

METHODS

Plasmid resuspension

Quickly spin the tube containing the lyophilized plasmid to pellet the DNA. To obtain a plasmid solution at 1 µg/µl, resuspend the DNA in 20 µl of sterile H₂O. Store resuspended plasmid at -20°C.

Plasmid amplification and cloning

Plasmid amplification and cloning can be performed in *E. coli* GT116 or other commonly used laboratory *E. coli* strains, such as DH5α.

Blasticidin usage

Blasticidin should be used at 25-100 µg/ml in bacteria and 1-30 µg/ml in mammalian cells. Blasticidin is supplied at 10 mg/ml in HEPES buffer.

References:

1. Kim, D.W. et al., 1990. Use of the human elongation factor 1 alpha promoter as a versatile and efficient expression system. *Gene* 2: 217-223. 2. Takebe, Y. et al., 1988. SR alpha promoter: an efficient and versatile mammalian cDNA expression system composed of the simian virus 40 early promoter and the R-U5 segment of human T-cell leukemia virus type 1 long terminal repeat. *Mol. Cell Biol.* 1: 466-472. 3. Carswell, S., & Alwine, J.C., 1989. Efficiency of utilization of the simian virus 40 late polyadenylation site: effects of upstream sequences. *Mol. Cell Biol.* 10: 4248-4258. 4. Yu J & Russell JE., 2001. Structural and functional analysis of an mRNP complex that mediates the high stability of human beta-globin mRNA. *Mol Cell Biol.* 21(17):5879-88.

RELATED PRODUCTS

Product	Catalog Code
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pSELECT-NHA-zeo	psetz-nha
pSELECT-CHA-zeo	psetz-cha
pSELECT-CHis-blasti	psetb-chis
pSELECT-NHis-zeo	psetz-nhis
pSELECT-CHis-zeo	psetz-chis

TECHNICAL SUPPORT

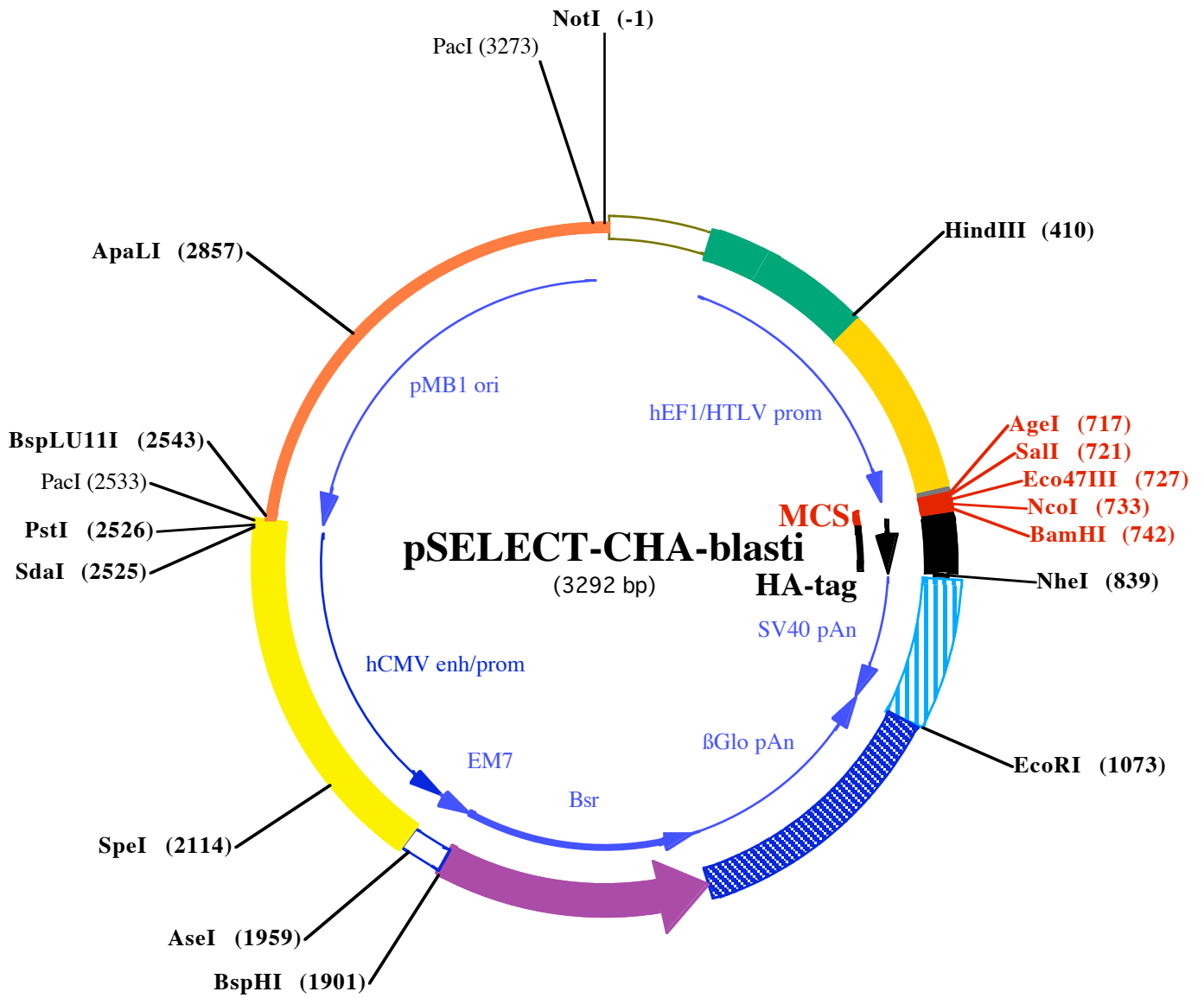
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NotI (-1)

