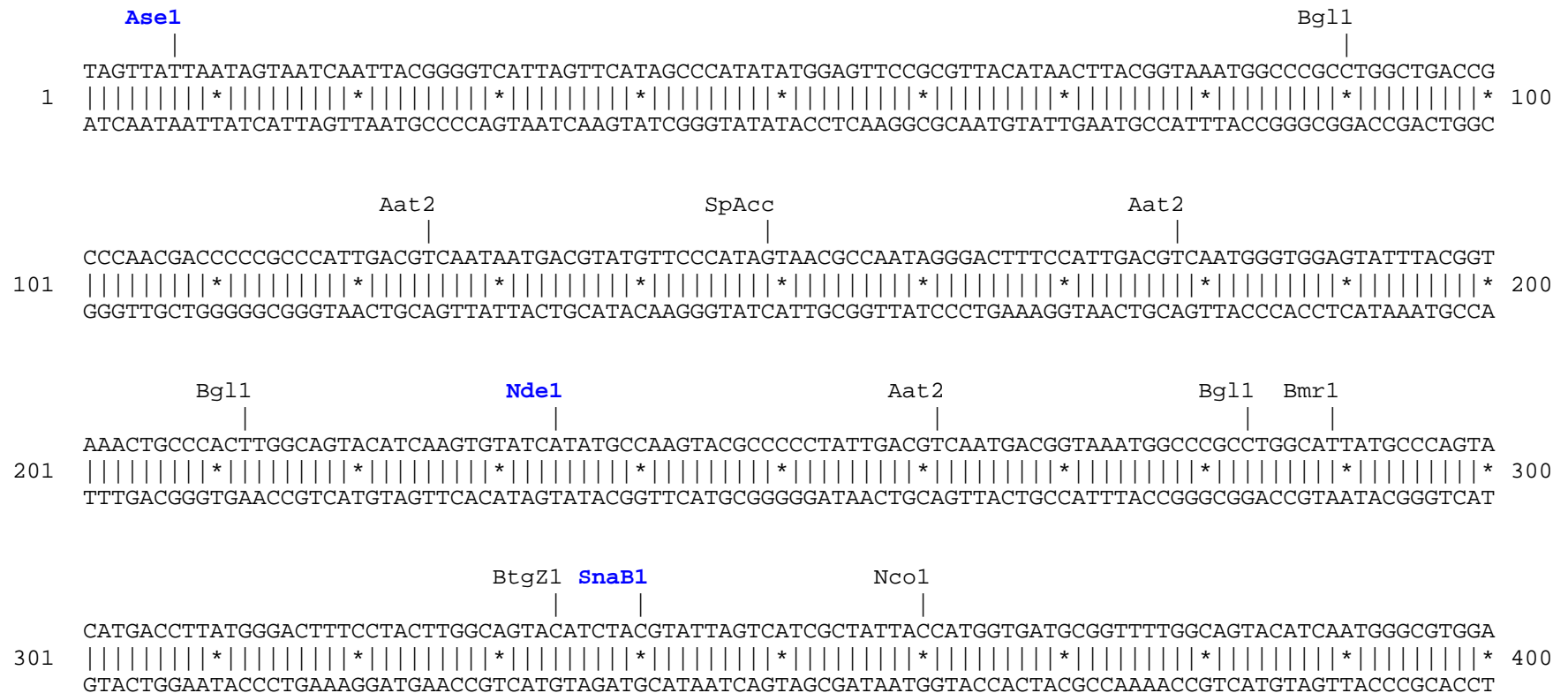


pFusionRed-ER vector restriction map

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).

FusionRed amino acids are shown in red, endoplasmic reticulum (ER) targeting sequence (calreticulin signal sequence) and ER retention signal (KDEL sequence) amino acids are shown in green, linker amino acids are shown in black.



