

# pDRIVE-hUbiquitin B

A plasmid with the native human Ubiquitin B promoter

Catalog # pdrive-hubiquitinb

For research use only

Version # 02I18-SV

## PRODUCT INFORMATION

### Content:

- 1 disk of lyophilized GT100 *E. coli* bacteria transformed by pDRIVE-hUbiquitin B.
- GT100 genotype is: *F*-, *mcrA*,  $\Delta$ (*mrr-hsdRMS-mcrBC*),  $\Delta$ *80lacZ* $\Delta$ *M15*,  $\Delta$ *MacX74*, *recA1*, *endA1*.
- 4 pouches of *E. coli* FastMedia™ Zeo

### Shipping and storage:

- Products are shipped at room temperature.
- Transformed bacteria should be stored at -20°C. Bacteria are stable up to one year when properly stored.
- Store *E. coli* FastMedia™ Zeo at room temperature. FastMedia™ is stable 18 months when stored properly.

### Quality control:

- Plasmid construct has been confirmed by restriction analysis and sequencing.
- Bacteria have been lyophilized, and their viability upon resuspension has been verified.
- Promoter activity has been confirmed by transient transfection of 293 cells as well as other selected cell lines.

## GENERAL PRODUCT USE

pDRIVE is an expression plasmid containing a native or composite promoter of interest. pDRIVE may be used to:

- **Subclone a promoter of interest into another vector.** Unique restriction sites are present at each end of the promoter allowing convenient excision. The 5' sites include *Sda* I, *Pst* I, and *Spe* I. *Sda* I is compatible with *Nsi* I and *Pst* I. *Spe* I is compatible with *Avr* II, *Nhe* I and *Xba* I. The 3' restriction site is *Nco* I which includes the ATG start codon, and is compatible with *BspH* I and *BspLU11* I.
- **Compare the activity of different promoters** in transient transfection experiments. Each pDRIVE promoter drives the expression of the *LacZ* reporter gene which allows for testing of the promoter's activity in transient transfection experiments. Furthermore, the *LacZ* gene is flanked by unique restriction sites (*Nco* I and *EcoR* I) for easy replacement with a different gene of interest.

## PROMOTER CHARACTERISTICS

Element	Name	Origin	Size bp
Promoter	Ubiquitin B	Human	300
5'UTR	Ubiquitin B	Human	793*
Intron*	Ubiquitin B	Human	701

\*Intron is contained within the 5' UTR. ^Size of 5'UTR without intron is 92 bp.

### hUbiquitin B promoter

Ubiquitin B is a small highly conserved protein that is abundantly expressed in all eukaryotic cells. Ubiquitin B is involved in the degradation of short-lived regulatory proteins as well as abnormal/mutated proteins and in the processing of major histocompatibility class I-restricted antigens<sup>1</sup>. A 1.1 kb fragment from the ubiquitin B gene was shown to display sustained expression of a transgene *in vitro* and *in vivo*. Twenty days post-injection, the levels of expression were similar to those obtained with the CMV promoter and were higher 35 days post-injection<sup>2</sup>.

## PLASMID FEATURES

- **LacZ gene** encodes  $\beta$ -galactosidase an enzyme that catalyzes the hydrolysis of X-Gal, producing a blue precipitate that can be easily visualized under a microscope.
  - **SV40 pAn:** The Simian Virus 40 late polyadenylation signal enables efficient cleavage and polyadenylation reactions resulting in high levels of steady-state mRNA.
  - **pMB1 Ori** is a minimal *E. coli* origin of replication with the same activity as the longer Ori.
  - **EM7** is a bacterial promoter that enables the constitutive expression of the antibiotic resistance gene in *E. coli*.
  - **Sh ble** gene confers zeocin resistance therefore allowing the selection of transformed *E. coli* carrying a pDRIVE plasmid.
- Note: Stable transfection of clones cannot be performed due to the absence of an eukaryotic promoter upstream of the Sh ble gene.*

