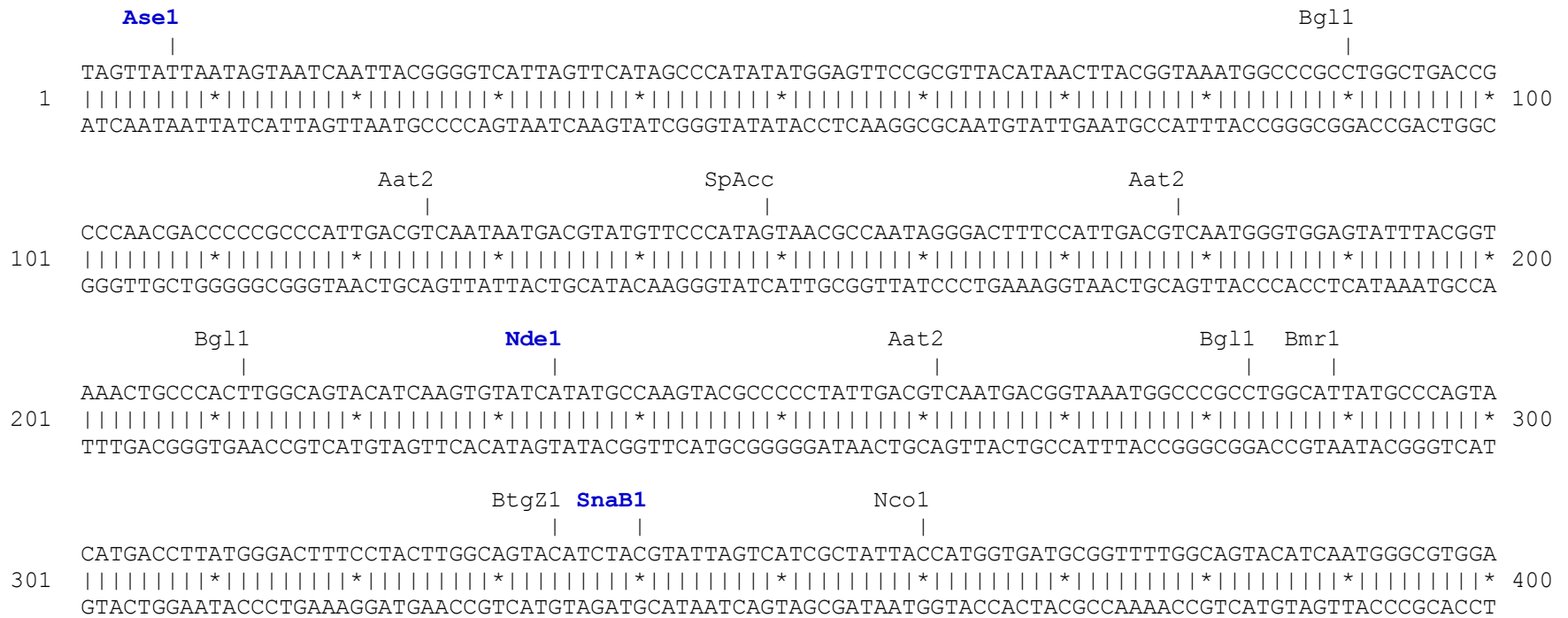


pTagFP635-Cx32 vector restriction map

The data has not been verified by restriction digestion with each enzyme listed and does not take into account possible methylation sites. Enzymes that recognize unambiguous sequences less than 6 basepairs long are not included – for the more complete enzyme list please refer to the Table of restriction sites.