BseY1 BseY1 StuI BsmB1

```

1001 CCAGGACGGCTGCCTCATCTACAACGTCAAGGTTAGAGGGGTGAACTTCCCAGCCAACGGCCCTGTGATGCAGAAGAAAACACTCGGCTGGGAGGCCTCC
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1100
    GGTCTGCGGACGGAGTAGATGTTGCAGTTCCAATCTCCCCACTTGAAGGGTCGGTTGCCGGGACACTACGTCTTCTTTTGTGAGCCGACCTCCGGAGG
FusionRed > Q D G C L I Y N V K V R G V N F P A N G P V M Q K K T L G W E A S
    
```

EgI1 BssS1 Eco57 Bsa1

```

1101 ACCGAGACGATGTACCCCGCTGACGGCGGCCTGGAAGGCGCATGTGACATGGCCCTGAAGCTCGTGGGCGGGGGCCACCTGATCTGCAACCTTGAGACCA
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1200
    TGGCTCTGCTACATGGGGCGACTGCCGCCGGACCTTCCGCGTACACTGTACCGGGACTTCGAGCACCCCGCCCCGGTGGACTAGACGTTGGAACCTCTGGT
FusionRed > T E T M Y P A D G G L E G A C D M A L K L V G G G H L I C N L E T T
    
```

BpuE1 SpAcc Bsa1 PshA1

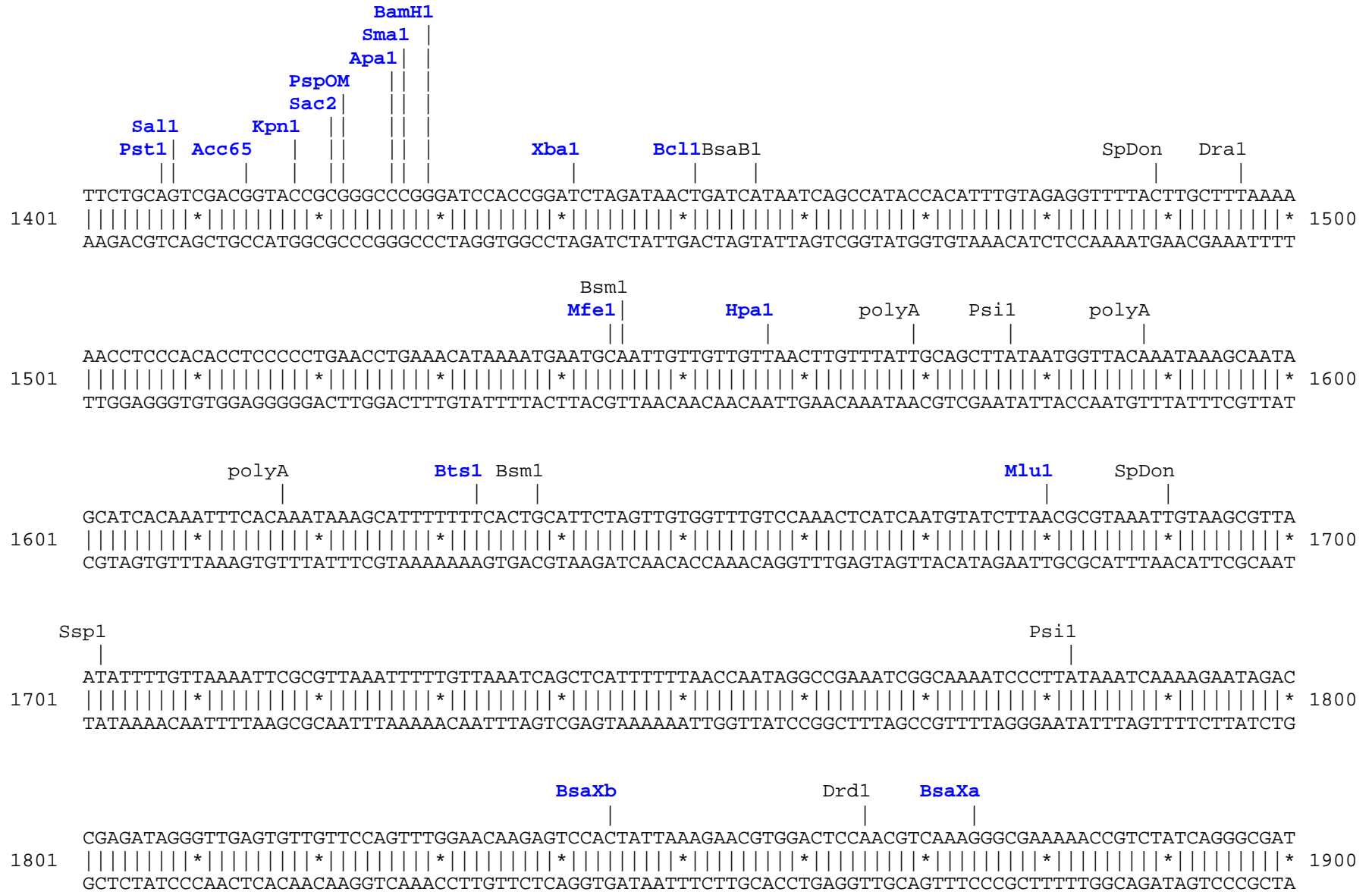
```

1201 CATAcAGATCCAAGAAACCCGCTACGAACCTCAAGATGCCCGGCGTCTACAACGTGGACCACAGACTGGAAAGAATCAAGGAGGCCGACGATGAGACCTA
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1300
    GTATGTCTAGGTTCTTTGGGCGATGCTTGGAGTTCTACGGGCCGAGATGTTGCACCTGGTGTCTGACCTTTCTTAGTTCCTCCGGCTGCTACTCTGGAT
FusionRed > Y R S K K P A T N L K M P G V Y N V D H R L E R I K E A D D E T Y
    
```