## METHODS

### Growth of pDRIVE-transformed bacteria:

Use sterile conditions to do the following:

- 1- Resuspend the lyophilized *E. coli* by adding 1 ml of LB medium in the tube containing the disk. Let sit for 5 minutes. Mix gently by inverting the tube several times.
- 2- Streak bacteria taken from this suspension on a zeocin LB agar plate prepared with the *E. coli* FastMedia™ Zeo agar provided (see below).
- 3- Place the plate in an incubator at 37°C overnight.
- 4- Isolate a single colony and grow the bacteria in TB supplemented with zeocin using the FastMedia™ Zeo liquid provided (see below).
- 5- Extract the pDRIVE plasmid DNA using the method of your choice.

### Selection of bacteria with *E. coli* FastMedia™ Zeo:

*E. coli* FastMedia™ Zeo is a **new, fast and convenient** way to prepare liquid and solid media for bacterial culture by using only a microwave. *E. coli* FastMedia™ Zeo is a TB (liquid) or LB (solid) based medium with zeocin, and contains stabilizers.

*E. coli* FastMedia™ Zeo can be ordered separately (catalog code # fas-zn-1, fas-zn-s).

### Method:

- 1- Pour the contents of a pouch into a clean borosilicate glass bottle or flask.
- 2- Add 200 ml of distilled water to the flask
- 3- Heat in a microwave on MEDIUM power setting (about 400Watts), until bubbles start appearing (approximately 3 minutes). **Do not heat a closed container. Do not autoclave FastMedia™.**
- 4- Swirl gently to mix the preparation. **Be careful, the bottle and media are hot, use heatproof pads or gloves and care when handling.**
- 5- Reheat the media for 30 seconds and gently swirl again. Repeat as necessary to completely dissolve the powder into solution. But be careful to avoid overboiling and volume loss.
- 6- Let agar medium cool to 45°C before pouring plates. Let liquid media cool to 37°C before seeding bacteria.

*Note: Do not reheat solidified FastMedia™ as the antibiotic will be permanently destroyed by the procedure.*