Unique sites are shown in bold blue. The location given specifies the 3' end of the cut DNA (the base to the left of the cut site). TagFP635 amino acids are shown in red, Connexin 32 amino acids are shown in green, linker amino acids are shown in black.



```
                                     Aat2                                     Eci1
                                     |                                     |
TAGCGGTTTACTCAGGGGATTTCCAAGTCTCCACCCATTGACGTCAATGGGAGTTTGTGGTGGCACCACAAAATCAACGGGACTTTCCAAAATGTCGTA
401  |||||*||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 500
ATCGCCAAACTGAGTGCCCTAAAGGTTTCAGAGGTGGGGTAACTGCAGTTACCCTCAAACAAAACCGTGGTTTTAGTTGCCCTGAAAGTTTTTACAGCAT
```

```
                                     Nhe1 Afel
                                     |   |
ACA ACTCCGCCCCATTGACGCAAATGGGCGGTAGGCGTGTACGGTGGGAGGTCTATATAAGCAGAGCTGGTTTTAGTGAACCGTCAGATCCGCTAGCGCTA
501  |||||*||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 600
TGTTGAGGCGGGGTAAGTGCCTTACC CGCCATCCGCACATGCCACCCTCCAGATATATTTCGTCTCGACCAAATCACTTGCCAGTCTAGGCGATCGCGAT
```

```
      Xho1 Hind3 EcoR1
      |   |   |   |
BpuE1 Bgl2 | Sac1 | BstB1 | SpAcc |               Bts1 | SpDon
      |   |   |   |   |   |   |               |   |   |
CCGACTCAGATCTCGAGCTCAAGCTTCGAATTCCCTCTGGGAAAGGGCAGCAGCACCCAGGTGTGGCAGTGACAGGGAGGTGTGAATGAGGCAGGATGA
601  |||||*||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 700
GGCCTGAGTCTAGAGCTCGAGTTCGAAGCTTAAGGGAGACCCTTTCCCGTCGTCGTCGGTCCACACCGTCACTGTCCCTCCACACTTACTCCGCCTACT
```

Connexin 32 >

M N

```
      BsrG1               Bsm1               Eco57
      |                   |                   |
ACTGGACAGGTTTGTACACCTTGCTCAGTGGCGTGAACCGGCATTCTACTGCCATTGGCCGAGTATGGCTCTCGGTCATCTTCATCTTCAGAATCATGGT
701  |||||*||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 800
TGACCTGTCCAAACATGTGGAACGAGTCACCGCACTTGGCCGTAAGATGACGGTAACCGGCTCATAACCGAGAGCCAGTAGAAGTAGAAGTCTTAGTACCA
```

Connexin 32 > W T G L Y T L L S G V N R H S T A I G R V W L S V I F I F R I M V

```
      SpAcc
      _Chi | Pst1
      |   |   |
GCTGGTGGTGGCTGCAGAGAGTGTGTGGGGTGATGAGAAAATCTTCCCTTCATCTGCAACACACTCCAGCCTGGCTGCAACAGCGTTTGTCTATGACCAATTC
801  |||||*||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 900
CGACCACCACCGACGTCTCTCACACACCCCACTACTCTTTAGAAGGAAGTAGACGTTGTGTGAGGTCCGACCGACGTTGTTCGCAAACGATACTGGTTAAG
```

Connexin 32 > L V V A A E S V W G D E K S S F I C N T L Q P G C N S V C Y D Q F

```

          SpAcc          BseY1          Pml1
          Pst1|          BseR1 |          BssS1  Msc1  BstX1  SpDon
          ||          ||          |          |          |          |
1901  TTCCCCATCTCCCATGTGCGGCTGTGGTCCCTGCAGCTCATCCTAGTTTCCACCCCAGCTCTCCTCGTGGCCATGCACGTGGCTCACCAACACATAG
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1000
      AAGGGGTAGAGGTACACGCCGACACCAGGGACGTCGAGTAGGATCAAAGGTGGGGTTCGAGAGGAGCACCGGTACGTGCACCGAGTGGTTCGTTGTGTATC
Connexin 32 > F P I S H V R L W S L Q L I L V S T P A L L V A M H V A H Q Q H I E
    
```

```

                          SpDon
                          Ear1 |
                                EcoN1  | |          Bpm1
          Nco1 SanD1  BpuE1SpAcc  | | |          BseR1 |          Ale1  PshA1
          |          |          |          |          |          |          |
1001  AGAAGAAAATGCTACGGCTTGAGGGCCATGGGGACCCCCTACACCTGGAGGAGGTGAAGAGGCACAAGGTCCACATCTCAGGGACACTGTGGTGGACCTA
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1100
      TCTTCTTTTACGATGCCGAACCTCCCGGTACCCCTGGGGGATGTGGACCTCCTCCACTTCTCCGTGTTCCAGGTGTAGAGTCCCTGTGACACCACCTGGAT
Connexin 32 > K K M L R L E G H G D P L H L E E V K R H K V H I S G T L W W T Y
    
```

```

                          Bbs1          Nco1  Xcm1
                          |          |          |
1101  TGTTCATCAGCGTGGTGTTCGGGCTGTGTTTGGAGCCGTCTTCATGTATGTCTTTTATCTGCTCTACCCTGGCTATGCCATGGTGGCGGCTGGTCAAGTGC
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1200
      ACAGTAGTCGCACCACAAGGCCGACAACAAACTCCGGCAGAAGTACATACAGAAAATAGACGAGATGGGACCGATAACGGTACCACGCCGACCAGTTACG
Connexin 32 > V I S V V F R L L F E A V F M Y V F Y L L Y P G Y A M V R L V K C
    
```

```

          Aat2          BstX1          Bbs1  Bbs1          Nhe1
          |          |          |          |          |
1201  GACGTCTACCCCTGCCCAACACAGTGGACTGCTTCGTGTCCCGCCCCACCGAGAAAACCGTCTTCACCGTCTTCATGCTAGCTGCCTCTGGCATCTGCA
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1300
      CTGCAGATGGGGACGGGGTTGTGTACCTGACGAAGCACAGGGCGGGGTGGCTCTTTTGGCAGAAGTGGCAGAAGTACGATCGACGGAGACCGTAGACGT
Connexin 32 > D V Y P C P N T V D C F V S R P T E K T V F T V F M L A A S G I C I
    
```

```

                          BseY1  BsrB1
                          |          |
1301  TCATCCTCAATGTGGCCGAGGTGGTGTACCTCATCATCCGGGCTGTGCCCGCCGAGCCCAGCGCCGCTCCAATCCACCTTCCCGAAGGGCTCGGGCTT
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1400
      AGTAGAGTTACACCGGCTCCACCACATGGAGTAGTAGGCCCGGACACGGGCGGCTCGGGTTCGCGGCGAGGTTAGGTGGAAGGGCGTTCCCAGCCCGAA
Connexin 32 > I L N V A E V V Y L I I R A C A R R A Q R R S N P P S R K G S G F
    
```

