

Appendix A

DNA SEQUENCES AND PLASMID MAPS

TABLE OF CONTENTS

DNA and amino acid sequences

pEC28cyl-pher*	A-2
pEC2-RGD, RDG, and SC5	A-9
pEC2-SC5-ELF ₅ and ELF ₆	A-14

A.1 pETcyl-phes* plasmid information

Submitted by: Stacey Maskarinec, Marissa Mock

Date 4/22/04

Strain name: XL-1 blue (*SupE44 hsdR17 recA1 endA1 gyrA46 thi relA1 lac⁻*
F'*[proAB⁺ lacI^q lacZΔM15 Tn10(tet^r)]* (From Stratagene)) / **pET28cyl-phes***

Vector (kb): pET28cyl-phes* (7.0)

Cloning site: See plasmid map.

Construction of pET28cyl-phes* plasmid:

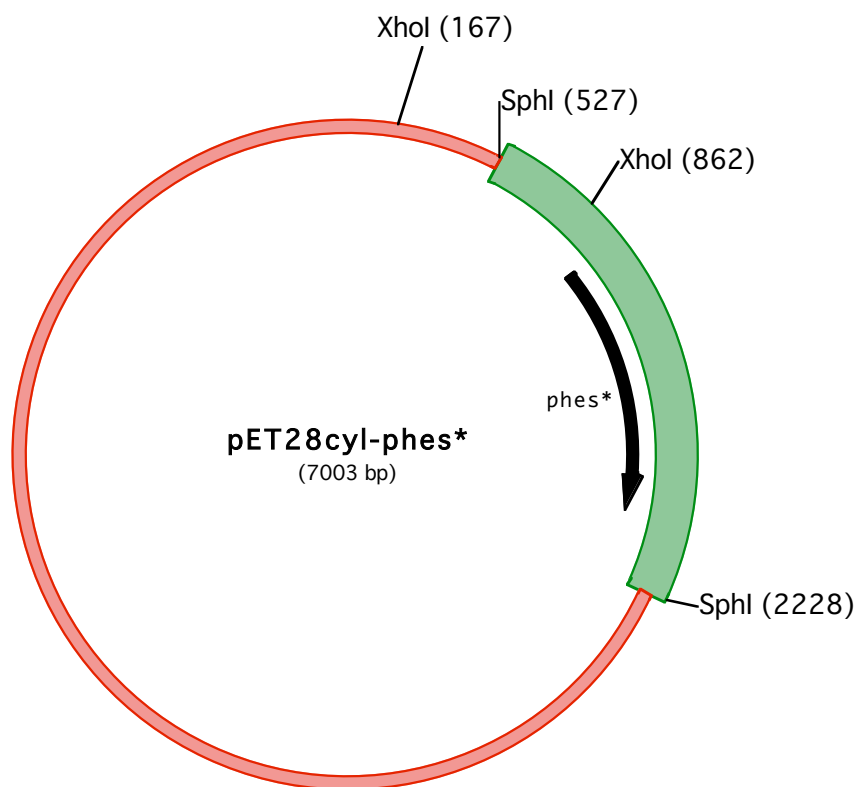
The phes* gene was cut from pUC19-FS (Nandita Sharma) with Sph I. It was ligated into the Sph I site of pET28cyl (Charles Liu).

Source available :

12 % Glycerol culture in Marissa-3 freezer box

Culture conditions: 2xYT, 37°C

A.1.1 Plasmid map



A.1.2 Position of elements

	<u>bp</u>
Vector size	7003
Full insert region	532-2233
PheRS* gene	992-1073

A.1.3 Full sequence (pET28cyl-phes*)

1 ATCCGGATATAGTTCCTCCTTTTCAGCAAAAAACCCCTCAAGACCCGTTTAGAGGCCCAAGGGGTTATGCTAGTTATT
TAGGCCTATATCAAGGAGGAAAGTCGTTTTTGGGGAGTTCTGGGCAAATCTCCGGGGTCCCAATACGATCAATAA
79 GCTCAGCGGTGGCAGCAGCCAACCTCAGCTTCCTTTTCGGGCTTTGTTAGCAGCCGGATCTCAGTGGTGGTGGTGGT
CGAGTCGCCACCGTCGTCGGTTGAGTCGAAGGAAAGCCCGAAACAATCGTCGGCCTAGAGTCACCACCACCACCACCA

XhoI (167)

157 GCTCGA ctt act cga gaa gct t CAT GTG GTG GTG GTG GTG ACC CAT TTG CTG TCC
CGAGCT gaa tga gct ctt cga a GTA CAC CAC CAC CAC CAC TGG GTA AAC GAC AGG
215 ACC AGT CAT GCT CGC CAT gGTATATCTCCTTCTTAAAGTTAAACAAAATTATTTCTAGAGGGGAATTGTTAT
TGG TCA GTA CGA GCG GTA cCATATAGAGGAAGAATTTCAATTTGTTTTAATAAAGATCTCCCCTTAACAATA
287 CCGCTCACAAATCCCCTATAGTGAGTCGTATTAATTTTCGGGGATCGAGATCTCGATCCTCTACGCCGACGCATCGT
GGCGAGTGTAAAGGGGATATCACTCAGCATAATTAAGCGCCCTAGCTCTAGAGCTAGGAGATGCGGCCTGCGTAGCA
365 GGCCGCGCATACCCGGCGCCACAGGTGCGGTTGCTGGCGCCTATATCGCCGACATCACCGATGGGGAAGATCGGGCTCG
CCGGCCGTAGTGGCCGGGTGCCACGCCAACGCCGGATATAGCGGCTGTAGTGGCTACCCCTTCTAGCCCGAGC
443 CCACTTCGGGCTCATGAGCGCTTGTTCGGCGTGGGTATGGTGGCAGGCCCCGTGGCCGGGGGACTGTTGGGCGCCAT
GGTGAAGCCCCGAGTACTCGGAACAAAGCCGCACCCATACCACCGTCCGGGGCACC GGCCCCCTGACAACCCGCGTA

SphI (527)

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599 ctccccgcgcttgccgattcattaatgcagctggcagcaggtttcccactggaagcgggcagtgagcgaac
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677 gcaattaatgtgagttagctcactcattaggcaccaggctttacactttatgcttccggctcgtatggtgtgga
cgtaattacactcaatcgagtgagtaatccgtgggtccgaaatgtgaaatacgaaggccgagcatacaacacact
755 attgtgagcggataacaatttcacacaggaacagctatgaccatgattacgccaagctcgaattaaccctcactaa
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XhoI (862)

833 agggaaacaaaagctggtaccgggccccccctcgaggtcgacggtatcgataagcttgatatcgaattccccgggacc
tccttgttttcgaccatggccgggggggagctccagctgcatagctattcgaactatagcttaagggggcctgg
911 aaaatggcaagtaaaaatagcctgatgggataggctctaagtcacaacgaaccagtgaccactg
ttttaccgttcattttatcggactaccctatccgagattcaggttgcttggtcacagtggtgac