### References:

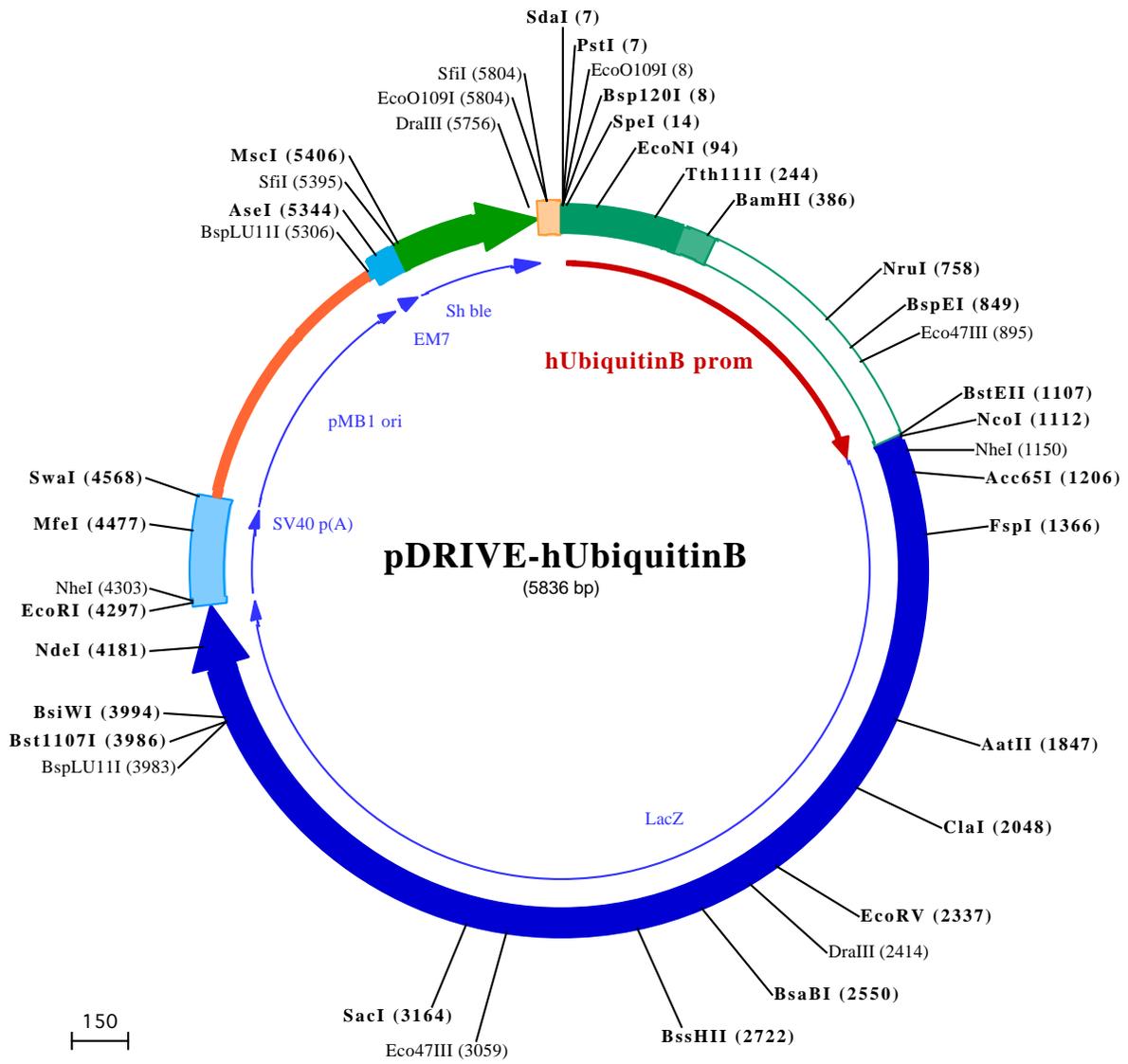
1. Ciechanover A. and Schwartz AL. (1998) Proc Natl Acad Sci USA 95(6):2727-30.
2. Yew NS *et al.* (2001) Mol Ther. 4(1):75-82.

## TECHNICAL SUPPORT

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Bsp120I (8)  
EcoO109I (8)  
PstI (7)  
SdaI (7) SpeI (14) EcoNI (94)

1 CCTGCAGGGCCCACTAGTAAGTTTCCAGAGCTTTTCGAGGAAGGTTTCTTCAACTCAAATTCATCCGCTGATAATTTTCTTATTTTCTCTAAAGAAGGA

101 AGAGAAGCGCATAGAGGAGAAGGAAATAATTTTTTAGGAGCCTTTCTTACGGCTATGAGGAATTTGGGGCTCAGTTGAAAAGCCTAAACTGCCTCTCGG

Tth111I (244)

201 GAGGTTGGGCGCGGCGAACTACTTTTCAGCGGCGCACGGAGACGGCTCTACGTGAGGGGTGATAAGTGACGCAACACTCGTTGCATAAATTTGCGCTCCG

BamHI (386)

301 CCAGCCCGAGCATTAGGGGCGGTTGGCTTTGTTGGGTGAGCTTGTGTCCCTGTGGGTGGACGTGGTTGGTGATTGGCAGGATCCTGGTATCCGC

401 TACAGgtactggccacagccgtaaaagacctgccccggcgtagagggggaatgggtgaggtcaagctggaggcttcttgggggtgggtgggcccgctg

500 aggggaggggagggcgaggtgacgcgacacccggcctttctgggagagtgggccttgttgacctaaagggggcgagggcagttggcacgcgcacgcgc