SpDon
 |
 CGGCCACCGCCTCTCACCTGAATACAAGCAGAATGAGATCAACAAGCTGCTGAGTGAGCAGGATGGCTCCCTGAAAGACATACTGCGCCGCAGCCCTGGC
 1401 ||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||| 1500
 GCCGGTGGCGGAGAGTGGACTTATGTTTCGTCTTACTCTAGTTGTTTCGACGACTCACTCGTCTTACCGAGGGACTTTCTGTATGACGCGCGCTCGGGACCG
Connexin 32 > **G H R L S P E Y K Q N E I N K L L S E Q D G S L K D I L R R S P G**

BseY1 SpAcc BamH1 Age1 Ale1 Sap1
 | | | | | | | |
 ACCGGGGCTGGGCTGGCTGAAAAGAGCGACCGCTGCTCGGCCTGCGCGGATCCACCGGTGCGCCACCATGGTGTCTAAGGGCGAAGAGCTGATTAAGGAGA
 1501 ||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||| 1600
 TGGCCCCGACCCGACCGACTTTTCTCGCTGGCGACGAGCCGGACGCGCCTAGGTGGCCAGCGGTGGTACCACAGATTCCCCTCTCGACTAATTCCTCT
Cx32/TagFP635 > **T G A G L A E K S D R C S A C A D P P V A T M V S K G E E L I K E N**

BsrG1 ApaL1 SpAcc
 | | | | | | | |
 ACATGCACATGAAGCTGTACATGGAGGGCACCCTGAACAACCACCCTTCAAGTGCACATCCGAGGGCGAAGGCAAGCCCTACGAGGGCACCAGACCAT
 1601 ||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||| 1700
 TGTACGTGTACTTTCGACATGTACCTCCCGTGGCACTTGTGGTGGTGAAGTTCACGTGTAGGCTCCCGCTTCCGTTCCGGGATGCTCCCGTGGGTCTGGTA
TagFP635 > **M H M K L Y M E G T V N N H H F K C T S E G E G K P Y E G T Q T M**

GAGAATCAAGGTGGTGCAGGGCGGCCCTCTCCCTTCGCCTTCGACATCCTGGCTACCAGCTTCATGTACGGCAGCAAAACCTTCATCAACCACACCCAG
 1701 ||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||| 1800
 CTCTTAGTTCACCAGCTCCCGCCGGGAGAGGGGAAGCGGAAGCTGTAGGACCGATGGTTCGAAGTACATGCCGTCGTTTTGGAAGTAGTTGGTGTGGGTC
TagFP635 > **R I K V V E G G P L P F A F D I L A T S F M Y G S K T F I N H T Q**

Bsu36 SpDon Bbs1 Bpm1
 | | | | | | | |
 GGCATCCCCGACTTCTTTAAGCAGTCCTTCCCTGAGGGCTTCACATGGGAGAGAGTCAACACATACGAAGACGGGGGCGTGTGACCGCTACCCAGGACA
 1801 ||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||| 1900
 CCGTAGGGGCTGAAGAAATTCGTCAGGAAGGGACTCCCGAAGTGTACCCTCTCTCAGTGGTGTATGCTTCTGCCCCGACGACTGGCGATGGGTCTCTGT
TagFP635 > **G I P D F F K Q S F P E G F T W E R V T T Y E D G G V L T A T Q D T**

