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Dralll
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280

AloI'
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AloI
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350

Psil
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630

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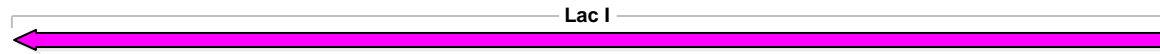
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BGH30_B01b-pET32-DEST

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o		
	FspAI	
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o	++++ ++++ ++++ ++++ ++++ ++++ ++++ ++++ ++++ ++++ ++++ ++++	3640
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o		
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XbaI



5180

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BamHI

BamHI'

Trx Tag

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5250

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o
o

RsrII

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

5320

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o
o
o

Trx Tag

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5390

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o
o
o

Trx Tag

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5460

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o
o
o

Trx Tag

G E V A A T K V G A L S K G Q L K E F L D A N L
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5530

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His6-tag
Thrombin

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5600

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BstBI
BgIII
Acc65I
KpnI

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

5670

5' acgacgacaaggccatggcgatATCAACAAGTTTGTACAAAAAAGCAGGCTctgaaaacttgtactttca
 3' tgctgctgttccggtaccgctaTAGTTGTTCAAACATGTTTTTTCGTCAGacttttgaacatgaaagt

Enterokinase
attB1
TEV site

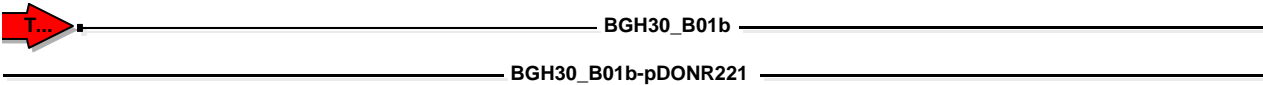
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5740

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' aggcACCAGGCCGTGGACACCGGCCGCAAAAAACCTTGCAGGTCTACACCACCGCCAAAGACACAAAA
 3' tccgTGGGTCCGGCACCTGTGGCCGGCGTTTTTTTGAACGTCCAGATGTGGTGGCGTTTTCTGTGTTTT

5810



G T Q A V D T G P Q K T L Q V Y T T A K D T K
 179 180 181 182 183 184 185 186 187 188 189 190 191 192 193 194 195 196 197 198 199 200 201

5' TTGCGTCTCACCTTGAGCGATAAACTCTCGCTCAAACCCGCGCAACAGGCGCCGAAACCGACACCGCCA
 3' AACGCAGAGTGGAACTCGCTATTTGAGAGCGAGTTTGGGCGCGTTGTCCGCGGCCTTTGGCTGTGGCGGT

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5880



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

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5' TTTTCGTCAACCCGACAAAGCGCTACCAAACCCCTGCTCGGTATTGGCGGCGCTATCACCGATGCCAGCGC
 3' AAAAGCAGTTGGGCCTGTTTCGCGATGGTTTGGGACGAGCCATAACCGCCGCGATAGTGGCTACGGTTCGCG

5950

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

5' CGAGGTGTATGCAACACTCGATCAAAAAACGCAGCGGAATTTATGCAGGCTTACTATGACCAGGACAAG
 3' GCTCCACATACGTTGTGAGCTAGTTTTTTGCGTCGCCCTTAAATACGTCCGAATGATACTGGTCCTGTTC

6020

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

5' GGTCTGGGCTACAGCCTGCTGCGCACCACCATCCACAGCTCCGACTTTGCCAGCAGCAGCTACACCTATA
 3' CCAGACCCGATGTCGGACGACGCGTGGTGGTAGGTGTCGAGGCTGAAACGGTCGTCGATGTGGATAT

6090

BGH30_B01b

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G L G Y S L L R T T I H S S D F A S S S Y T Y
 272 273 274 275 276 277 278 279 280 281 282 283 284 285 286 287 288 289 290 291 292 293 294

5' TCGAAGAGGGCGACAAAGCGCTTAACACCTTTGATATAACGCCGATAAAAAACACCGCATCCCCATGAT
 3' AGCTTCTCCCGCTGTTTCGCGAATTGTGGAACTATATTGCGGCCATTTTTTTGTGGCGTAGGGGTACTA