BssS1 Dra3 MscI SpDon BspE1 BseR1 SpAcc EcoR1

```

1301 CGTCGAGCAGCACGAGGTGGCTGTGGCCAGATACTCTACTGGTGGCGCTGGTGTGATGGAGGTAAAGGTGGAGGAGGTTCCGGAAAGGACGAGCTGTAAGAA
    |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||* 1400
    GCAGCTCGTCGTGCTCCACCGACACCGGTCTATGAGATGACCACCGGACCACTACCTCCATTTCCACCTCCTCCAAGGCCTTTCTGCTCGACATTCTT
FusionRed > V E Q H E V A V A R Y S T G G A G D G G K G G G S G K D E L *
    
```



```

                BtgZ1
                Dra3 |
                ||
1901  GGCCCACTACGTGAACCATCACCCCTAATCAAGTTTTTTTTGGGGTTCGAGGTGCCGTAAAGCACTAAATCGGAACCCTAAAGGGAGCCCCCGATTTAGAGCTT 2000
         |||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*||
         CCGGGTGATGCACTTGGTAGTGGGATTAGTTCAAAAAACCCCAGCTCCACGGCATTTCGTGATTTAGCCTTGGGATTTCCCTCGGGGGCTAAATCTCGAA

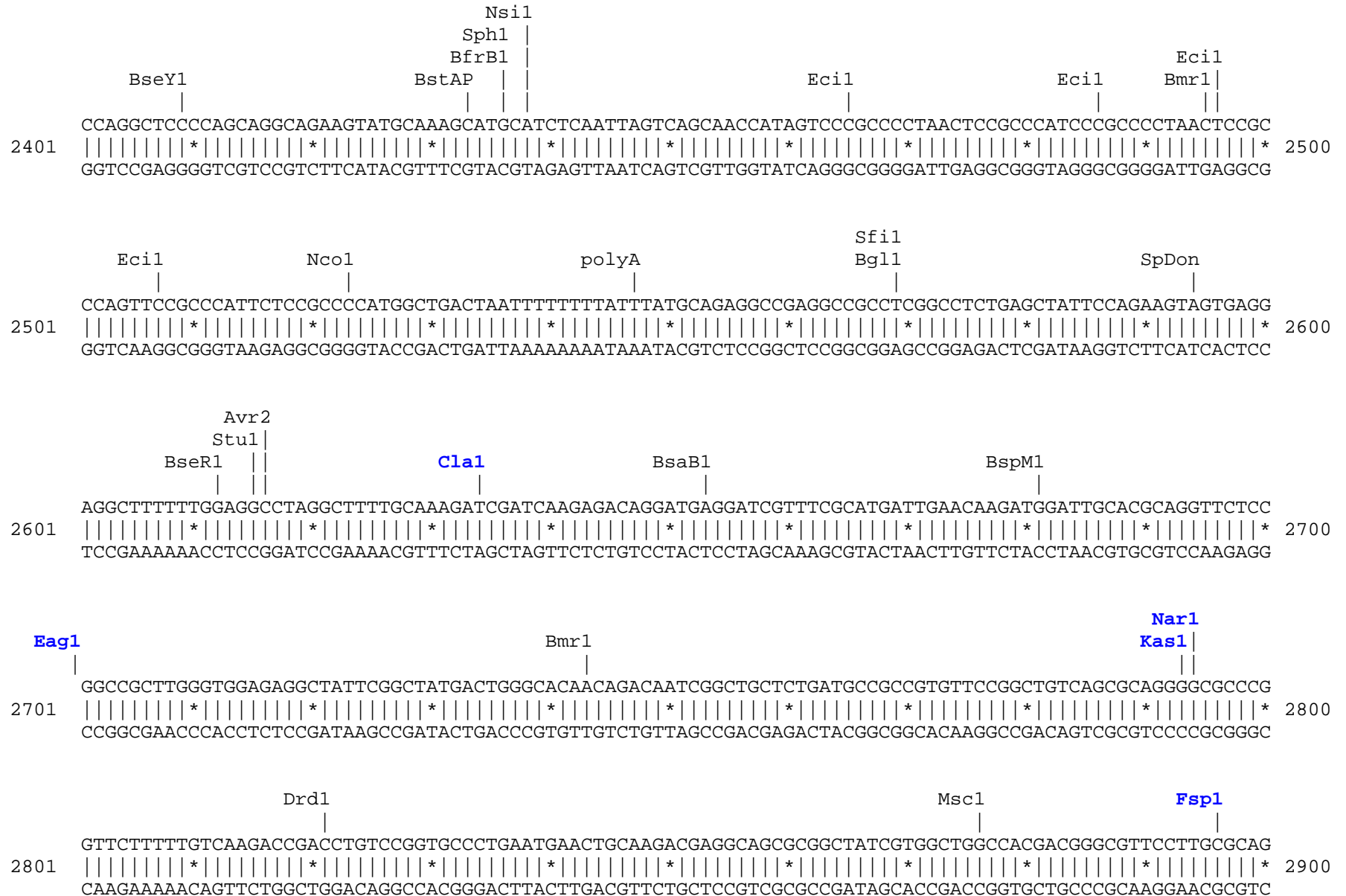
                NaeI
                NgoM4 |
                ||
                BsrB1
                |