SpDon BseY1
 | | | | | | | |
 CCAGCCTCCAGGACGGCTGCCTCATCTACAACGTCAAGATCAGAGGGGTGAACTTCCCATCCAACGGCCCTGTGATGCAGAAGAAAACACTCGGCTGGGA
 1901 ||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||||||||*||| 2000
 GGTCGGAGGTCTGCCGACGGAGTAGATGTTGCAGTTCTAGTCTCCCCACTTGAAGGGTAGGTTGCCGGGACACTACGCTCTTTTTGTGAGCCGACCCT
TagFP635 > **S L Q D G C L I Y N V K I R G V N F P S N G P V M Q K K T L G W E**

```

StuI          EglI          BssS1          Eco57
|             |             |             |
GGCCTCACCGAGATGCTGTACCCCGCTGACGGCGGCTTGAAGGCAGAAGCGACATGGCCCTGAAGCTCGTGGGGCGGGGCCCACCTGATCTGCAACTTG
2001  |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 2100
CCGAGGTGGCTCTACGACATGGGGCGACTGCCGCCGACCTTCCGTCTTCGCTGTACCGGGACTTCGAGCACCCGCCCGGTGGACTAGACGTTGAAC
TagFP635  >  A S T E M L Y P A D G G L E G R S D M A L K L V G G G H L I C N L

          Bbs1          BpuE1          Bbs1          SpAcc          BsaI
          |             |             |             |             |
AAGACCACATACAGATCCAAGAAACCCGCTAAGAACCTCAAGATGCCCGGCTCTACTATGTGGACAGAAGACTGGAAAGAATCAAGGAGGCCGACAAAG
2101  |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 2200
TTCTGGTGTATGTCTAGGTTCTTTGGGCGATTCTTGAGTTCTACGGGCCGAGATGATACACCTGTCTTCTGACCTTTCTTAGTTCTCCGGCTGTTTC
TagFP635  >  K T T Y R S K K P A K N L K M P G V Y Y V D R R L E R I K E A D K E

                                     NotI
                                     EagI
          PshA1          BssS1  Dra3          MscI  AlwN1          SpAcc          BsrB1|          XbaI
          |             |             |             |             |             |             |
AGACCTACGTCGAGCAGCACGAGGTGGCTGTGGCCAGATACTGCGACCTCCCTAGCAATCTGGGGCACAACTTAATTGAGCGGCCGCGACTCTAGATCA
2201  |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 2300
TCTGGATGCAGCTCGTCGTGCTCCACCGACACCGGTCTATGACGCTGGAGGGATCGTTAGACCCCGTGTGTTGAATTAAGTAACTCGCCGGCGCTGAGATCTAGT
TagFP635  >  T Y V E Q H E V A V A R Y C D L P S N L G H K L N *

                                     BsmI
          BsaB1          SpDon  DraI          MfeI|
          |             |             |             |
TAATCAGCCATACCACATTTGTAGAGGTTTTACTTGGCTTTAAAAAACCTCCCACACCTCCCCCTGAACCTGAAACATAAAATGAATGCAATTGTTGTTGT
2301  |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 2400
ATTAGTCGGTATGGTGTAAACATCTCCAAAATGAACGAAATTTTTTGGAGGGTGTGGAGGGGGACTTGGACTTTGTATTTTACTTACGTTAACAACAACA

HpaI          polyA  PstI          polyA          polyA          BtsI  BsmI
|             |             |             |             |             |             |
TAACTTGTATTATTCAGCTTATAATGGTTACAAAATAAAGCAATAGCATCACAAATTTACAAAATAAAGCATTTTTTTCACTGCATTCTAGTTGTGGTTTG
2401  |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 2500
ATTGAACAAAATAACGTCGAATATTACCAATGTTTATTTTCGTTATCGTAGTGTTTAAAGTGTTTATTTTCGTAAAAAAGTGACGTAAGATCAACACCAAAC