975 acacaatgaggaaaaccatgtcacatctcgcagaactgggtgccagtgcggaaggcggccattagccaggcgtcagat
 tgtgttactccttttggtagagtgtagagcgtcttgaccaacggtcacgcttccgccggaatcggtccgcagtcta
 1▶ M S H L A E L V A S A K A A I S Q A S D
 1052 gttgccgcttagataatgtgcgcgctgaatatttgggtaaaaaagggcacttaacccttcagatgacgaccctgac
 caacggcgcaatctattacacgcgcagcttataaaccatttttcccgtaattgggaagtctactgctgggacgc
 21▶ V A A L D N V R V E Y L G K K G H L T L Q M T T L R
 1129 tgagctgccgccagaagagcgtccggcagctgggtgcggttatcaacgaagcgaagagcaggttcagcaggcgtga
 actcgacggcgggtcttctcgcaggccgctcgaccacgccaatagttgcttcgctttctcgccaagtctcgcgact
 46▶ E L P P E E R P A A G A V I N E A K E Q V Q Q A L
 1206 atgcgcgtaaagcggaaactgaaaagcgtgcaactgacgctgctggcggcggaacgattgatgtctctctgcc
 tacgcatcttgccttgaccttctcgcagcgtgacttacgcgagaccgccccttctgtaactacagagagacggt
 72▶ N A R K A E L E S A A L N A R L A A E T I D V S L P
 1283 ggtcgtcgcattgaaaaagggcggctgcatccggttaccggtaccatcgaccgctatcgaaagtttctcggtgagct
 ccagcagcgtaaacttttccgcccagacgtaggccaatgggcatggtagctggcatagctttcaagaagccactcga
 98▶ G R R I E N G G L H P V T R T I D R I E S F F G E L
 1360 tggctttaccgtggcaaccgggcccggaaatcgaagacgattatcataacttcgatgctctgaacattcctggtcacc
 accgaatggcaccgttggcccggccttttagcttctgctaatagtattgaagctacgagacttgaaggaccagtgg
 123▶ G F T V A T G P E I E D D Y H N F D A L N I P G H
 1437 acccggcgcgctgaccacgacacttctggtttgacactaccgctgctgctgctaccagacctctggcgtacag
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 149▶ H P A R A D H D T F W F D T T R L L R T Q T S G V Q
 1514 atccgcaccatgaaagcccagcagccaccgattcgtatcatcgcgcctggcctggtttatcgtaacgactacgacca
 taggcgtggtactttcgggtcgtcgggtgctaagcatagtagcgcggaccggcacaatagcattgctgatgctggt
 175▶ I R T M K A Q Q P P I R I I A P G R V Y R N D Y D Q
 1591 gactcacagccgatgttccatcagatggaaggtctgattggtgataccaacatcagctttaccaactgaaaggca
 ctgagtgctgggctacaaggtagctaccttccagactaacaactatggtttagctgaaatggttggactttccgt
 200▶ T H T P M F H Q M E G L I V D T N I S F T N L K G
 1668 cgctgcagcacttctcgcgtaacttctttaggaagatttgcagattcgttccgtccttctacttcccgtttacc
 gcgacgtgctgaaggacgattgaagaaactccttctaaacgtctaagcgaaggcaggaaggatgaagggcaaatgg
 226▶ T L H D F L R N F F E E D L Q I R F R P S Y F P F T
 1745 gaaccttctgcagaagtggacgtcatgggtaaaaaaggtaaatggctggaagtgctgggctgcccgatggtgcatcc
 cttggaagacgtcttccactgcagtagcatttttgcatttaccgaccttcacgaccgacgcccctaccacgtagg
 252▶ E P S A E V D V M G K N G K W L E V L G C G M V H P
 1822 gaacgtggtgctaacgttggcctcgcaccgggaagtttactctggtttcggcttcgggatggggatggagcgtctga
 cttgcacaacgcattgcaaccgtagctgggcttcaaatgagaccaaagccgaagccctaccctacctcgcagact
 277▶ N V L R N V G I D P E V Y S G F G F G M G M E R L
 1899 ctatgttgcgttacggcgtcaccgacctgcgttattcttgcgaaacgatctgcgtttcctcaaacagtttaataa
 gatacaacgcaatgccgcagtggtggcagcaagtaagaagcttttctagacgcaaggagtttgcataatttatt
 303▶ T M L R Y G V T D L R S F F E N D L R F L K Q F K
 1976 ggcaggaatagattatgaaattcagtgaactgtggttacgcgaatgggtgaaccggcgattgatagcgtgctgctg
 ccgtccttatctaataactttaagtcaacttgacaccaatgcgcttaccacttgggcccgtaactatcgctacgcgac

1976 ggcaggaatagattatgaattcagtgactgtggttacgcgaatgggtgaaccggcgattgatagcgatgcgctg
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 2053 gcaaatcaaatcactatggcgggctggaagttgggggatccactagttctagagcggccgccaccgcggtggagct
 cgtttagtttagtgataccgcccggacctcaacccccctaggtgatcaagatctcgccggcggtggcgccaccctcga
 2130 ccaattcgccctatagtgagtcgtattacaattcactggccgctgttttacaacgctcgtgactgggaaaaccctggc
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SphI (2228)

2207 gttaccceaacttaatcgccttgcacgCACCATTCTTGGCGGGCGGTGCTCAACGGCCTCAACCTACTACTGGGCT
 caatggggttgaattagcggaacgtacGTGGTAAGGAACGCCGCCACGAGTTGCCGGAGTTGGATGATGACCCGA
 2284 GCTTCCTAATGCAGGAGTCGCATAAGGGGAGAGCGTCGAGATCCCGGACACCATCGAATGGCGCAAAACCTTTCCGCGG
 CGAAGGATTACGTCCTCAGCGTATTCCCTCTCGCAGCTCTAGGGCCTGTGGTAGCTTACCGCGTTTTGAAAAGCGCC
 2361 TATGGCATGATAGCGCCCGAAGAGAGTCAATTCAGGGTGGTGAATGTGAAACAGTAACGTTATACGATGTCGCGAG
 ATACCGTACTATCGCGGGCCTTCTCAGTTAAGTCCCACCACTTACACTTTGGTCATTGCAATATGCTACAGCGTC
 2438 AGTATGCCGGTGTCTTTATCAGACCGTTTCCCGCGTGGTGAACCAGGCCAGCCACGTTTCTGCGAAAACCGGGAA
 TCATACGGCCACAGAGAATAGTCTGGCAAAGGGCGCACCACCTTGGTCCGGTCCGTGCAAAGACGCTTTTGCGCCCTT
 2515 AAAGTGGAAAGCGGCGATGGCGGAGCTGAATTACATTCCAAACCGCGTGGCACAACAACCTGGCGGGCAAACAGTGGT
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 2592 GCTGATTGGCGTTGCCACCTCCAGTCTGGCCCTGCACGCGCCGTGCGAAATTGTGCGGGCGATTAAATCTCGCGCCG
 CGACTAACCGCAACGGTGGAGGTGAGACCGGGACGTGCGCGGACGCTTAAACAGCGCCGTAATTTAGAGCGCGGC
 2669 ATCAACTGGGTGCCAGCGTGGTGTGATGGTAGAACGAAGCGGCGTGAAGCCTGTAAAGCGGCGGTGCACAAAT
 TAGTTGACCCACGGTCGCACCACACAGCTACCATCTTGCTTCGCCGACGTTCCGGACATTTCCGCCCCACGTGTTA
 2746 CTTCTCGCGCAACGCGTCAGTGGGCTGATCATTAACTATCCGCTGGATGACCAGGATGCCATTGCTGTGGAAGCTGC
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 2823 CTGCACTAATGTTCCGGCGTTATTTCTTGATGTCTCTGACAGACACCCATCAACAGTATTATTTCTCCCATGAAG
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 3131 TGCTGTTGCCAACGATCAGATGGCGCTGGGCGCAATGCGCGCCATTACCGAGTCCGGGCTGCGCGTTGGTGCAGAT
 ACGACCAACGGTTGCTAGTCTACCGCGACCCGCGTTACGCGCGGTAATGGCTCAGGCCCGACGCGCAACCACGCCTA
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 3670 GGTCAATTTTCGGCGAGGACCGCTTTCGCTGGAGCGGACGATGATCGGCCTGTGCTTGGCGTATTCCGAATCTTGC
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 3747 ACGCCCTCGCTCAAGCCTTGTCACTGGTCCCGCCACCAAACGTTTTGGCGAGAAGCAGGCCATTATCGCCGGCATG
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 3824 GCGGCCCCACGGGTGCGCATGATCGTGCTCTGTGCTTGGAGACCCGGCTAGGCTGGCGGGGTTGCTTACTGGTTA
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 3901 GCAGAATGAATCACCGATACGCGAGCGAACGTGAAGCGACTGCTGCTGCAAAACGCTGCGACCTGAGCAACAACAT
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4132 GATTTTTCTCTGGTCCCGCCGATCCATACCGCCAGTTGTTTACCCTCACAAACGTTCCAGTAACCGGCATGTTTCAT
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5980 GAAAAATATTGTTGATGCGCTGGCAGTGTTCTGCGCCGGTTCATTGATTCCCTGTTTGTAAATTGTCCTTTTAAACAG
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6519 CCCGAAAAGTGCCACCTGAAATTGTAAACGTTAATATTTTTGTTAAAATTCGCGTTAAATTTTTGTTAAATCAGCTCA
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6750 ATGGCCCACTACGTGAACCATCACCTAATCAAGTTTTTTGGGGTCGAGGTGCCGTAAAGCACTAAATCGGAACCT
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6827 AAAGGGAGCCCCGATTTAGAGCTTGACGGGGAAAGCCGGCGAACGTGGCGAGAAAGGAAGGGAAAGCGAAAGG
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6904 AGCGGGCGCTAGGGCGCTGGCAAGTGTAGCGGTACGCTGCGCGTAACCACCACACCCGCCGCGCTTAATGCGCCGC
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ATGTCCCGCGCAGGGTAAGCGGT