6160

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

SfiI

5' CAAGCAGGCCATCCGGGCCGCGGCGGCAGCATCACCACCTACGCCAGCCCTGGAGCGCACCGGCGTTT
 3' GTTCGTCCGGTAGGCCCGGCGGCCGCGTCTAGTGGTGGATGCGGTTCGGGACCTCGCGTGGCCGCAA

6230

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K Q A I R A A G G S I T T Y A S P W S A P A F
 319 320 321 322 323 324 325 326 327 328 329 330 331 332 333 334 335 336 337 338 339 340 341

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5' ATGAAAGACACCAACAATATGCTGCAAGGCGGCAAGCTGCTGCCCGAATACCGCGACACCTGGGCCACCT
 0 ++++++
 3' TACTTTCTGTGGTTGTTATACGACGTTCCGCCGTTTCGACGACGGGCTTATGGCGCTGTGGACCCGGTGA

6300

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M K D T N N M L Q G G K L L P E Y R D T W A T
 342 343 344 345 346 347 348 349 350 351 352 353 354 355 356 357 358 359 360 361 362 363 364

5' ACTTCACCAAATTTATCCGCGCCTATGAGCAGGAAGGCATTCCGATTTGGGGGATTACCGTACAAAACGA
 0 ++++++
 3' TGAAGTGGTTTAAATAGGCGCGGATACTCGTCCTTCCGTAAGGCTAAACCCCTAATGGCATGTTTTGCT

6370

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Y F T K F I R A Y E Q E G I P I W G I T V Q N E
 365 366 367 368 369 370 371 372 373 374 375 376 377 378 379 380 381 382 383 384 385 386 387 388

SacII

5' ACCCATGGCCAAGCAAACCTGGGAATCGATGATTTTCACCGCGGAAGAAGAGCGCGATTTCTCAAAAAC
 0 ++++++
 3' TGGGTACCGGTTTCGTTTGGACCCTTAGCTACTAAAAGTGGCGCCTTCTTCTCGCGCTAAAGGAGTTTTTG

6440

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P M A K Q T W E S M I F T A E E E R D F L K N
 389 390 391 392 393 394 395 396 397 398 399 400 401 402 403 404 405 406 407 408 409 410 411

5' CACCTGGGCCCCGATCATGCACACATCCGGCCTGGGCGATAAAAAATCATCGTGTGGGATCACAACCGCG
 0 ++++++
 3' GTGGACCCGGGCTAGTACGTGTGTAGGCCGACCCGCTATTTTTTTAGTAGCACACCCTAGTGTGGCGC

6510

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H L G P I M H T S G L G D K K I I V W D H N R
 412 413 414 415 416 417 418 419 420 421 422 423 424 425 426 427 428 429 430 431 432 433 434

BbvCI

5' ACCTGATCACGCACCGCGCCAACACCATCCTCAGCGATCCCGAAGCGGCTAAATATGTCTGGGGCGTGGG
 0 ++++++
 3' TGGACTAGTGCGTGGCGCGGTTGTGGTAGGAGTCGCTAGGGCTTCGCCGATTTATACAGACCCCGCACCC

6580

BGH30_B01b

BGH30_B01b-pDONR221

D L I T H R A N T I L S D P E A A K Y V W G V G
 435 436 437 438 439 440 441 442 443 444 445 446 447 448 449 450 451 452 453 454 455 456 457 458

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5' CTTCCACTGGTACGAAACCTGGACCGGCGGCCAACCCATGTTTCGGCAACGTGGCCGAAGTCAAGCAGGCA
 6650
 3' GAAGGTGACCATGCTTTGGACCTGGCCCGGTTGGGTACAAGCCGTTGCACCGGCTTCAGTTCGTCCGT

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F H W Y E T W T G G Q P M F G N V A E V K Q A
 459 460 461 462 463 464 465 466 467 468 469 470 471 472 473 474 475 476 477 478 479 480 481

5' TTCCCGCATATCAACCTGCTGCTCACCGAAGCCACGGTGGAAAAATTTGATGCGGCGCGCTACCACCCT
 6720
 3' AAGGGCGTATAGTTGGACGACGAGTGGCTTCGGTGCACCTTTTTAACTACGCCGCGCGATGGTGGTGA

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F P H I N L L L T E A T V E K F D A A R Y H H
 482 483 484 485 486 487 488 489 490 491 492 493 494 495 496 497 498 499 500 501 502 503 504