```



```
              Ear1      Bsu36          Eci1          Pvu2 |          BseY1  
              |         |             |       |             |  
TTGAAAAAGGAAGAGTCTGAGGCGGAAAGAACCAGCTGTGGAATGTGTGTCAGTTAGGGTGTGGAAAGTCCCCAGGCTCCCCAGCAGGCAGAAGTATGC  
3101 | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | 3200  
AACTTTTCTCTCAGGACTCCGCCCTTCTTGGTCGACACCTTACACACAGTCAATCCCACACCTTTCAGGGGTCCGAGGGGTTCGTCCGTCTTCATACG
```

```
          Nsi1          Nsi1  
          Sph1 |          Sph1 |  
          BfrB1 |          BfrB1 |  
BstAP | |          SexA1          BseY1          BstAP | |  
          | |          |         |         |         |         |         |  
AAAGCATGCATCTCAATTAGTCAGCAACCAGGTGTGGAAAGTCCCCAGGCTCCCCAGCAGGCAGAAGTATGCAAAGCATGCATCTCAATTAGTCAGCAAC  
3201 | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | 3300  
TTTCGTACGTAGAGTTAATCAGTCGTTGGTCCACACCTTTCAGGGGTCCGAGGGGTTCGTCCGTCTTCATACGTTTCGTACGTAGAGTTAATCAGTCGTTG
```

```
              Eci1          Eci1          Eci1          Bmr1 |          Eci1          Nco1          polyA  
              |         |         |         ||          |         |         |  
CATAGTCCCGCCCTAACTCCGCCATCCCGCCCTAACTCCGCCCAGTTCCGCCCATTTCCGCCCATGGCTGACTAATTTTTTTTATTTATGCAGAG  
3301 | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | 3400  
GTATCAGGGCGGGGATTGAGGCGGGTAGGGCGGGATTGAGGCGGGTCAAGGCGGGTAAGAGGCGGGTACCGACTGATTAATAAATAAATACGTCTC
```

```
          Sfi1          Avr2          Stu1 |  
          Bgl1          SpDon          BseR1 | |          Cla1          BsaB1  
          |         |         |         |         |         |         |         |  
GCCGAGGCCGCTCGGCCTCTGAGCTATTCCAGAAGTAGTGAGGAGGCTTTTTTTGGAGGCCTAGGCTTTTGCAAAGATCGATCAAGAGACAGGATGAGGA  
3401 | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | 3500  
CGGCTCCGGCGGAGCCGGAGACTCGATAAGGTCTTCATCACTCTCCGAAAAACCTCCGGATCCGAAAAACGTTTCTAGCTAGTTCTCTGTCTACTCCT
```

```
          BspM1          BfuA1          Eag1          Bmr1  
          |         |         |         |  
TCGTTTCGCATGATTGAACAAGATGGATTGCACGCAGGTTCTCCGGCCGCTTGGGTGGAGAGGCTATTCGGCTATGACTGGGCACAACAGACAATCGGCT  
3501 | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | | | | | * | 3600  
AGCAAAGCGTACTAACTTGTCTACCTAACGTGCGTCCAAGAGGCCGGCGAACCCACCTCTCCGATAAGCCGATACTGACCCGTGTTGTCTGTTAGCCGA
```

```

                                Nar1
                                Kas1 |
                                ||
                                Drd1
                                |
3601 GCTCTGATGCCGCCGTGTTCCGGCTGTCAGCGCAGGGGCGCCCGGTTCTTTTTGTCAAGACCGACCTGTCCGGTGCCCTGAATGAACTGCAAGACGAGGC
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 3700
    CGAGACTACGGCGGCACAAGGCCGACAGTCGCGTCCCCGCGGGCCAAGAAAAACAGTTCTGGCTGGACAGGCCACGGGACTTACTTGACGTTCTGCTCCG

                                Msc1
                                |
                                Pvu2
                                | |
                                Fsp1 |
                                | |
                                PflF1
                                |
                                Eco57
                                |
3701 AGCGCGGCTATCGTGGCTGGCCACGACGGGCGTTCCTTGCGCAGCTGTGCTCGACGTTGTCACTGAAGCGGGAAGGGACTGGCTGCTATTGGGGCAAGTG
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 3800
    TCGCGCCGATAGCACCGACCGGTGCTGCCCGCAAGGAACGCGTCGACACGAGCTGCAACAGTGACTTCGCCCTTCCCTGACCGACGATAACCCGCTTCAC

                                SpDon
                                |
                                BciV1
                                |
                                BsrD1
                                |
3801 CCGGGCAGGATCTCCTGTCATCTCACCTTGCTCCTGCCGAGAAAGTATCCATCATGGCTGATGCAATGCGGCGGCTGCATACGCTTGATCCGGCTACCT
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 3900
    GGCCCCGTCTAGAGGACAGTAGAGTGGAACGAGGACGGCTCTTTCATAGGTAGTACCGACTACGTTACGCCCGGACGTATGCGAACTAGGCCGATGGA

                                BtgZ1
                                BspM1 |
                                BfuA1 |
                                | |
                                Sap1
                                Ear1
                                |
3901 GCCCATTCGACCACCAAGCGAAACATCGCATCGAGCGAGCACGTACTCGGATGGAAGCCGGTCTTGTCGATCAGGATGATCTGGACGAAGAGCATCAGGG
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 4000
    CGGGTAAGCTGGTGGTTCGCTTTGTAGCGTAGCTCGCTCGTGCATGAGCCTACCTTCGGCCAGAACAGCTAGTCCTACTAGACCTGCTTCTCGTAGTCCC

                                BpuE1
                                |
                                Sph1
                                |
                                Nco1
                                |
                                BtgZ1
                                |
4001 GCTCGCGCCAGCCGAACTGTTCCGCCAGGCTCAAGGCGAGCATGCCCCGACGGCGAGGATCTCGTTCGTGACCCATGGCGATGCCTGCTTGCCGAATATCATG
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 4100
    CGAGCGGGTTCGGCTTGACAAGCGGTCCGAGTTCGCTCGTACGGGCTGCCGCTCCTAGAGCAGCACTGGGTACCGCTACGGACGAACGGCTTATAGTAC