1 CCGGCCGCAATAAAATATCTTTATTTTCATTACATCTGTGTGTTGGTTTTTTTGTGTGAATCGTAACTAACATACGCTCTCCATCAAACAAA
93 ACGAAACAAAACAACTAGCAAATAGGCTGTCCCCAGTGCAAGTGCAGGTGCCAGAACATTTCTCTATCGAAGGATCTGCGATCGCTCCGG
185 TGCCCGTCAGTGGGCAGAGCGCACATCGCCACAGTCCCCGAGAAGTTGGGGGAGGGGTCCGCAATTGAACGGGTGCCTAGAGAAGGTGGC
277 GCGGGGTAAACTGGGAAAGTGATGTCGTGTACTGGCTCCGCCTTTTCCCGAGGGTGGGGGAGAACCGTATATAAGTGCAGTAGTCCGCCGTG

HindIII (410)

369 AACGTTCTTTTTTCGCAACGGGTTTGCCGCCAGAACACAGCTGAAGCTTCGAGGGGCTCGCATCTCTCCTTCACGCGCCCGCCCTACCTG
461 AGGCCGCCATCCACGCCGGTTGAGTTCGCGTTCTGCCGCCCTCCCGCTGTGGTGCCTCCTGAACTGCGTCCGCCGTCTAGGTAAGTTTAAAGC
553 TCAGGTCGAGACCGGGCCTTTGTCCGGCGCTCCCTTGGAGCCTACCTAGACTCAGCCGGCTCTCCACGCTTTGCCTGACCCTGCTTGCTCAA

**SalI (721) NeoI (735+)
AgeI (717) Eco47III (727)**

645 CTCTACGCTTTTGTTCGTTTTCTGTCTGCGCCGTTACAGATCCAAGCTGTGACCGGCGCCTACCTGAGATCACCGGTCGACAGCGCTCCA

BamHI (742)

737 TGGCTCGGATCCATCCCTATGATGTGCCAGACTATGCTGGCTATCCATATGATGTTCTGATTATGCTGGATACCCTTATGATGTGCCAGA
1▶ GlySerTyrProTyrAspValProAspTyrAlaGlyTyrProTyrAspValProAspTyrAlaGlyTyrProTyrAspValProAs

NheI (839)

829 CTATGCCATAAGCTAGCTGGCCAGACATGATAAGATACATTGATGAGTTTGGACAAACCACAACCTAGAATGCAGTGAAAAAATGCTTTTATT
29▶ pTyrAla...

EcoRI (1073)

1013 GGTTCAAGGGGAGGTGTGGGAGGTTTTTTAAAGCAAGTAAAACCTCTACAAATGTGGTATGGAATCTAAAATACAGCATAGCAAACCTTTA
1105 ACCTCAAATCAAGCCTCTACTTGAATCCTTTTCTGAGGGATGAATAAGGCATAGGCATCAGGGGCTGTTGCCAATGTGCATTAGCTGTTTG
1197 CAGCCTCACCTTCTTTCATGGAGTTTAAAGATATAGTGTATTTTCCCAAGGTTTGAAGTACTGCTCTTCATTTCTTTATGTTTTAAATGCACTGA
1289 CCTCCACATTCCTTTTTTAGTAAAATATTAGAAATAATTTAAATACATCATTGCAATGAAAATAAATGTTTTTTATTAGGCAGAATCCAG
1381 ATGCTCAAGGCCCTTCATAATATCCCCAGTTTAGTAGTTGGACTTAGGGAACAAAGGAACCTTTAATAGAAATTGGACAGCAAGAAAGCGA
1473 GCTTCTAGCTTTAGTTCCCTGGTGTACTTGAGGGGGATGAGTTCCCAATGGTGGTTTTGACCAGCTTGCCATTTCATCTCAATGAGCACAAAAG