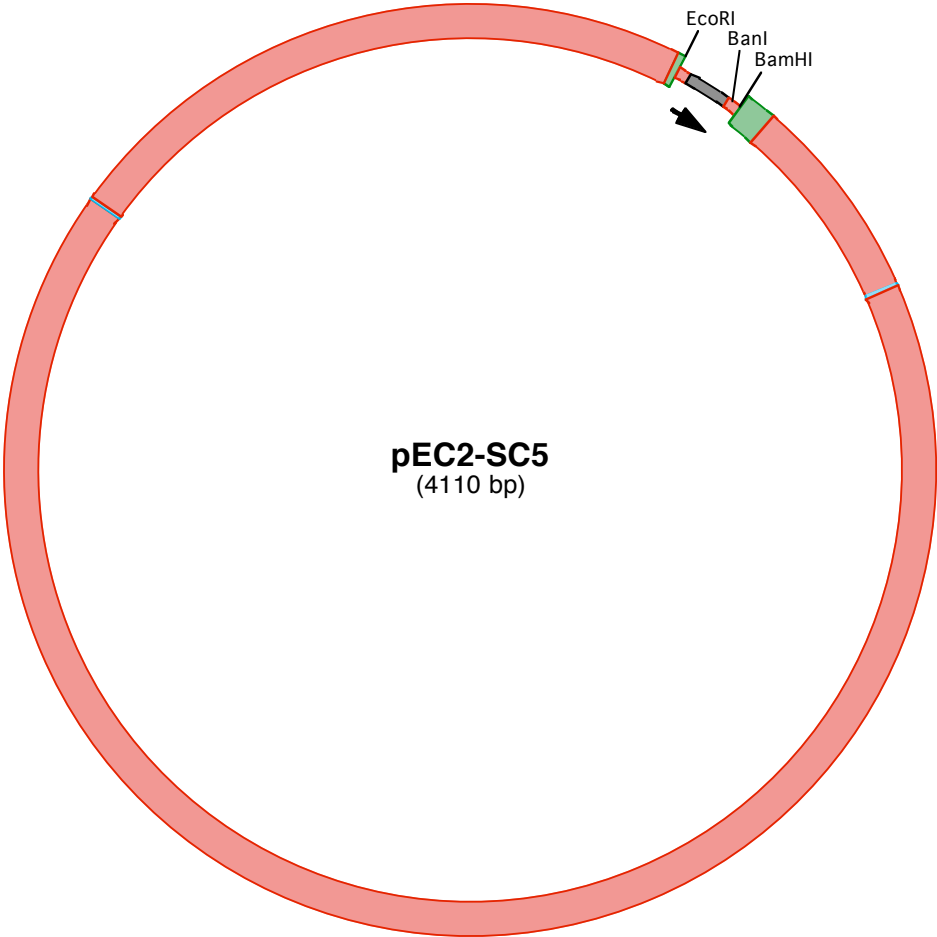
A.2 pEC2-SC5, -RGD, -RDG plasmid information**Submitted by** Marissa Mock **Date 4/22/04****Strain name/** XL-1 blue (*SupE44 hsdR17 recA1 endA1 gyrA46 thi relA1 lac⁻*
F'*[proAB⁺ lacI^q lacZΔM15 Tn10(tet^r)]* (From Stratagene))/ **pEC2-SC5**
pEC2-RGD
pEC2-RDG**Vector (kb) :** **pEC2-SC5**
pEC2-RGD
pEC2-RDG**Cloning site:** See plasmid map.**Construction of pEC2-CBD plasmids**

The cell binding domain region was cut out of the appropriate pUC19-SC5, pUC19-RGD, or pUC19-RDG vector with EcoR I/ BamH I digestion. The pEC2 (Eric Cantor) vector was cut with EcoR I/BamH I and ligated with the inserts to produce pEC2-SC5, pEC2-RGD, and pEC2-RDG.

Plasmid map and full sequence are shown only for pEC2-SC5; others are similar.

Source available :**12 % Glycerol culture in Marissa freezer box****Culture conditions:** 2xYT, 37°C

A.2.1 Plasmid map



A.2.2 Position of elements

	<u>bp</u>
Vector size	4110
SC5 insert	314-420
RGD insert	314-414
RDG insert	314-414

A.2.3 Full sequence (pEC2-SC5)

1 GTTGACGCCGGGCAAGAGCAACTCGGTCGCCGATACACTATTCTCAGAATGACTTGGTTGAGTACTCACCAGTCACAGAAA
CAACTGCGGCCGTTCTCGTTGAGCCAGCGCGTATGTGATAAGAGTCTTACTGAACCAACTCATGAGTGGTCAGTGTCTTT

83 AGCATCTTACGGATGGCATGACAGTAAGAGAATTATGCAGTGTGCCATAACCATGAGTGATAAACTGCGGCCAACTTACT
TCGTAGAATGCCTACCGTACTGTCATTCTCTTAATACGTCACGACGGTATTGGTACTCACTATTGTGACGCCGGTTGAATGA

165 TCTGACAACGATCGGAGGACCGAAGGAGCTAACCCTTTTTTGCAACAACATGGGGGATCATGTAACCGCCTTGATCGTTGG
AGACTGTTGCTAGCCTCCTGGCTTCCTCGATTGGCGAAAAACGTGTTGTACCCCTAGTACATTGAGCGAACTAGCAACC

247 GAACCGGAGCTGAATGAAGCCATACCAAACGACGAGCGTGACACCACGATGCCTGCAgatctgatcag aat tcg tcg
CTTGGCCTCGACTTACTTCGGTATGGTTTGCTGCTCGACTGTGGTGCTACGGACGTctagactagtc tta agc agc

PstI EcoRI

324 NheI
acg cta gct tcc tcg atg gtg aag aga tcc aga tcg gcc aca tcc cgc gtg aag ttg atg
tgc gat cga agg agc tac cac ttc tct agg tct agc cgg tgt agg gcg cac ttc aac tac
1► L D G E E I Q I G H I P R E V D

384 NheI Bani XhoI BamHI
att acc acc tgt acg cta cgc cgg tgc cgc tcg agg gatccatctagagtcgacgctcggccggttaaccta
taa tgg tgg aca tgc gat cgc gcc acg gcg agc tcc ctaggtagatctcagctgcagccggcaattggat
17► D Y H L Y

PstI

454 ggagatctgcaGCAATGGCAACAACGTTGCGCAAATATTAAGTGGCAACTACTTACTCTAGCTTCCCGCAACAATTAAT
cctctagacgctCGTTACCGTTGTTGCAACGCGTTTGATAATTGACCGCTTGATGAATGAGATCGAAGGCCGTTGTTAATTA

536 AGACTGGATGGAGGCGGATAAAGTTGCAAGCACTTCTGCGCTCGGCCCTTCCGGCTGGCTGGTTTTATTGCTGATAAACT
TCTGACCTACCTCCGCTATTTCAACGTCCTGGTGAAGACGCGAGCCGGGAAGGCCGACCGACCAATAACGACTATTTAGA

618 GGAGCCGGTGAAGCGTGGTCTCGCGGTATCATTGCAGCACTGGGGCCAGATGGTAAGCCCTCCCGTATCGTAGTTATCTACA
CCTCGGCCACTCGACCCAGAGCGCCATAGTAACGTCGTGACCCCGGTCTACCATTCCGGAGGGCATAGCATCAATAGATGT