```



```

                        BseY1
                        NaeI  |
                        NgoM4 | |           Rsr2           EciI           SapI
                                   |                   |                   |
                                   |                   |                   |
    4101  GTGAAAAATGGCCGCTTTTCTGGATTCATCGACTGTGGCCGGCTGGGTGTGGCGGACCGCTATCAGGACATAGCGTTGGCTACCCGTGATATTGCTGAAG
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 4200
    CACCTTTTACCGGCGAAAAGACCTAAGTAGCTGACACCGGCCGACCCACACCGCCTGGCGATAGTCCTGTATCGCAACCGATGGGCACTATAACGACTTC

                               BtgZ1
                               BsrB1 |
                               BssS1 |
                               Eco57 |
    4201  AGCTTGGCGGCGAATGGGCTGACCGCTTCTCGTGCTTTACGGTATCGCCGCTCCCGATTTCGAGCGCATCGCCTTCTATCGCCTTCTTGACGAGTTCTT
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 4300
    TCGAACCGCCGCTTACCCGACTGGCGAAGGAGCACGAAATGCCATAGCGGCGAGGGCTAAGCGTTCGCGTAGCGGAAGATAGCGGAAGAACTGCTCAAGAA

                                       BssS1
                                       BspM1
                                       BfuA1
    BsrB1       BstB1
    |           |
    4301  CTGAGCGGGGACTCTGGGGTTCGAAATGACCGACCAAGCGACGCCCAACCTGCCATCACGAGATTTCGATTCCACCGCCGCCTTCTATGAAAGGTTGGGCT
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 4400
    GACTCGCCCTGAGACCCCAAGCTTTACTGGCTGGTTCGCTGCGGGTTGGACGGTAGTGCTCTAAAGCTAAGGTGGCGGCGGAAGATACTTTCCAACCCGA

                                       Bpm1
                                       SpAcc
                                       Avr2
                                       |
                                       |
    4401  TCGGAATCGTTTTCCGGGACGCCGGCTGGATGATCCTCCAGCGCGGGGATCTCATGCTGGAGTTCTTCGCCCACCCTAGGGGGAGGCTAACTGAAACACG
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 4500
    AGCCTTAGCAAAAAGGCCCTGCGGCCGACCTACTAGGAGGTCGCGCCCCTAGAGTACGACCTCAAGAAGCGGGTGGGATCCCCCTCCGATTGACTTTGTGC

                              polyA          polyA
                              |              |
    4501  GAAGGAGACAATACCGGAAGGAACCCGCGCTATGACGGCAATAAAAAGACAGAATAAAACGCACGGTGTGGGTCGTTTGTTCATAAACGCGGGGTTCCG
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 4600
    CTTCCTCTGTATGGCCTTCCTTGGGCGGATACTGCCGTTATTTTTCTGTCTATTTTTCGCGTGCCACAACCCAGCAAACAAGTATTTGCGCCCCAAGCC
  