2001  GACGGGGAAAGCCGGCGAACGTGGCGAGAAAGGAAGGAAGAAAGCGAAAGGAGCGGGCGCTAGGGCGCTGGCAAGTGTAGCGGTCCAGCTGCGCGTAAC 2100
         |||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*||
         CTGCCCTTTTCGGCCGCTTGACCAGCTCTTTCCTTCCCTTCTTTCGCTTTCTCGCCCGGATCCCACGACCGTTACATCGCCAGTGCACGCGCATTG

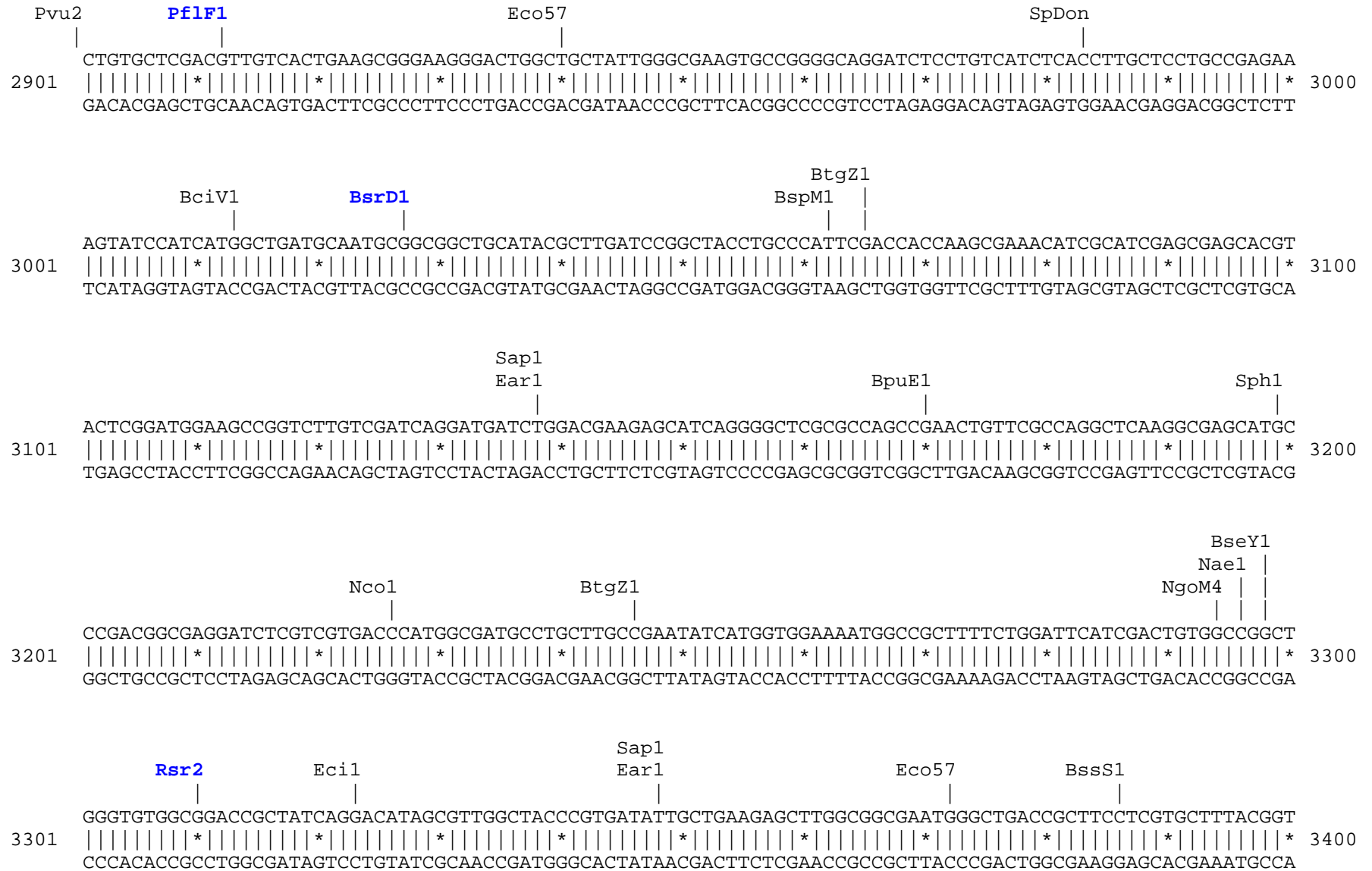
                                                                polyA
                                                                |
2101  CACCACACCCGCGCGCTTAATGCGCCGCTACAGGGCGCGTCAGGTGGCACTTTTCGGGGAAATGTGCGCGGAACCCCTATTTGTTTATTTTCTAAATA 2200
         |||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*||
         GTGGTGTGGGCGGCGGAATTACGCGCGATGTCCCGCGCAGTCCACCGTGAAAAGCCCTTTACACGCGCCTTGGGGATAAAACAAATAAAAAGATTTAT

                BciV1
                BspH1 |
                BsrB1 | |
                | |
                Ssp1 |
                Ear1 |
                Bsu36 |
                Ecil
                Pvu2 |
                |
2201  CATTCAAATATGTATCCGCTCATGAGACAATAACCCTGATAAATGCTTCAATAATATTGAAAAGGAAGAGTCCTGAGGCGGAAAGAACCAGCTGTGGAA 2300
         |||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*||
         GTAAGTTTATACATAGGCGAGTACTCTGTTATTGGGACTATTTACGAAGTTATTATAACTTTTTCTTCTCAGGACTCCGCTTTCTTGGTTCGACACCTT

                                                                NsiI
                                                                Sph1 |
                                                                BfrB1 |
                BseY1 |          BstAP |          SexA1
                |          |          |
2301  TGTGTGTCAGTTAGGGTGTGGAAAGTCCCCAGGCTCCCCAGCAGGCAGAAGTATGCAAAGCATGCATCTCAATTAGTCAGCAACCAGGTGTGGAAAGTCC 2400
         |||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*||
         ACACACAGTCAATCCCACACCTTTCAGGGGTCCGAGGGGTTCGTCCGTTTCATACGTTTTCGTACGTAGAGTTAATCAGTCGTTGGTCCACACCTTTCAGG