700 CGACGGGGAGTCAGGCAACTATGGATGAACGAAATAGACAGATCGCTGAGATAGGTGCgtgcCTCACTGATTAAGCATTGGT
GCTGCCCTCAGTCCGTTGATACCTACTTGTCTTATCTGTCTAGCGACTCTATCCACGcacgGAGTGACTAATTCGTAACCA

782 AACTGTCAGACCAAGTTTACTCATATATACTTTAGATTGATTTAAAACCTTATTTTTAATTTAAAAGGATCTAGGTGAAGAT
TTGACAGTCTGGTTCAAATGAGTATATATGAAATCTAACTAAATTTGAAGTAAAAATTAATTTCTAGATCCACTTCTA

864 CCTTTTTGATAATCTCATGACAAAATCCCTTAACGTGAGTTTTGTTCCACTGAGCGTCAGACCCCTAATAAGATGATCT
GGAAAACTATTAGAGTACTGGTTTTAGGGAATTGCACTCAAAGCAAGGTGACTCGCAGTCTGGGAATTATTCTACTAGA

946 TCTTGAGATCGTTTTGGTCTGCGGTAATCTCTTGCTCTGAAAACGAAAAACCGCCTTGCAGGGCGGTTTTTCGAAGGTT
AGAAGTCTAGCAAAACCGACGCGCATTAGAGAACGAGACTTTTTGCTTTTTGGCGAACGTCCCGCCAAAAGCTTCCAAG

1028 TCTGAGCTACCAACTCTTTGAACCGAGGTAACCTGGCTTGGAGGAGCGCAGTCACCAAACTTGTCTTTTTCAGTTTAGCCTTA
AGACTCGATGGTTGAGAAAATTGGCTCCATTGACCGAACCTCCTCGCTCAGTGGTTTTGAAACAGGAAAGTCAAATCGGAAT

1110 ACCGGCGCATGACTTCAAGACTAACTCCTCTAAATCAATTACCAGTGGCTGCTGCCAGTGGTGCTTTTTGCATGTCTTTCCGG
TGCCCGCTACTGAAGTTCTGATTGAGGAGATTTAGTTAATGGTCACCAGACGGTCACCACGAAAACGTACAGAAAGGCC

1192 GTTGGACTCAAGACGATAGTTACCGGATAAGGCGCAGCGGTGGACTGAACGGGGGTTCTGTCATACAGTCCAGCTTGGAG
CAACTGAGTTCTGCTATCAATGGCTATTCCGCGTCGCCAGCCTGACTTGCCCCCAAGCAGTATGTCAGGTGCAACCTC

1274 CGAACTGCCTACCCGAACTGAGTGTCAAGCGTGAATGAGACAAAACCGGCCATAACAGCGGAATGACACCGGTAACCGA
GCTTGACGGATGGCCCTTACTCACAGTCCGACCTTACTCTGTTGCGCCGGTATTGTGCTTACTGTGGCATTGGCT

1356 AAGGCAGGAACAGGAGAGCGCACGAGGGAGCCGCCAGGGGAAACGCCTGGTATCTTTATAGTCCTGTGGGTTTCGCCACC
TTCCGTCCTTGTCTCTCGCGTGCTCCCTCGGGGTCCCCCTTTGCGGACCATAGAAATATCAGGACAGCCAAAGCGGTGG

1438 ACTGATTTGAGCGTCAGATTTCTGTGATGCTTGTGAGGGGGCGGAGCCTATGGAAAAACGGCTTTGCCGCGGCCCTCTCACT
TGACTAAACTCGCAGTCTAAAGCACTACGAACAGTCCCCCGCTCGGATACCTTTTTGCCGAAACGGCGCCGGGAGAGTGA

1520 TCCCTGTTAAGTATCTTCTGGCATCTTCCAGGAAATCTCCGCCCGTTTCTAAGCCATTTCCGCTCGCCGAGTCGAACGA
AGGGACAATTCATAGAAGGACCGTAGAAGGTCTTTAGAGGGGGGCAAGCATTCCGTAAGGCGAGCGGCGTCAGCTTGT

1602 CCGAGCGTAGCGAGTCAGTGAGCGAGGAAGCGGAATATATCCTGTATCACATATTCTGCTGACGCACCGGTGCAGCCTTTTT
GGCTCGCATCGCTCAGTCACTCGCTCCTTCGCCTTATATAGGACATAGTGATAAGACGACTGCGTGCCACGTCGGAAAA

1684 TCTCCTGCCACATGAAGCACTTCACTGACACCCTCATCAGTGCCAACATAGTAAGCCAGTATACACTCCGCTAGCGCTGAGG
AGAGGACGGTGTACTTCGTGAAGTACTGTGGGAGTAGTCACGGTTGTATCATTCCGTCATATGTGAGGCGATCGCGACTCC

1766 TCTGCCTCGTGAAGAAGGTGTTGCTGACTCATACCAGGCCTGAATCGCCCCATCATCCAGCCAGAAAGTGAGGGAGCCACGG
AGACGGAGCACTTCTCCACAACGACTGAGTATGGTCCGACTTAGCGGGGTAGTAGGTCGGTCTTTCACTCCCTCGGTGCC

1848 TTGATGAGAGCTTTGTTGTAGGTGGACCAGTTGGTGATTTTTGAACTTTTGCTTTGCCACGGAACGGTCTGCGTTGTGGGAA
AACTACTCTCGAAACAACATCCACCTGGTCAACCACTAAAACCTTGAAAACGAAACGGTGCCTTGCCAGACGCAACAGCCCTT

1930 GATGCGTGATCTGATCCTTCAACTCAGCAAAAGTTCGATTTATTCAACAAAGCCACGTTGTGTCTCAAATCTCTGATGTTA
CTACGCACTAGACTAGGAAGTTGAGTCGTTTTCAAGCTAAATAAGTTGTTTCGGTGCAACACAGAGTTTTAGAGACTACAAT

2012 CATTGCACAAGATAAAAAATATATCATCATGAACAATAAAACTGTCTGCTTACATAAACAGTAATACAAGGGGTGTTATGAGC
GTAACGTGTTCTATTTTTATATAGTAGTACTTGTATTTTTGACAGACGAATGTATTTGTCAATTATGTTCCCCACAATACTCG

2094 CATATTCAACGGGAAACGTCTTGCTCGAGGCCGCGATTAATTTCAACATGGATGCTGATTTATATGGGTATAAATGGGCTC
GTATAAGTTGCCCTTTGAGAACGAGCTCCGGCGCTAATTTAAGGTTGTACCTACGACTAAATATACCCATATTTACCCGAG

2176 GCGATAATGTCGGGCAATCAGGTGCGACAATCTATCGATTGTATGGGAAGCCCGATGCGCCAGAGTTGTTTCTGAAACATGG
CGCTATTACAGCCCGTTAGTCCACGCTGTTAGATAGCTAACATACCCTTCGGGCTACGCGGTCTCAACAAAGACTTTGTACC

2258 CAAAGGTAGCGTTGCCAATGATGTTACAGATGAGATGGTCAGACTAAACTGGCTGACGGAATTTATGCCTCTTCCGACCATC
GTTTCCATCGCAACGGTTACTACAATGTCTACTCTACCAGTCTGATTTGACCGACTGCCTTAAATACGGAGAAGGCTGGTAG

2340 AAGCATTTTATCCGTACTCCTGATGATGCATGGTACTCACCCTGCGATCCCCGGGAAAAACAGCATTCCAGGTATTAGAAG
TTCGTAATAATAGGCATGAGGACTACTACGTACCAATGAGTGGTGACGCTAGGGGCCCTTTTGTGTAAGGTCCATAATCTTC

2422 AATATCCTGATTCAGGTGAAAATATTGTTGATGCGCTGGCAGTGTTCTGCGCCGTTGCATTGATTCCCTGTTTGTAAATTG
TTATAGGACTAAGTCCACTTTTATAACAACCTACGCGACCGTCAAGGACGCGGCCAACGTAAGCTAAGGACAAACATTAAC