141▶ ...AsnArgThrTyrLysLeuProIleLeuGluGluIleThrThrLysValLeuLysGlyAsnMetGluIleLeuValPheC

1565 CAGTCAGGAGCATAGTCAGAGATGAGCTCTCTGCACATGCCACAGGGGCTGACCACCTGATGGATCTGTCCACCTCATCAGAGTAGGGGTG

113▶ ysAspProAlaTyrAspSerIleLeuGluArgCysMetGlyCysProSerValValArgIleSerArgAspValGluAspSerTyrProHis
1657 CCTGACAGCCACAATGGTGTCAAAGTCTTCTGCCCGTTGCTCACAGCAGACCCAATGGCAATGGCTTCAGCACAGACAGTGACCCTGCCAA

83▶ ArgValAlaValIleThrAspPheAspLysGlnGlyAsnSerValAlaSerGlyIleAlaIleAlaGluAlaCysValThrValArgGlyIle
1749 TGTAGGCTCAATGTGGACAGCAGAGATGATCTCCCGCTTGGTCTGATGGCCGCCCGACATGGTGTCTGTGTCTCATAGAGCATG

52▶ eTyrAlaGluIleHisValAlaSerIleIleGluGlyThrLysThrArgIleAlaAlaGlyValHisHisLysAsnAspGluTyrLeuMetT
1841 GTGATCTTCTCAGTGGCGACCTCCACCAGCTCCAGATCTGCTGAGAGATGTTGAAGGTCTTCATGATGGCCCTCTATAGTGCATCGTATT

BspHI (1901)

21▶ hrIleLysGluThrAlaValGluValLeuGluLeuAspGlnGlnSerIleAsnPheThrLysMet

AseI (1959)

1933 ATACTATGCCGATATACTATGCCGATGATTAATTGTCAAACAGCGTGGATGGCGTCTCCAGCTTATCTGACGGTTCACTAAACGAGCTCTG

2025 CTTATATAGACCTCCCACCGTACACGCCTACCGCCATTTGCGTCAATGGGGCGGAGTTGTTACGACATTTTGAAAGTCCCCTTGATTTAC

2117 TAGTCAAAACAAACTCCCATTTGACGTCAATGGGGTGGAGACTTGAAATCCCCGTGAGTCAAACCGCTATCCACGCCATTGATGTACTGCC

2209 AAAACCGCATCATCATGGTAATAGCGATGACTAATACGTAGATGTACTGCCAAGTAGGAAAGTCCCATAGGTTCATGTACTGGGCATAATGC

2301 CAGGCGGGCCATTTACCGTCATTGACGTCAATAGGGGGCGTACTTGGCATATGATACACTTGATGTACTGCCAAGTGGGCAGTTTACCGTAA

2393 ATACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACATACGTCAATTATTGACGTCAATGGGCGGGGTCTGTTGG

PacI (2533)
PstI (2526)
SdaI (2525) BspLU11I (2543)

2485 GCGGTCAGCCAGGCGGGCCATTTACCGTAAGTTATGTAACGCCCTGCAGGTTAATTAAGAACATGTGAGCAAAAGGCCAGCAAAAGGCCAGGA

2577 ACCGTAAAAGGCCCGCTTGCTGGCGTTTTTCCATAGGCTCCGCCCCCTGACGAGCATCACAAAATCGACGCTCAAGTCAGAGGTGGCGA

2669 AACCCGACAGGACTATAAAGATACCAGGCGTTTCCCCCTGGAAGCTCCCTCGTGCCTCTCCTGTTCCGACCCCTGCCGCTTACCGGATACCT

2761 GTCCGCTTTCTCCCTTCGGAAGCGTGGCGCTTTCTCATAGCTCACGCTGTAGGTATCTCAGTTCGGTGTAGGTGCTCGCTCCAAGCTGG

ApaLI (2857)

2853 GCTGTGTGCACGAACCCCCGTTTCAGCCCACCGCTGCGCTTATCCGGTAACTATCGTCTTGAGTCCAACCCGGTAAGACACGACTTATCG

2945 CCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGAGGTATGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCCTAACTACGGCTA

3037 CACTAGAAGAACAGTATTTGGTATCTGCGCTCTGCTGAAGCCAGTTACCTTCGGAAAAAGAGTTGGTAGCTCTTGATCCGGCAAACAAACCA

3129 CCGCTGGTAGCGGTGGTTTTTTTGTGTTGCAAGCAGCAGATTACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTTCTACGGGG

PacI (3273)

3221 TCTGACGCTCAGTGGAACGAAAACCTCACGTTAAGGGATTTTGGTCATGGCTAGTTAATTAACATTTAAATCA