```





BtgZ1
 BsrB1 |
 |
 ATCGCCGCTCCCGATTTCGAGCGCATCGCCTTCTATCGCCTTCTTGACGAGTTCTTCTGAGCGGGACTCTGGGGTTTCGAAATGACCGACCAAGCGACGCC
 3401 |||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||* 3500
 TAGCGGCGAGGGCTAAGCGTCGCGTAGCGGAAGATAGCGGAAGAACTGCTCAAGAAGACTCGCCCTGAGACCCCAAGCTTTACTGGCTGGTTTCGCTGCGG

BssS1
 BspM1 |
 |
 CAACCTGCCATCAGGAGATTTTCGATTCCACCGCCGCCTTCTATGAAAGGTTGGGCTTCGGAATCGTTTTTCGGGACGCCGGCTGGATGATCCTCCAGCGC
 3501 |||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||* 3600
 GTTGGACGGTAGTGCTCTAAAGCTAAGGTGGCGGCGGAAGATACTTTCCAACCCGAAGCCTTAGCAAAAGGCCCTGCGGCCGACCTACTAGGAGGTTCGG

Bpm1
 SpAcc |
 Avr2 |
 |
 GGGGATCTCATGCTGGAGTTCTTCGCCCACCCTAGGGGGAGGCTAACTGAAACACGGAAGGAGACAATACCGGAAGGAACCCGCGCTATGACGGCAATAA
 3601 |||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||* 3700
 CCCCTAGAGTACGACCTCAAGAAGCGGGTGGGATCCCCCTCCGATTGACTTTGTGCCTTCTCTGTTATGGCCTTCTTGGGGCGGATACTGCCGTTATT

polyA
 |
 AAAGACAGAATAAAAACGCACGGTGTTGGGTTCGTTTGTTCATAAAACGCGGGGTTTCGGTCCCAGGGCTGGCACTCTGTTCGATAACCCACCGAGACCCCATTTG
 3701 |||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||* 3800
 TTTCTGTCTTATTTTTCGCTGCCACAACCCAGCAAACAAGTATTTGCGCCCAAGCCAGGGTCCCAGCGTGCAGACAGCTATGGGGTGGCTCTGGGGTAAC

BstAP
 AlwN1 |
 |
 GGGCCAATACGCCCCGCTTCTTCTTTTCCCCACCCCAAGTTTCGGGTGAAGGCCAGGGCTCGCAGCCAACGTCGGGGCGGCAGGCCCTGCC
 3801 |||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||*|||||||* 3900
 CCCGGTTATGCGGGCGCAAAGAAGGAAAAGGGGTGGGGTGGGGGTTCAAGCCACTTCCGGGTCCCAGCGTTCGGTTGCAGCCCCGCCGTCCGGGACGG


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      Bsu36                      DraI          DraI                      BspH1
      |                          |            |                          |
3901  ATAGCCTCAGGTTACTCATATATACTTTAGATTGATTTTAAAACCTTCATTTTTTAATTTAAAAGGATCTAGGTGAAGATCCTTTTTTGATAATCTCATGACCA 4000
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*
      TATCGGAGTCCAATGAGTATATATGAAATCTAACTAAATTTTGAAGTAAAAATTAATTTTCTAGATCCACTTCTAGGAAAAACTATTAGAGTACTGGT

                                     BpuE1
                                     |
4001  AAATCCCTTAACGTGAGTTTTTCGTTCCACTGAGCGTCAGACCCCGTAGAAAAGATCAAAGGATCTTCTTGAGATCCTTTTTTTCTGCGCGTAATCTGCTG 4100
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*
      TTTAGGGAATTGCACTCAAAGCAAGGTGACTCGCAGTCTGGGGCATCTTTTCTAGTTTCTAGAAAGAACTCTAGGAAAAAAGACGCGCATTAGACGAC

                                               Eco57
                                               |
4101  CTTGCAAACAAAAAACCACCGCTACCAGCGGTGGTTTTGTTTGC CGGATCAAGAGCTACCAACTCTTTTTCCGAAGGTAAGTGGCTTCCAGCAGAGCGCAG 4200
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*
      GAACGTTTGTTTTTTGGTGGCGATGGTCGCCACCAAACAAACGGCCTAGTTCTCGATGGTTGAGAAAAAGGCTTCCATTGACCGAAGTCGTCTCGCGTC

                SpAcc
                |
4201  ATACCAAATACTGTCCTTCTAGTGTAGCCGTAGTTAGGCCACCACTTCAAGAACTCTGTAGCACCGCTACATACCTCGCTCTGCTAATCCTGTTACCAG 4300
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*
      TATGGTTTATGACAGGAAGATCACATCGGCATCAATCCGGTGGTGAAGTTCTTGAGACATCGTGGCGGATGTATGGAGCGAGACGATTAGGACAATGGTC

AlwN1                      BpuE1                      ApaL1
|                          |                          |
4301  TGGCTGCTGCCAGTGGCGATAAGTCGTGTCTTACCGGGTTGGACTCAAGACGATAGTTACCGGATAAGGCGCAGCGGTCTGGGCTGAACGGGGGGTTCGTG 4400
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*
      ACCGACGACGGTCACCGCTATTTCAGCACAGAATGGCCAACTGAGTTCTGCTATCAATGGCCTATTCCGCGTCGCCAGCCCGACTTGCCCCCAAGCAC