2504 TCCTTTTAAACAGCGATCGCGTATTTCTGCTCGCTCAGGCGCAATCACGAATGAATAACGGTTTGGTTGATGCGAGTGATTTT
AGGAAAATTGTCGCTAGCGCATAAAGCAGAGCGAGTCCGCGTTAGTGCTTACTTATTGCCAAACCAACTACGCTCACTAAAA

2586 GATGACGAGCGTAATGGCTGGCCTGTTGAACAAGTCTGGAAGAAATGCATAAGCTTTTGCCATTCTACCGGATTGAGTCG
CTACTGCTCGCATTACCGACCGACAACCTTGTTCAGACCTTTCTTTACGTATTCGAAAACGGTAAGAGTGGCCTAAGTCAGC

2668 TCACTCATGGTGATTTCTCACTTGATAACCTTATTTTTGACGAGGGGAAATTAATAGGTTGTATTGATGTTGGACGAGTCGG
AGTGAGTACCACTAAAGAGTGAACCTATTGGAATAAAAACCTGCTCCCTTTAATTATCCAACATAACTACAACCTGCTCAGCC

2750 AATCGCAGACCGATAACCAGGATCTTGCCATCCTATGGAAGTGCCTCGGTGAGTTTTCTCCTTCATTACAGAAACGGCTTTTT
TTAGCGTCTGGCTATGGTCTAGAACGGTAGGATACCTTGACGGAGCCACTCAAAGAGGAAGTAATGTCTTTGCCGAAAA

2832 CAAAAATATGGTATTGATAATCCTGATATGAATAAATTGAGTTTTCAATTTGATGCTCGATGAGTTTTTCTAATCAGAATTGG
GTTTTTATACCATAACTATTAGGACTATACTTATTTAACGTCAAAGTAACTACGAGCTACTCAAAAAGATTAGTCTTAACC

2914 TTAATTGGTTGTAACACTGGCAGAGCATTACGCTGACTTGACGGGACGGCGGCTTTGTTGAATAAAATCGAACTTTTGCTGAG
AATTAACCAACATTGTGACCGTCTCGTAATGCGACTGAACTGCCCTGCCGCCGAAACAACCTATTTAGCTTGAAAACGACTC

2996 TTGAAGGATCAGATCACGCATCTTCCCGACAACGCAGACCGTTCCGTGGCAAAGCAAAGTTCAAATCACCAACTGGTCCA
AACTTCCTAGTCTAGTGCGTAGAAGGGCTGTTGCGTCTGGCAAGGCACCGTTTCGTTTTCAAGTTTTAGTGTTGACCAGGT

3078 CCTACAACAAAGCTCTCATCAACCGTGGCTCCCTCACTTTCTGGCTGGATGATGGGGCGATTGAGGCTGGTATGAGTCAGC
GGATGTTGTTTCGAGAGTAGTTGGCACCGAGGGAGTGAAAGACCGACCTACTACCCCGCTAAGTCCGGACCATACTCAGTCG

3160 AACACCTTCTTACGAGGCAGACCTCAGCGCTCAAAGATGCAGGGGTAAAAGCTAACCGCATCTTTACCGACAAGGCATCCG
TTGTGGAAGAAGTGCTCCGTCTGGAGTCGCGAGTTTCTACGTCCCATTTCGATTGGCGTAGAAATGGCTGTTCCGTAGGC

3242 GCAGTTCAACAGATCGGGAAGGGCTGGATTTGCTGAGGATGAAGGTGGAGGAAGGTGATGTCATTCTGGTGAAGAAGCTCGA
CGTCAAGTTGTCTAGCCCTTCCCGACCTAAACGACTCCTACTTCCACCTCCTTCCACTACAGTAAGACCACTTCTTCGAGCT

3324 CCGTCTTGGCCGCGACACCGCCGACATGATCCAACCTGATAAAAGAGTTTGTGCTCAGGGGTAGCGGTTCCGTTTATTGAC
GGCAGAACCGCGCTGTGGCGGCTGTACTAGTTGACTATTTTCTCAAACCTACGAGTCCCACATCGCCAAGCCAATAACTG

3406 GACGGGATCAGTACCGACGGTGATATGGGGCAAATGGTGGTCACCATCCTGTCCGCTGTGGCACAGGCTGAACGCCGGAGGA
CTGCCCTAGTCATGGCTGCCACTATACCCCGTTTACCACCAGTGGTAGGACAGCCGACACCGTGTCCGACTTGGCGCCTCCT

3488 TCgatcCTAGAGCGCACGAATGAGGGCCGACAGGAAGCAAAGCTGAAAGGAATCAAATTTGGCCGAGGCGTACCGTGGACA
AGctagGATCTCGCGTCTTACTCCGGCTGTCTTCTGACTTTCCTTAGTTTAAACCGGCGTCCGCATGGCACCTGT

3570 GGAACGTGCTGCTGACGCTTATCAGAAGGGCACTGGTGCAACGGAAATTGCTCATCAGCTCAGTATTGCCGCTCCACGGT
CCTTGACGACGACTGCGAAGTAGTCTTCCCGTGACCAGTTGCCTTAAACGAGTAGTCGAGTCATAACGGGCGAGGTGCCA

3652 TTATAAAATCTTGAAGACGAAAGGGCCTCGTGATACGCCTATTTTTATAGGTTAATGTCATGATAATAATGGTTTCTTAGA
AATATTTAAGAACTTCTGCTTTCGCGAGCACTATGCGGATAAAAATATCCAATTACAGTACTATTATTACCAAAGAATCT

3734 CGTCAGGTGGCACTTTTCGGGGAAATGTGCGCGGAACCCCTATTTGTTATTTTTCTAAATACATTCAAATATGTATCCGCT
GCAGTCCACCGTGAAAAGCCCCTTACACGCGCCTTGGGGATAAACAAATAAAAAGATTTATGTAAGTTTATACATAGGCGA

3816 CATGAGACAATAACCCTGATAAATGCTTCAATAATATTGAAAAAGGAAGAGTATGAGTATTCAACATTTCCGTGTGCGCCTT
GTACTCTGTTATTGGGACTATTTACGAAGTATTATACTTTTTCTTCTCATACTCATAAGTTGTAAAGGCACAGCGGGAA

3898 ATTCCCTTTTTGCGGCATTTTGCCTTCTGTTTTGCTACCCAGAAACGCTGGTGAAAGTAAAAGATGCTGAAGATCAGT
TAAGGGAAAAACGCCGTAAACGGAAGGACAAAAACGAGTGGGTCTTTCGCGACCACTTTCATTTTCTACGACTTCTAGTCA

3980 TGGGTGCACGAGTGGGTTACATCGAACTGGATCTCAACAGCGGTAAGATCCTTGAGAGTTTTCGCCCCGAAGAAGTTTTCC
ACCCACGTGCTCACCAATGTAGCTTGACCTAGAGTTGTCGCCATTCTAGGAACTCTCAAAGCGGGGCTTCTTGCAAAGG

4062 AATGATGAGCACTTTTAAAGTTCTGCTATGTGGCGCGGTATTATCCCGT
TTACTACTCGTAAAATTTCAAGACGATACACCGCGCCATAATAGGGCA

A.3 pEC2-SC5-ELF₅ and -ELF₆ information

Submitted by Marissa Mock **Date 4/22/04**

Strain name: JM109 (*e14-(McrA-) recA1 endA1 gyrA96 thi-1 hsdR17 (rK-mK+) supE44 relA1 fG(lac-proAB) [F' traD36 proAB lacIqZfGM15]*). (From Zymo Research))/ pEC2-SC5-ELF₅
or / pEC2-SC5-ELF₆

Vector (kb): pEC2-SC5-ELF₅ (X)
 pEC2-SC5-ELF₆ (X)

Construction of pEC2-CBD plasmids

ELF monomer (see sequence below) was obtained through Ban I digestion of pUC19-ELF (Nandita Sharma).

pEC2-SC5-ELF₅:

The 75 bp fragment was multimerized through ligation with T4 DNA ligase for 3 minutes at 0°C. The ligation mixture was run on a 2% agarose gel, and the band corresponding to pentamer (375 bp) was cut out and extracted from the gel. The pentamer DNA was ligated with pEC2-SC5 vector that had been digested with Ban I and dephosphorylated with CIP, to yield pEC2-SC5-ELF₅.

pEC2-SC5-ELF₆:

The 75 bp ELF fragment was mixed with T4 ligase at 0°C for 30 s and added directly to pEC2-SC5 vector that had been digested with Ban I and dephosphorylated with CIP. The entire ligation mixture was transformed in JM109 competent cells; screened colonies revealed a strain containing pEC2-SC5-ELF₆.