598 cgacagaactaacagacat taaccaacagcgattccgtcgegttaacttgggaggaaggcgaaaagaggtagttgtgtggcttctggaacccta

NruI (758)

696 aatttggaaatcccagtatgagaat ggtgt cccttctgtgtttcaatgggatttttacttcgagctcttgggttgggtttgtttcagtttgctt

BspEI (849) Eco47III (895)

795 aacaccgtgcttaggtttgaggcagattggagttcggtcgggggagttgaaatccgggaacagttagtggggaaagctgtggacgcttggtaagagagc

895 gctctggatcttccgctgttgacgttgaaccttgaatgacgaatttcgtattaagtgacttagccttgaattgaggggaggttgcggaatattaa

995 cgtatattaaggcattttgaaggaatagtgctaattttgaagaatattaggtgtaaaagcaagaatacaatgatcctgaggtgacacgcttatgtttta

NcoI (1112)  
BstEII (1107) NheI (1150)

1095 ctttttaactagGTACCATGGGGGTTCTCATCATCATCATCATGGTATGGCTAGCATGACTGGTGACAGCAAAATGGGTCGGATCTGTACGACG

1 MetGlyGlySerHisHisHisHisHisHisGlyMetAlaSerMetThrGlyGlyGlnGlnMetGlyArgAspLeuTyrAspA

Acc65I (1206)

1195 ATGACGATAAGGTACCTAAGGATCAGCTTGGAGTTGATCCCGTCGTTTTACAACGTCGTGACTGGGAAAACCTGGCGTTACCCAACCTAATCGCCTTGC

28 spAspAspLysValProLysAspGlnLeuGlyValAspProValValLeuGlnArgArgAspTrpGluAsnProGlyValThrGlnLeuAsnArgLeuAl

FspI (1366)

1295 AGCACATCCCCCTTTCGCCAGCTGGCGTAATAGCGAAGAGGCCCGCACCGATCGCCCTTCCCAACAGTTGCGCAGCCTGAATGGCGAATGGCGCTTTGCC

61 aAlaHisProProPheAlaSerTrpArgAsnSerGluGluAlaArgThrAspArgProSerGlnGlnLeuArgSerLeuAsnGlyGluTrpArgPheAla

1395 TGGTTTCCGGCACCAGAAGCGGTGCGCGAAAGCTGGCTGGAGTGCAGTCTTCTGAGGCCGATACTGTCTGCTGCCCTCAAACCTGGCAGATGCACGGTT

95 TrpPheProAlaProGluAlaValProGluSerTrpLeuGluCysAspTrpLeuProGluAlaAspThrValValValProSerAsnTrpGlnMetHisGlyT

1495 ACGATGGCCCATCTACACCAACGTAACCTATCCATTACGGTCAATCCGCGTTTTGTTCCACGGAGAATCCGACGGGTTGTTACTGCTCACATTTAA

128 yrAspAlaProl leTyrThrAsnValThrTyrProl leThrValAsnProProPheValProThrGluAsnProThrGlyCysTyrSerLeuThrPheAs

1595 TGTGATGAAAGCTGGCTACAGGAAGGCCAGACGCGAATTATTTTTGATGGCGTAACTCGCGTTTTATCTGTGGTGAACGGGCGCTGGGTGGTTAC

161 nValAspGluSerTrpLeuGlnGluGlyGlnThrArgI leI lePheAspGlyValAsnSerAlaPheHisLeuTrpCysAsnGlyArgTrpValGlyTyr

1695 GGCAGGACAGCTGTTGCGCTCTGAATTTGACCTGAGCGCATTTTTACGCGCCGGAGAAAACCGCCTCGCGGTGATGGTGTGCGTTGGAGTGACGGCA

195 GlyGlnAspSerArgLeuProSerGluPheAspLeuSerAlaPheLeuArgAlaGlyLeuAsnArgLeuAlaValMetValLeuArgTrpSerAspGlyS

AatII (1847)

