ++++++
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 0
 0
 T... BGT41_T01c3 BGT41_T01a3b-pDONR221

5810

G K A I N P W G L L S L E N N P L N H L K R S
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BGT41_T01a3b-pET32-DEST

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 3' TTCTTTAAAATATTTCTTTTATATATGCTGGTTGAGTTATATAATTTGGAAGGGTTTTTCAAATTAAG

————— BGT41_T01c3 —————
 ————— BGT41_T01a3b-pDONR221 —————

K K F Y K E K Y I R P T Q Y I K P S P K S L I
 202 203 204 205 206 207 208 209 210 211 212 213 214 215 216 217 218 219 220 221 222 223 224

5880

BseRI

5' ATATAGGTTATTTCTCTGCTGATTTCTGAATCATCCTGTAATGCAACTAATTGCTCCTCTACTTGAGCT
 3' TATATCCAATAAAGAGACGACTAAAGGACTTAGTAGGACATTACGTTGATTAACGAGGAGATGAACTCGA

————— BGT41_T01c3 —————
 ————— BGT41_T01a3b-pDONR221 —————

H I G Y F S A D F L N H P V M Q L I A P L L E L
 225 226 227 228 229 230 231 232 233 234 235 236 237 238 239 240 241 242 243 244 245 246 247 248

5950

5' ACATGATAAATATAGGTTTAAAATATATTTATACTCATTTGTGCCAAAAGAAGATGAATATACTGAAAGA
 3' TGTACTATTTATATCCAAATTTTATATAAATATGAGTAAACACGGTTTTTCTTCTACTTATATGACTTTCT

————— BGT41_T01c3 —————
 ————— BGT41_T01a3b-pDONR221 —————

H D K Y R F K I Y L Y S F V P K E D E Y T E R
 249 250 251 252 253 254 255 256 257 258 259 260 261 262 263 264 265 266 267 268 269 270 271

6020

Nsil

Swal

5' GCAAAAAGTCTGGATGCATATTTAGAAACATCAAAAATTTAAATGATATTGAAGCAGTTGAATTAGCAA
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————— BGT41_T01c3 —————
 ————— BGT41_T01a3b-pDONR221 —————

A K K S G C I F R N I K N L N D I E A V E L A
 272 273 274 275 276 277 278 279 280 281 282 283 284 285 286 287 288 289 290 291 292 293 294

6090

5' GAAGTGATCAGTTAGATATTGCAGTAGATCTCATGGGATATAACCAGACACAATAGAATGCCTATATTCTC
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————— BGT41_T01c3 —————
 ————— BGT41_T01a3b-pDONR221 —————

R S D Q L D I A V D L M G Y T R H N R M P I F S
 295 296 297 298 299 300 301 302 303 304 305 306 307 308 309 310 311 312 313 314 315 316 317 318

6160

BGT41_T01a3b-pET32-DEST

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————— BGT41_T01c3 —————

————— BGT41_T01a3b-pDONR221 —————

K S K E T S P L F N S K L F T Q D L E N I Y L

552 553 554 555 556 557 558 559 560 561 562 563 564 565 566 567 568 569 570 571 572 573 574

6930

5' ATCTGGTAAAAAATAGGACCCAGCTTTCTTGTACAAAGTGGTTGATatcggatccgaattcgagctccg
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 3' TAGACCATTTTTTATCCTGGGTCGAAAGAACATGTTTCACCAACTatagcctaggcttaagctcgaggc

————— BGT41_T01c3 —————

————— attB2 —————

————— BGT41_T01a3b-pDONR221 —————

D L V K K .

575 576 577 578 579 580

7000

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Sall NotI EagI PspXI XhoI Aval BmeT110I

5' aaaggaagctgagttggctgctgccaccgctgagcaataactagcataacccttggggcctctaaacgg
 0
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————— T7 Terminator —————

7070

7140

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 0
 3' cagaactccccaaaaaacgactttcctccttgatataggccta

————— T7 Terminator —————

7183