ELF monomer sequence:

ggtgccgggtgtggcggtccgggcgtgggtgtaccgggcttcgggtgtccgggcgtaggtgttccgggtgtcggggtgcc

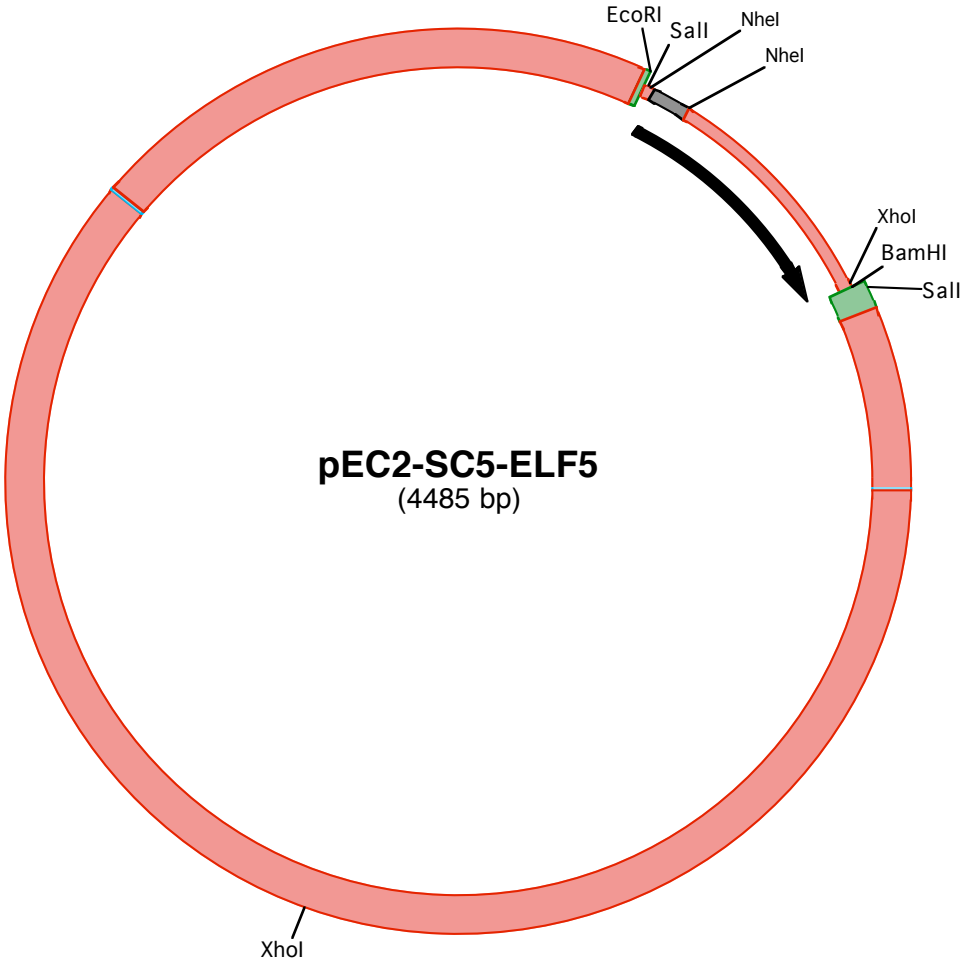
Plasmid map and full sequence shown only for pEC2-SC5-ELF₅; other is similar.

Source available :

12 % Glycerol culture in Marissa freezer box

Culture conditions: 2xYT, 37°C

A.3.1 Plasmid map



A.3.2 Position of elements

	<u>bp</u>
Vector size	4485
SC5 cell binding domain	314-406
ELF pentamer	407-782

A.3.3 Full sequence (pEC2-SC5-ELF₅)

1 GTTGACGCCGGGCAAGAGCAACTCGGTCGCCGCATACACTATTCTCAGAATGACTTGGTTGAGTACTCACCAGTCACA
CAACTGCCGCCCGTTCTCGTTGAGCCAGCGCGTATGTGATAAGAGTCTTACTGAACCAACTCATGAGTGGTCAGTGT

79 GAAAAGCATCTTACGGATGGCATGACAGTAAGAGAATTATGCAGTGCTGCCATAACCATGAGTGATAACACTGCCGGC
CTTTTCGTAGAATGCCTACCGTACTGTCATTCTTAATACGTCACGACGGTATTGGTACTACTATTGTGACGCCGG

157 AACTTACTTCTGACAACGATCGGAGGACCGAAGGAGCTAACCGCTTTTTTGCACAACATGGGGGATCATGTAACCTGC
TTGAATGAAGACTGTTGCTAGCCTCTGGCTTCTCGATTGGCGAAAAACGTGTTGTACCCCTAGTACATTGAGCG

235 CTTGATCGTTGGGAACCGGAGCTGAATGAAGCCATACCAAACGACGAGCGTGACACCACGATGCCTGCAgatctgac
GAACTAGCAACCCTTGGCCTCGACTTACTTCGGTATGGTTTGTGCTCGCACTGTGGTGTACGGACGTctagactag

EcoRI NheI

313 ag aat tcg tcg acg cta gct tcc tcg atg gtg aag aga tcc aga tcg gcc aca tcc cgc
tc tta agc agc tgc gat cga agg agc tac cac ttc tct agg tct agc cgg tgt agg gcg
1► L D G E E I Q I G H I P

NheI

372 gtg aag ttg atg att acc acc tgt acg cta gcg cg gtgccgggtgtgggcgttcggggcgtgggtg
cac ttc aac tac taa tgg tgg aca tgc gat cgc gc cacggccacacccgcaaggcccgacccac
13► R E V D D Y H L Y A S A V P G V G V P G V G

438 taccgggcttcggtgtcccgggcgtaggtgttcgggtgtcgggtgccgggtgtgggcgttcggggcgtgggtgtac
atggccgaagccacagggcccgcatccacaaggccacagccccagggccacacccgcaaggcccgacccacatg
35► V P G F G V P G V G V P G V G V P G V G V P G V G V

516 cgggcttcggtgtcccgggcgtaggtgttcgggtgtcgggtgccgggtgtgggcgttcggggcgtgggtgtaccgg
gcccgaagccacagggcccgcatccacaaggccacagccccagggccacacccgcaaggcccgacccacatggcc
61► P G F G V P G V G V P G V G V P G V G V P G V G V P

594 gttcgggtgtcccgggcgtaggtgttcgggtgtcgggtgccgggtgtgggcgttcggggcgtgggtgtaccgggt
cgaagccacagggcccgcatccacaaggccacagccccagggccacacccgcaaggcccgacccacatggccgga
87► G F G V P G V G V P G V G V P G V G V P G V G V P G

672 tcggtgtcccgggcgtaggtgttcgggtgtcgggtgccgggtgtgggcgttcggggcgtgggtgtaccgggttcg
agccacagggcccgcatccacaaggccacagccccagggccacacccgcaaggcccgacccacatggccggaagc
113► F G V P G V G V P G V G V P G V G V P G V G V P G F

XhoI BamHI

750 gtgtcccgggcgtaggtgttcgggtgtcggg gtg ccg ctc gag g gatccatctagagtcgacgtcggccgt
cacagggcccgcatccacaaggccacagccc cac ggc gag ctc c ctaggtagatctcagctgcagccggca
139► G V P G V G V P G V G

822 taacctaggagatctgcaGCAATGGCAACAACGTTGCGCAAATATTAAGTGGCGAACTACTTACTCTAGCTTCCCGG
attggatcctctagacgtCGTTACCGTTGTTGCAACGCGTTTGATAATTGACCGTTGATGAATGAGATCGAAGGGCC