1795 GTTATCTGGAAGATCAGGATATGTGGCGGATGAGCGGCATTTTCCGTGACGTCCTGTTGCTGCATAAACCGACTACACAAATCAGCGATTTCCATGTTGC

228 erTyrLeuGluAspGlnAspMetTrpArgMetSerGlyI lePheArgAspValSerLeuHisLysProThrThrGlnI leSerAspPheHisValAl

1895 CACTCGCTTAATGATGATTTACAGCCGCTGTACTGGAGCTGAAGTTGAGATGTGCGGCAAGTTGCGTACTACCTACCGGTAACAGPTTCTTTATGG

261 aThrArgPheAsnAspPheSerArgAlaValLeuGluAlaGluValGlnMetCysGlyGluLeuArgAspTyrLeuArgValThrValSerLeuTrp

ClaI (2048)

1995 CAGGGTAAACGCAGGTGCCAGCGCACCGCCTTTCCGCGGTGAAATATCGATGAGCGTGGTGGTTATGCCGATCGCGTCACACTACGCTGAACG

295 GlnGlyGluThrGlnValAlaSerGlyThrAlaProPheGlyGlyGluI leI leAspGluArgGlyGlyTyrAlaAspArgValThrLeuArgLeuAsnV

2095 TCGAAAACCCGAAACTGTGGAGCGCCGAAATCCCGAATCTCTATCGTGGCGGTTGAACTGCACACCGCCGACGCGCTGATTGAAGCAGAAGCCTG

328 alGluAsnProLysLeuTrpSerAlaGluI leProAsnLeuTyrArgAlaValValGluLeuHisThrAlaAspGlyThrLeuI leGluAlaGluAlaCy

2195 CGATGTCGGTTTCCGCGAGGTGCGGATTGAAAATGGTCTGCTGCTGTAACGGCAAGCCGTTGCTGATTTCGAGGCGTTAACCGTCACGAGCATCATCT

361 sAspValGlyPheArgGluValArgI leGluAsnGlyLeuLeuLeuAsnGlyLysProLeuLeuI leArgGlyValAsnArgHisGluHisHisPro

EcoRV (2337)

2295 CTGCATGGTCAGGTCATGGATGAGCAGACGATGGTGCAGGATATCTGCTGATGAAGCAGAACAACCTTAAACGCCGTGCGCTGTTTCGATTATCCGAACC

395 LeuHisGlyGlnValMetAspGluGlnThrMetValGlnAspI leLeuLeuMetLysGlnAsnAsnPheAsnAlaValArgCysSerHisTyrProAsnH

DraIII (2414)

2395 ATCCGCTGTTGATACAGCTGTGCGACCGCTACGGCCTGTATGTTGGTGAAGCCAATATTGAAACCCACGGCATGGTCCAATGAATCGTCTGACCGA

428 isProLeuTrpTyrThrLeuCysAspArgTyrGlyLeuTyrValValAspGluAlaAsnI leGluThrHisGlyMetValProMetAsnArgLeuThrAs

BsaBI (2550)

2495 TGATCCGCGCTGGCTACCGCGATGAGCGAACCGTAACCGGAATGGTGCAGCGGATCGTAATCACCCGAGTGTGATCATCTGGTCTGGGGAATGAA

461 pAspProArgTrpLeuProAlaMetSerGluArgValThrArgMetValGlnArgAspArgAsnHisProSerValI leI leTrpSerLeuGlyAsnGlu

2595 TCAGGCCACGGCGCTAATCACGACGCGCTGTATCGCTGGATCAAATCTGTCGATCCTTCCGCGCGGTGCAGTATGAAGCGCGGAGCCGACACCACGG

495 SerGlyHisGlyAlaAsnHisAspAlaLeuTyrArgTrpI leLysSerValAspProSerArgProValGlnTyrGluGlyGlyAlaAspThrThrA

BssHIII (2722)