```

```

                                Bsa1                               SpDon
                                |                               |
TCCCAGGGCTGGCACTCTGTGATAACCCACCGAGACCCCATTTGGGGCCAATACGCCC GCGTTTTCTTCTTTTTCCCCACCCCAAGTTCGGGTG
4601  |||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 4700
AGGGTCCCGACCGTGAGACAGCTATGGGGTGGCTCTGGGGTAACCCCGTTATGCGGGCGCAAAGAAGGAAAAGGGGTGGGGTGGGGGTTCAAGCCCAC

                                BstAP
                                AlwN1         Bsu36                               Dra1
                                |               |                               |
AAGGCCCAAGGGCTCGCAGCCAACGTCGGGGCGGCAGGCCCTGCCATAGCCTCAGGTTACTCATATATACTTTAGATTGATTTAAAACTTCATTTTAAATT
4701  |||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 4800
TTCCGGGTCCCGAGCGTCGGTTCAGCCCCGCCGTCGGGACGGTATCGGAGTCCAATGAGTATATATGAAATCTAACTAAATTTTGAAGTAAAATTA

Dra1                               BspH1
|                                   |
TAAAAGGATCTAGGTGAAGATCCTTTTTTGATAATCTCATGACCAAATCCCTTAACGTGAGTTTTTCGTTCCACTGAGCGTCAGACCCCGTAGAAAAGATC
4801  |||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 4900
ATTTTCTAGATCCACTTCTAGGAAAACTATTAGAGTACTGGTTTTAGGGAATTGCACTCAAAGCAAGGTGACTCGCAGTCTGGGGCATCTTTTCTAG

                                BpuE1
                                |
AAAGGATCTTCTTGAGATCCTTTTTTCTGCGGTAATCTGCTGCTTGCAAACAAAAAAACCACCGCTACCAGCGGTGGTTTTGTTTGCCGGATCAAGAGC
4901  |||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 5000
TTTCTAGAAAGAACTCTAGGAAAAAAGACGCGCATTAGACGACGAACGTTTGTTTTTTTTGGTGGCGATGGTCGCCACCAAACAAACGGCCTAGTTCTCG

                                Eco57                               SpAcc
                                |                                   |
TACCAACTCTTTTTCCGAAGGTAAGTGGCTTCAGCAGAGCGCAGATACCAAATACTGTCTTCTAGTGTAGCCGTAGTTAGGCCACCACTTCAAGAAGTC
5001  |||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 5100
ATGGTTGAGAAAAAGGCTTCCATTGACCGAAGTCGCTCTCGCGTCTATGGTTTTATGACAGGAAGATCACATCGGCATCAATCCGGTGGTGAAGTCTTGAG

                                AlwN1                               BpuE1
                                |                                   |
TGTAGCACCGCCTACATACCTCGCTCTGCTAATCCTGTTACCAGTGGCTGCTGCCAGTGGCGATAAGTCGTGTCTTACCGGGTTGGACTCAAGACGATAG
5101  |||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 5200
ACATCGTGGCGGATGTATGGAGCGAGACGATTAGGACAATGGTCACCGACGACGGTCACCGCTATTTCAGCACAGAATGGCCCAACCTGAGTTCCTGCTATC

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                                     ApaL1      BseY1
                                     |          |
5201 TTACCGGATAAGGCGCAGCGGTCTGGGCTGAACGGGGGTTTCGTGCACACAGCCCAGCTTGGAGCGAACGACCTACACCGAACTGAGATACCTACAGCGTG
   |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 5300
   AATGGCCTATTCCGCGTCGCCAGCCGACTTGCCCCCAAGCACGTGTGTCTGGGTCTGAACCTCGCTTGCTGGATGTGGCTTGACTCTATGGATGTTCGCAC

                                     BciV1
                                     |          |
   SpAcc                               Eci1      BssS1
   |                                   |          |
5301 AGCTATGAGAAAAGCGCCACGCTTCCCGAAGGGAGAAAAGGCGGACAGGTATCCGGTAAGCGGCAGGGTCTGGAAACAGGAGAGCGCACGAGGGAGCTTCCAGG
   |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 5400
   TCGATACTCTTTCGCGGTGCGAAGGGCTTCCCTCTTTCGCGCTGTCCATAGGCCATTCGCCGTCCCAGCCTTGTCCTCTCGCGTGCTCCCTCGAAGGTCC

                                     SpAcc      Drd1      BpuE1      SpAcc
                                     |          |          |          |
5401 GGGAAACGCCCTGGTATCTTTATAGTCCTGTCTGGGTTTCGCCACCTCTGACTTGAGCGTCGATTTTTGTGATGCTCGTCAGGGGGCGGAGCCTATGGAAA
   |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 5500
   CCCTTTCGCGGACCATAGAAATATCAGGACAGCCCAAAGCGGTGGAGACTGAACTCGCAGCTAAAAAACTACGAGCAGTCCCCCGCCTCGGATACCTTT

                                     SpDon
                                     |          |
   Eci1                               BspLU|
   |                                   ||
5501 AACGCCAGCAACGCGGCCTTTTACGGTTCTTGGCCTTTTGCTGCACATGTTCTTCTGCGTTATCCCCTGATTCTGTGGATAACCGTA
   |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 5600
   TTGCGGTCGTTGCGCCGAAAAATGCCAAGGACCGGAAAACGACCGGAAAACGAGTGTACAAGAAAGGACGCAATAGGGGACTAAGACACCTATTGGCAT

                                     Nsi1
                                     BfrB1 |
                                     | |
5601 TTACCGCCATGCAT
   |||||*|||| 5614
   AATGGCGGTACGTA
```