900 CAACAATTAATAGACTGGATGGAGGCGGATAAAGTTGCAGGACCATTCTGCGCTCGGCCCTTCCGGCTGGCTGGTTT
GTTGTTAATTATCTGACCTACCTCCGCCTATTCAACGTCCTGGTGAAGACGCGAGCCGGGAAGGCCGACCGACAAA

978 ATTGCTGATAAATCTGGAGCCGGTGAAGCGTGGTCTCGCGGTATCATTGCAGCACTGGGGCCAGATGGTAAGCCCTCC
TAACGACTATTTAGACCTCGGCCACTCGACCCAGAGCGCCATAGTAACGTCGTGACCCCGTCTACCATTCCGGGAGG

1056 CGTATCGTAGTTATCTACACGACGGGGAGTCAGGCAACTATGGATGAACGAAATAGACAGATCGTGAGATAGGTGCG
GCATAGCATCAATAGATGTGCTGCCCTCAGTCCGTTGATACTACTTGTCTTAGCGACTCTATCCACGC

1134 tgcCTCACTGATTAAGCATTGGTAACTGTCAGACCAAGTTTACTCATATATACTTTAGATTGATTTAAAACCTTCATTT
acgGAGTGACTAATTCGTAACCATTGACAGTCTGGTCAAATGAGTATATATGAAATCTAACTAAATTTTGAAGTAAA

1212 TTAATTTAAAAGGATCTAGGTGAAGATCCTTTTTGATAATCTCATGACCAAAATCCCTTAACGTGAGTTTTCGTTCCA
AATTAATTTTCTAGATCCACTTCTAGGAAAACTATTAGAGTACTGGTTTTAGGGAATTGCACTCAAAGCAAGGT

1290 CTGAGCGTCAGACCCCTTAATAAGATGATCTTCTTGAGATCGTTTTGGTCTGCGCGTAATCTCTTGCTCTGAAAACGA
GACTCGCAGTCTGGGAATTATTCTACTAGAAGAACTCTAGCAAAACAGACGCGCATTAGAGAACGAGACTTTTGCT

1368 AAAAACCGCCTTGACGGGCGTTTTTCGAAGTTCTCTGAGCTACCAACTCTTTGAACCGAGGTAACCTGGCTTGGAGG
TTTTTGGCGGAACGTCCCGCCAAAAAGCTTCAAGAGACTCGATGGTTGAGAACTTGGCTCCATTGACCGAACCTCC

1446 AGCGCAGTCACCAAACTTGCCTTTCAGTTTAGCCTTAACGGCGCATGACTTCAAGACTAACTCCTCTAAATCAAT
TCGGTCAGTGGTTTTGAACAGGAAAGTCAAATCGGAATTGGCCGCTACTGAAGTTCTGATTGAGGAGATTTAGTTA

1524 TACCAGTGGCTGCTGCCAGTGGTGTCTTTGCATGTCTTTCGGGTTGGACTCAAGACGATAGTTACCGGATAAGGCGC
ATGGTCACCGACGACGGTCAACAGAAAACGTACAGAAAAGGCCAACCTGAGTTCTGCTATCAATGGCCTATTCCGCG

1602 AGCGGTGCGGACTGAACGGGGGTTCTGTCATACAGTCCAGCTTGGAGCGAACTGCCTACCCGGAACCTGAGTGTCAGGC
TCGCCAGCCTGACTTGCCCCCAAGCACGTATGTCAAGTGAACCTCGCTTGACGGATGGGCCTTGACTCACAGTCCG

1680 GTGGAATGAGACAAACCGGCCATAACAGCGGAATGACACCGGTAACCGAAAGGCAGGAACAGGAGAGCGCACGAGG
CACCTTACTCTGTTTGCGCCGTATTGTGCCTTACTGTGGCCATTTGGCTTTCGTCCTTGTCTCTCGCGTGCTCC

1758 GAGCCGCCAGGGGAAACGCCTGGTATCTTTATAGTCTGTGGGTTTTGCCACCCTGATTTGAGCGTCAGATTTCCG
CTCGCGGTCCCTTTCGCGACCATAGAAATATCAGGACAGCCAAAGCGGTGGTACTAACTCGCAGTCTAAAGC

1836 TGATGCTTGTGAGGGGGCGGAGCCTATGAAAAACGGCTTTCGCGCGGCCCTCTCACTTCCCTGTTAAGTATCTTCC
ACTACGAACAGTCCCCCGCCTCGGATACTTTTTGCAGAACGGCGCGGGGAGAGTGAAGGGACAATTCATAGAAGG

1914 TGGCATCTTCCAGGAAATCTCCGCCCGTTCTGTAAGCCATTTCCGCTCGCCGAGTGAACGACCGAGCGTAGCGAGT
ACCGTAGAAGGTCCTTTAGAGGCGGGCAAGCATTCCGTAAGGGCGAGCGGCGTCAGCTTGTGGCTCGCATCGCTCA

1992 CAGTGAGCGAGGAAGCGGAATATATCCTGTATCACATATTCTGCTGACGCACCGGTGCAGCCTTTTTTCTCCTGCCAC
GTCACTCGCTCCTTCCGCTTATATAGGACATAGTGTATAAGACGACTGCGTGGCCAGTTCGAAAAAAGAGGACGGTG

2070 ATGAAGCACTTCACTGACACCCTCATCAGTGCCAACATAGTAAGCCAGTATACTCCGCTAGCGCTGAGGTCTGCCT
TACTTCGTGAAGTACTGTGGGAGTAGTCACGGTTGATCATTCCGGTCATATGTGAGGCGATCGCGACTCCAGACGGA

2148 CGTGAAGAAGGTGTTGCTGACTCATACCAGGCCTGAATCGCCCCATCATCCAGCCAGAAAGTGAAGGGAGCCACGGTTG
GCACTTCTTCCACAACGACTGAGTATGGTCCGACTTAGCGGGGTAGTAGGTGGTCTTTCCTCCCTCGGTGCCAAC

2226 ATGAGAGCTTTGTTGTAGGTGGACCAGTTGGTGATTTTGAACTTTTGCTTGGCCACGGAACGGTCTGCGTTGTCCGGGA
TACTCTGAAACAACATCCACCTGGTCAACCACTAAAACCTGAAAACGAAACGGTGCTTGGCAGACGCAACAGCCCT

2304 AGATGCGTGATCTGATCCTTCAACTCAGCAAAAGTTCGATTTATTCAACAAAGCCAGTTGTGTCTCAAATCTCTGA
TCTACGCACTAGACTAGGAAGTTGAGTCGTTTTCAAGCTAAATAAGTTGTTTTGGTGCAACACAGAGTTTTAGAGACT

2382 TGTTACATTGCACAAGATAAAAAATATATCATCATGAACAATAAACTGTCTGCTTACATAAACAGTAATAAAGGGGT
ACAATGTAACGTGTTCTATTTTTATATAGTAGTACTTGTATTTTGACAGACGAATGTATTTGTCATTATGTTCCCA

2460 GTTATGAGCCATATTCAACGGGAAACGTCTTGTCTGAGGCCGCGATTAAATCCAACATGGATGCTGATTTATATGGG
CAATACTCGGTATAAGTTGCCCTTGCAGAACGAGCTCCGGCGCTAATTTAAGGTTGTACCTACGACTAAATATACCC

2538 TATAAATGGGCTCGCGATAATGTGGGCAATCAGGTGCGACAATCTATCGATTGTATGGGAAGCCCGATGCGCCAGAG
ATATTTACCGAGCGCTATTACAGCCCGTTAGTCCACGCTGTTAGATAGCTAACATACCCTTCCGGGCTACGCGGTCTC

2616 TTGTTTCTGAAACATGGCAAAGGTAGCGTTGCCAATGATGTTACAGATGAGATGGTCAGACTAACTGGCTGACGGAA
AACAAAGACTTTGTACCGTTTCCATCGCAACGGTACTACAATGTCTACTCTACCAGTCTGATTTGACCGACTGCCTT