2695 CCACCGATATTATTTGCCGATGTACGCGCGGTGGATGAAGACCAGCCCTTCCCGCTGTGCCGAAATGGTCCATCAAAAAATGGCTTTCGCTACCTGG

528 laThrAspI leI leCysProMetTyrAlaArgValAspGluAspGlnProPheProAlaValProLysTrpSerI leLysLysTrpLeuSerLeuProG

2795 AGAGACGCGCCGCTGATCCTTTGCGAATACGCCACGCGATGGGTAAACAGTCTTGGCGGTTTCGCTAAATACTGGCAGGCGTTTCGTCAGTATCCCGT

561 yGluThrArgProLeuI leLeuCysGluTyrAlaHisAlaMetGlyAsnSerLeuGlyGlyPheAlaLysTyrTrpGlnAlaPheGlnTyrArg

2895 TTACAGGGCGGCTTCGTCTGGACTGGGTGGATCAGTCGCTGATTAATATGATGAAAACGGCAACCCGTGGTCCGGCTTACGGCGGTGATTTTGGCGATA  
595▶ LeuGlnGlyGlyPheValTrpAspTrpValAspGlnSerLeuI leLysTyrAspGluAsnGlyAsnProTrpSerAlaTyrGlyGlyAspPheGlyAspT  
Eco47III (3059)  
2995 CGCCGAACGATCGCCAGTTCTGTATGAACGGTCTGGTCTTTGCCGACCGCACGCCGCATCCAGCGCTGACGGAAAGCAAAAACACCAGCAGCAGTTTTTCCA  
628▶ hrProAsnAspArgGlnPheCysMetAsnGlyLeuValPheAlaAspArgThrProHisProAlaLeuThrGluAlaLysHisGlnGlnGlnPhePheG  
SacI (3164)  
3095 GTTCGGTTTATCCGGGCAAACCATCGAAGTGACCAGCGAATACCTGTTCCGTCATAGCGATAACGAGCTCCTGCACTGGATGGTGGCGCTGGATGGTAA  
661▶ nPheArgLeuSerGlyGlnThrI leGluValThrSerGluTyrLeuPheArgHisSerAspAsnGluLeuLeuHisTrpMetValAlaLeuAspGlyLys  
3195 CCGCTGGCAAGCGGTGAAGTGCCTCTGGATGTCGCTCCACAAGGTAACAGTTGATTGAACTGCCTGAACTACCGCAGCCGGAGAGCGCCGGCAACTCT  
695▶ ProLeuAlaSerGlyGluValProLeuAspValAlaProGlnGlyLysGlnLeuI leGluLeuValProGluLeuProGlnProGluSerAlaGlyGlnLeuT  
3295 GGCTCACAGTACGCGTAGTGCAACCGAACCGACCGCATGGTCAGAAGCCGGGCACATCAGCGCCTGGCAGCAGTGGCGTCTGGCGGAAAACCTCAGTGT  
728▶ rpLeuThrValArgValValGlnProAsnAlaThrAlaTrpSerGluAlaGlyHisI leSerAlaTrpGlnGlnTrpArgLeuAlaGluAsnLeuSerVa  
3395 GACGCTCCCCGCGCTCCACGCCATCCCGCATCTGACCACCAGCGAAATGGATTTTTGCATCGAGCTGGGTAATAAGCGTTGGCAATTTAACCGCCAG  
761▶ IThrLeuProAlaAlaSerHisAlaI leProHisLeuThrThrSerGluMetAspPheCysI leGluLeuGlyAsnLysArgTrpGlnPheAsnArgGln  
3495 TCAGGCTTTCTTTCACAGATGTGGATTGGCGATAAAAAACAACCTGCTGACCGCTGCGCGATCAGTTCACCCGTGCACCGCTGGATAACGACATTTGGCG  