Found:

Aat2	Afe1	Afl2	Age1	Ale1	AlwN1	ApaL1	Ase1	Avr2	BamH1	Bbs1	BciV1	BfrB1	BfuA1
Bgl1	Bgl2	Bmr1	Bpm1	BpuE1	Bsa1	BsaB1	BsaXa	BsaXb	BseR1	BseY1	Bsm1	BspH1	BspLU
BspM1	BsrB1	BsrD1	BsrG1	BssS1	BstAP	BstB1	BstX1	Bsu36	BtgZ1	Bts1	_Chi	Clal	Dra1
Dra3	Drd1	Eag1	Ear1	Eci1	Eco57	EcoN1	EcoR1	Fsp1	Hind3	Hpa1	Kas1	Mfe1	Msc1
Nae1	Nar1	Nco1	Nde1	NgoM4	Nhe1	Not1	Nsi1	PflF1	Pml1	polyA	PshA1	Psi1	Pst1
Pvu2	Rsr2	Sac1	SanD1	Sap1	SexA1	Sfi1	SnaB1	SpAcc	SpDon	Sph1	Ssp1	Stu1	Xba1
Xcm1	Xho1												

Unique:

Afe1	Afl2	Age1	Ase1	BamH1	Bgl2	BsaXa	BsaXb	BspLU	BsrD1	_Chi	Clal	EcoN1	EcoR1
Fsp1	Hind3	Hpa1	Kas1	Mfe1	Nar1	Nde1	Not1	PflF1	Pml1	Rsr2	Sac1	SanD1	SexA1
Sfi1	SnaB1	Xba1	Xcm1	Xho1									

Not found:

Aar1	Acc65	Ac11	Ahd1	Apa1	Asc1	AsiS1	Bae1a	Bae1b	BbvC1	Bcg1a	Bcg1b	Bcl1	Blp1
BmgB1	Bpu10	Bsg1	BsiW1	BsmB1	BspE1	BssH2	BstE2	BstZ1	EcoK	EcoRV	ScFRT	Fse1	FspA1
I_Ceu	Kpn1	loxP	Mlu1	Nru1	Pac1	PflM1	Pme1	PspOM	Pvu1	Sac2	Sal1	Sbf1	Sca1
Sgf1	SgrA1	Sma1	Spe1	Srf1	Swal	T3RNA	T7RNA	T7Ter	PISce	Xmn1			

Excluded by site complexity:

Acc1	Ac11	Afl3	Alu1	Alw1	Apo1	Ava1	Ava2	Ban1	Ban2	Bbv1	BceA1	Bfa1	Bme15
BsaA1	BsaH1	BsaJ1	BsaW1	BseM2	BsiE1	BsiH1	Bsl1	BsmA1	BsmF1	Bsp12	BspCa	BspCb	Bsr1
BsrF1	BssK1	BstF5	BstN1	BstU1	BstY1	Btg1	Cac8	CviJ1	Dde1	Eae1	EcoO1	Fau1	Fnu4H
Fok1	Hae2	Hae3	Hga1	Hha1	Hinc2	Hinf1	HinP1	Hpa2	Hph1	Hpy99	Hpy1	Hpy3	HpyC3
HpyC4	HpyC5	Mae3	Mbo2	Mnl1	Mse1	Msl1	MspA1	Mwo1	Nci1	Nla3	Nla4	Nsp1	Ple1
PpuM1	Rsa1	Sau3A	Sau96	SfaN1	Sfc1	Sml1	Sty1	Taq1	Tat1	Tfi1	Tse1	Tsp45	Tsp50
TspR1													