5' GGCCCAATGCCGAACGCTACGGCAACTCCATCGTCAATGACTTCAACGCCGGCGGCGAGGCTGGACCGA
 6790
 3' CCGGGTTACGGCTTGCGATGCCGTTGAGGTAGCAGTTACTGAAGTTGCGGCCGCGCCGTCCGACCTGGCT

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W P N A E R Y G N S I V N D F N A G A A G W T D
 505 506 507 508 509 510 511 512 513 514 515 516 517 518 519 520 521 522 523 524 525 526 527 528

5' CTGGAATATGCTGCTGGACGATAAAGGCGGCCCAACCATGTCAGCAATTTCTGCTTCGCCCCATTAC
 6860
 3' GACCTTATACGACGACCTGCTATTTCCGCCGGGTTGGTACAGTCGTTAAAGACGAAGCGGGGTAAGTG

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W N M L L D D K G G P N H V S N F C F A P I H
 529 530 531 532 533 534 535 536 537 538 539 540 541 542 543 544 545 546 547 548 549 550 551

5' GCCGACAGCAAAACCGGCAAGCTGATTTACACCCGACCTATTACTACCTCGGCCACTTCTCGCGCTTTA
 6930
 3' CGGCTGTCGTTTTGGCCGTTGACTAAATGTGGGGCTGGATAATGATGGAGCCGTTGAAGAGCGCGAAAT

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BGH30_B01b-pDONR221

A D S K T G K L I Y T P T Y Y Y L G H F S R F
 552 553 554 555 556 557 558 559 560 561 562 563 564 565 566 567 568 569 570 571 572 573 574

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5' TCCGCCCTGAGGCGCAGCGGGTTCAGTGCGGTCAGCAGCCGCAGCGTGCTGCAAACCACGTCGTTTATCAA
 3' AGGCGGGACTCCGCGTCGCCAGTCACGCCAGTCGTCGGCGTCGCACGACGTTTGGTGCAGCAAATAGTT

7000

Bsu36I

BmgBI

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I R P E A Q R V S A V S S R S V L Q T T S F I N
 575 576 577 578 579 580 581 582 583 584 585 586 587 588 589 590 591 592 593 594 595 596 597 598

5' CGCCGATAACCAACTGGCAACGGTGGTCTTGAATTTAAGCGATGCCGCCGTGACCTATCGCTTTTATATA
 3' GCGGCTATTGGTTGACCGTTGCCACCAGAACTTAAATTCGCTACGCGGCACCTGGATAGCGAAAATATAT

7070

BGH30_B01b

BGH30_B01b-pDONR221

A D N Q L A T V V L N L S D A A V T Y R F Y I
 599 600 601 602 603 604 605 606 607 608 609 610 611 612 613 614 615 616 617 618 619 620 621

5' GGCACACAGGAAACCACCGTTACTATCCCCGCGCGCCATCCAAACGCTGGTGTATTAGGACCCAGCTT
 3' CCGTGTGTCCTTGGTGGCAATGATAGGGGCGCGCGGTAGGTTTGCACCACATAATCCTGGGTCGAA

7140

MauBI

BGH30_B01b

attB2

BGH30_B01b-pDONR221

G T Q E T T V T I P A R A I Q T L V Y
 622 623 624 625 626 627 628 629 630 631 632 633 634 635 636 637 638 639 640 641

5' TCTTGTACAAAGTGGTTGATatcggatccgaattcgagctccgctcgacaagcttgcggccgactcgagc
 3' AGAACATGTTTCACCAACTAtagcctaggcttaagctcgaggcagctgttcgaacgccggcgtgagctcg

7210

attB2

BamHI

EcoRI

EcoICRI

SacI

Sall

HindIII

NotI

EagI

PspXI

XhoI

AvaI

BmeT110I

5' accaccaccaccactgagatccggctgctaacaaagcccgaaaggaagctgagttggctgctgccac
 3' tgggtggtggtggtggtgactctaggccgacgattgtttcgggctttccttcgactcaaccgacgacggtg

7280

B1pI

5' cgctgagcaataactagcataacccttggggcctctaaacgggtcttgaggggttttttgctgaaagga
 3' gcgactcgttattgatcgattgggggaaccccgagatttgcccagaactccccaaaaaacgactttcct

7350

T7 Terminator

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5' ggaactatatccggat
o +++++|++++|++++|+
3' ccttgatataggccta
o

7366