795▶ SerGlyPheLeuSerGlnMetTrpI leGlyAspLysLysGlnLeuLeuThrProLeuArgAspGlnPheThrArgAlaProLeuAspAsnAspI leGlyV  
3595 TAAGTGAAGCGACCCGATTGACCTAACGCCTGGGTGCAACGCTGGAAGCGCGGGCCATTACCAGGCCAAGCAGCGTTGTTGCAGTGCACGGCAGA  
828▶ alSerGluAlaThrArgI leAspProAsnAlaTrpValGluArgTrpLysAlaAlaGlyHisTyrGlnAlaGluAlaAlaLeuLeuGlnCysThrAlaAs  
3695 TACACTTGCTGATGCGGTGCTGATTACGACCGCTCACGCTGGCAGCATCAGGGGAAAACCTTATTTATCAGCCGAAAACCTACCAGATTGATGGTAGT  
861▶ pThrLeuAlaAspAlaValLeuI leThrThrAlaHisAlaTrpGlnHisGlnGlyLysThrLeuPheI leSerArgLysThrTyrArgI leAspGlySer  
3795 GGTCAAATGGCGATTACCGTTGATGTTGAAGTGGCGAGCGATACCCGCATCCGGCGGATTGGCCTGAACTGCCAGCTGGCGCAGGTAGCAGAGCGGG  
895▶ GlyGlnMetAlaI leThrValAspValGluValAlaSerAspThrProHisProAlaArgI leGlyLeuAsnCysGlnLeuAlaGlnValAlaGluArgV  
Bst1107I (3986)  
3895 TAAACTGGCTCGGATTAGGGCCGAAGAAAATATCCCGACCGCCTTACTGCCGCTGTTTTGACCGCTGGGATCTGCCATTGTCAGACATGTATACCCC  
928▶ alAsnTrpLeuGlyLeuGlyProGlnGluAsnTyrProAspArgLeuThrAlaAlaCysPheAspArgTrpAspLeuProLeuSerAspMetTyrThrPr  
3995 GTACGCTTCCCGAGCGAAAACGGTCTGCGCTGCGGGACGCGGAATTGAATTATGGCCACACCAGTGGCGCGGGACTTCCAGTTCAACATCAGCCGC  
961▶ oTyrValPheProSerGluAsnGlyLeuArgCysGlyThrArgGluLeuAsnTyrGlyProHisGlnTrpArgGlyAspPheGlnPheAsnI leSerArg  
NdeI (4181)  
4095 TACAGTCAACAGCAACTGATGGAACAGCCATCGCCATCTGCTGCACGCGGAAGAAGGCACATGGCTGAATATCGACGGTTTCCATATGGGAGATTGGTG  
995▶ TyrSerGlnGlnGlnLeuMetGluThrSerHisArgHisLeuLeuHisAlaGluGluGlyThrTrpLeuAsnI leAspGlyPheHisMetGlyI leGlyG  
4195 GCACGACTCCTGGAGCCCGTCAATCGCGGAAATACAGCTGAGCGCCGCTGCTACCATTACCAGTTGGTCTGGTGTCAAAAATAATAACTAGTGC  
1028▶ lyAspAspSerTrpSerProSerValSerAlaGluLeuGlnLeuSerAlaGlyArgTyrHisTyrGlnLeuValTrpCysGlnLys•••  
NheI (4303)  
EcoRI (4297)  
4295 AGAATTCGCTAGCTCGACATGATAAGATACATTGATGAGTTTGGACAACCCACAACCTAGAATGCAGTGAAAAAATGCTTTATTTGTGAAATTTGTGATG  
MfeI (4477)  
4395 CTATTGCTTTATTTGTGAAATTTGTGATGCTATTGCTTTATTTGTAACCATTATAAGCTGCAATAAACAAAGTTAACAAACAATTGCATTCTTTTATG  
SwaI (4568)  