2694 TTTATGCCTCTTCCGACCATCAAGCATTTTATCCGTA CTCTGATGATGCATGGTACTCACCCTGCGATCCCCGGG
AAATACGGAGAAGGCTGGTAGTTCGTA AAAATAGGCATGAGGACTACTACGTACCAATGAGTGGTGACGCTAGGGGCC

2772 AAAACAGCATTCCAGGTATTAGAAGAATATCCTGATT CAGGTGAAAATATTGTTGATGCGCTGGCAGTGTTCTGCGC
TTTTGTCGTAAGGTCCATAATCTTCTTATAGGACTAAGTCCACTTTTATAACAAC TACGCGACCGTCACAAGGACGCG

2850 CGGTTGCATTTCGATTCCTGTTTGTAAATGTCCTTTT AACAGCGATCGCGTATTTCTGCTCGCTCAGGCGCAATCACGA
GCCAACGTAAGCTAAGGACAAACATTAACAGGAAAATTGTCGCTAGCGCATAAAGCAGAGCGAGTCCGCGTTAGTGCT

2928 ATGAATAACGGTTTGGTTGATGCGAGTGATTTTTGATGACGAGCGTAATGGCTGGCCTGTTGAACAAGTCTGAAAGAA
TACTTATTGCCAAACCAACTACGCTCACTAAAAC TACTGCTCGCATTACCGACCGACAACCTTGTTCAGACCTTTCTT

3006 ATGCATAAGCTTTTGCCATTCTCACCGGATTCAGTCGTC ACTCATGGTGATTTCTCACTTGATAACCTTATTTTTGAC
TACGTATTCGAAAACGGTAAGAGTGGCCTAAGTCAGCAGTGAGTACCACTAAAGAGTGAAC TATTGGAATAAAAACCTG

3084 GAGGGGAAATTAATAGGTTGATTGATGTTGGACGAGTCGGAATCGCAGACCGATACCAGGATCTTGCCATCCTATGG
CTCCCCTTAATTATCCAACATAACTACAACCTGCTCAGCCTTAGCGTCTGGCTATGGTCTAGAACGGTAGGATACC

3162 AACTGCCTCGGTGAGTTTTCTCCTTATTACAGAAACGGCTTTTTCAA AAATATGGTATTGATAATCCTGATATGAAT
TTGACGGAGCCACTCAAAGAGGAAGTAATGTCTTTGCCGAAAAGTTTTTATACCATAACTATTAGGACTATACTTA

3240 AAATTGCAGTTTCATTTGATGCTCGATGAGTTTTTCTAATCAGAATTGGTTAATTGGTTGTAACACTGGCAGAGCATT
TTTAACTCAAAGTAACTACGAGCTACTCAAAGATTAGTCTTAAACCAATTAACCAACATTGTGACCGTCTCGTAA

3318 ACGCTGACTTGACGGGACGGCGGCTTTGTTGAATAAATCGA ACTTTTTGCTGAGTTGAAGGATCAGATCACGCATCTTC
TGCGACTGAACTGCCCTGCCGCCAAAACAAC TATTATTAGCTTGAAAACGACTCAACTTCTAGTCTAGTGCGTAGAAG

3396 CCGACAACGCAGACCGTTCCGTGGCAAAGCAAAGTTCAA AATCACCAACTGGTCCACCTACAACAAAGCTCTCATCA
GGCTGTTGCGTCTGGCAAGGCACCGTTTCGTTTTCAAGTTT TAGTGGTTGACCAGGTGGATGTTGTTTCGAGAGTAGT

3474 ACCGTGGCTCCCTCACTTTCTGGCTGGATGATGGGGCGATT CAGGCCTGGTATGAGTCAGCAACACCTTCTTCACGAG
TGGCACCGAGGGAGTGAAAGACCGACTACTACCCCGCTAAGTCCGGAC CATACTCAGTCGTTGTGGAAGAAGTGCTC

3552 GCAGACCTCAGCGCTCAAAGATGCAGGGGTAAAAGCTAACC GCATCTTTACCGACAAGGCATCCGGCAGTTCAACAGA
CGTCTGGAGTCGCGAGTTTCTACGTCCCCATTTTCGATTGGCGTAGAAATGGCTGTTCCGTAGGCGGTCAAGTTGTCT

3630 TCGGGAAGGGCTGGATTTGCTGAGGATGAAGGTGGAGGAAGGTGATGTCATTCTGGTGAAGAAGCTCGACCGTCTTGG
AGCCCTTCCCGACCTAAACGACTCCTACTTCCACCTCCTTCCACTACAGTAAGACCACTTCTTCGAGCTGGCAGAACC

3708 CCGCGACACCGCCGACATGATCCAAC TATAAAAAGAGTTTGTGCTCAGGGTGTAGCGGTTTCGGTTTATTGACGACGG
GGCGCTGTGGCGGCTGACTAGGTTGACTATTTTCTCAA ACTACGAGTCCACATCGCCAAGCCAAATAACTGCTGCC

3786 GATCAGTACCGACGGTGATATGGGGCAAATGGTGGTCACCATCCTGT CCGCTGTGGCACAGGCTGAACGCCGGAGGAT
CTAGTCATGGCTGCCACTATACCCCGTTTACCACCA GTGGTAGGACAGCCGACACCGTGTCCGACTTGCGGCCTCTA

3864 *Cgatc*CTAGAGCGCACGAATGAGGGCCGACAGGAAGCAAAGCTGAAAGGAATCAAATTTGGCCG CAGGCGTACCGTGG
*Gctag*GATCTCGCGTGCTTACTCCCGGCTGTCTTCGTTTTCGACTTTCCTTAGTTTAAACCGGCGTCCGCATGGCACC

3942 ACAGGAACGTCGTGCTGACGCTTCATCAGAAGGGCACTGGTGCAACGGAAATTGCTCATCAGCTCAGTATTGCCCGCT
TGCTCTTGACGACGACTGCGAAGTAGTCTTCCCGTGACCACGTTGCCTTTAACGAGTAGTCGAGTCATAACGGGCGA

4020 CCACGGTTTATAAAATTTCTGAAGACGAAAGGGCCTCGTGATACGCCTATTTTTATAGGTTAATGTCATGATAATAAT
GGTGCCAAATATTTTAAAGAACTTCTGCTTTCCCGGAGCACTATGCGGATAAAAATATCCAATTACAGTACTATTATTA

4098 GGTTCCTTAGACGTCAGGTGGCACTTTTCGGGGAAATGTGCGCGGAACCCCTATTTGTTTATTTTTCTAAATACATTC
CCAAAGAATCTGCAGTCCACCGTGAAAAGCCCTTTACACGCGCCTTGGGGATAAACAAATAAAAAGATTTATGTAAG

4176 AAATATGTATCCGCTCATGAGACAATAACCCTGATAAATGCTTCAATAATATTGAAAAAGGAAGAGTATGAGTATTCA
TTTATACATAGGCGAGTACTCTGTTATTGGGACTATTTACGAAGTTATTATAACTTTTTCTTCTCATACTCATAAGT

4254 ACATTTCCGTGTCGCCCTTATTCCCTTTTTGCGGCATTTTGCCTTCCTGTTTTTGTCTACCCAGAAACGCTGGTGAA
TGTAAGGCACAGCGGGAATAAGGGAAAAACGCCGTAAAACGGAAGGACAAAAACGAGTGGGTCTTTGCGACCACTT

4332 AGTAAAAGATGCTGAAGATCAGTTGGGTGCACGAGTGGGTTACATCGAACTGGATCTCAACAGCGGTAAGATCCTTGA
TCATTTTCTACGACTTCTAGTCAACCCACGTGCTCACCCAATGTAGCTTGACCTAGAGTTGTCGCCATTCTAGGAACT

4410 GAGTTTTCGCCCCGAAGAACGTTTTCCAATGATGAGCACTTTTAAAGTTCTGCTATGTGGCGGGTATTATCCCGT
CTCAAAGCGGGGCTTCTTGCAAAGGTTACTACTCGTGAAAATTTCAAGACGATACACCGCGCCATAATAGGGCA