4495 TTTCAAGTTTCAGGGGGAGGTGTGGGAGGTTTTTAAAGCAAGTAAAACCTCTACAAATGTGGTAGATCCATTTAAATGTTAATTAACCTAGCCATGACCAA  
4595 AATCCCTTAACGTGAGTTTTCTTCCACTGAGCGTCAGACCCGTAGAAAAGATCAAAGGATCTTCTTGAGATCCTTTTTTCTGCGCGTAATCTGCTGC  
4695 TTGCAAAACAAAAAACCCGCTACCAGCGGTGTTTTGTTTCCGGATCAAGAGCTACCAACTCTTTTTCCGAAGGTAAGTGGCTTACGAGAGCGCAGA  
4795 TACCAAATACTGTTCTTCTAGTGTAGCCGTAGTTAGGCCACCACTTCAAGAACTCTGTAGCACCGCCTACATACCTCGCTCTGCTAATCCTGTTACCAGT  
4895 GGCTGCTGCCAGTGGCGATAAGTCGTGCTTACCAGGTTGGACTCAAGACGATAGTTACCGGATAAGGCGCAGCGGTGCGGCTGAACGGGGGTTCTGTCG  
4995 ACACAGCCAGCTTGGAGCGAACGACCTACCCGAAGTACCTACAGCGTGGCTATGAGAAAGCGCCACGCTTCCGAAGGGAGAAAGCGGACA  
5095 GGTATCCGGTAAGCGCAGGGTGGAAACAGGAGAGCGCACGAGGGAGCTTCCAGGGGAAACGCCTGGTATCTTTATAGTCTGTGCGGTTTTGCCACCT  
5195 CTGACTTGAGCGTGCATTTTTGTGATGCTCGTCAAGGGGGCGGAGCCTATGGAAAAACGCCAGCAACCGCGCCTTTTTACGGTTCCTGGCCTTTTGTGCG  
BspLU11I (5306) AseI (5344)  
5295 CTTTTGCTCACTGTTCTTAATTAATTTTTCAAAGTAGTTGACAATTAATCATCGGCATAGTATATCGGCATAGTATAACGACTCACTATAGGAG  
SfiI (5395) MscI (5406)  
5395 GGCCATCATGGCCAAAGTTGACCAGTGTGTCCAGTGTCTCACAGCCAGGGATGTGGCTGGAGCTGTTGAGTTCTGGACTGACAGGTTGGGGTCTCCAGA  
1▶ MetAlaLysLeuThrSerAlaValProValLeuThrAlaArgAspValAlaGlyAlaValGluPheTrpThrAspArgLeuGlyPheSerArg  
5495 GATTTTGTGGAGGATGACTTTGCAAGTGTGGTCCAGAGATGATGTACCCTGTTTATCTCAGCAGTCCAGGACCAGGTGGTGCCTGACAACCCCTGGCTT  
32▶ AspPheValGluAspAspPheAlaGlyValValArgAspAspValThrLeuPheI leSerAlaValGlnAspGlnValValProAspAsnThrLeuAlaT  
5595 GGGTGTGGGTGAGAGGACTGGATGAGCTGATGCTGAGTGGAGTGGTGTCTCCACCACTTCCAGGATGCCAGTGGCCCTGCCATGACAGAGATTGG  
65▶ rpValTrpValArgGlyLeuAspGluLeuTyrAlaGluTrpSerGluValValSerThrAsnPheArgAspAlaSerGlyProAlaMetThrGluI leG  
DraIII (5756)  
5695 AGAGCAGCCTGGGGAGAGATTTCCCTGAGAGACCCAGCAGGCAACTGTGTGCACTTTGTGGCAGAGGAGCAGGACTGAGGATAAGAATTGAGTTTC  
98▶ yGluGlnProTrpGlyArgGluPheAlaLeuArgAspProAlaGlyAsnCysValHisPheValAlaGluGluGlnAsp•••  
SfiI (5804)  
EcoO109I (5804)  
5795 AGAAAAGGGGGCCTGAGTGGCCCTTTTTTCAACTTAATTA