      BseY1                      SpAcc
      |                          |
4401  CACACAGCCCAGCTTGGAGCGAACGACCTACACCGAACTGAGATACCTACAGCGTGAGCTATGAGAAAGCGCCACGCTTCCCGAAGGGAGAAAGGCGGAC 4500
      |||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*|||||*
      GTGTGTCGGGTCGAACCTCGCTTGCTGGATGTGGCTTGACTCTATGGATGTGCACTCGATACTCTTTTCGCGGTGCGAAGGGCTTCCCTCTTTCCGCTG
```

```
           BciV1
           EciI      |
           |         |
           |         |
AGGTATCCGGTAAGCGGCAGGGTCGGAACAGGAGAGCGCACGAGGGAGCTTCCAGGGGGAAAACGCCTGGTATCTTTATAGTCCTGTTCGGGTTTCGCCACC
4501  |||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 4600
TCCATAGGCCATTTCGCCGTCCCAGCCTTGTCTCTCGCGTGCTCCCTCGAAGGTCCCCCTTTGCGGACCATAGAAATATCAGGACAGCCCCAAAGCGGTGG
```

```
           DrdI      |
           |         |
           |         |
           |         |
TCTGACTTGAGCGTCGATTTTTGTGATGCTCGTCAGGGGGCGGAGCCTATGGAAAAACGCCAGCAACGCGGCCTTTTTACGGTTCCTGGCCTTTTGTCTG
4601  |||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 4700
AGACTGAACTCGCAGCTAAAAAACTACGAGCAGTCCCCCGCCTCGGATACCTTTTTGCGGTCGTTGCGCCGGAAAAATGCCAAGGACCGGAAAAACGAC
```

```
           SpDon      |
           BspLU      |
           ||         |
           |         |
           |         |
           |         |
GCCTTTTGCTCACATGTTCTTTCTGCGTTATCCCCTGATTCTGTGGATAACCGTATTACCGCCATGCAT
4701  |||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*||||||*|| 4770
CGGAAAACGAGTGTACAAGAAAGGACGCAATAGGGGACTAAGACACCTATTGGCATAATGGCGGTACGTA
```

Found:

Aat2	Acc65	Age1	Ale1	AlwN1	Apa1	ApaL1	Ase1	Avr2	BamH1	Bbs1	BciV1	Bcl1	BfrB1
Bgl1	Bmr1	Bpm1	BpuE1	Bsa1	BsaB1	BsaXa	BsaXb	BseR1	BseY1	Bsm1	BsmB1	BspE1	BspH1
BspLU	BspM1	BsrB1	BsrD1	BsrG1	BssS1	BstAP	BstB1	Bsu36	BtgZ1	Bts1	Clal	Dra1	Dra3
Drd1	Eag1	Ear1	Ecil	Eco57	EcoR1	Fsp1	Hpa1	Kas1	Kpn1	Mfe1	Mlu1	Msc1	Nae1
Nar1	Nco1	Nde1	NgoM4	Nhe1	Nsi1	PflF1	PflM1	polyA	PshA1	Psi1	PspOM	Pst1	Pvu2
Rsr2	Sac2	Sall	Sap1	SexA1	Sfi1	Sma1	SnaB1	SpAcc	SpDon	Sph1	Ssp1	Stu1	Xba1

Unique:

Acc65	Age1	Ale1	Apa1	Ase1	BamH1	Bbs1	Bcl1	BsaXa	BsaXb	BsmB1	BspE1	BspLU	BsrD1
BsrG1	BstB1	Bts1	Clal	Eag1	EcoR1	Fsp1	Hpa1	Kas1	Kpn1	Mfe1	Mlu1	Nar1	Nde1
Nhe1	PflF1	PflM1	PshA1	PspOM	Pst1	Rsr2	Sac2	Sall	SexA1	Sma1	SnaB1	Xba1	

Not found:

Aar1	Acl1	Afe1	Afl2	Ahd1	Asc1	AsiS1	Baela	Baelb	BbvC1	BcglA	Bcglb	Bgl2	Blp1
BmgB1	Bpu10	Bsg1	BsiW1	BssH2	BstE2	BstX1	BstZ1	BxatB	BxatL	BxatR	BxatP	_Chi	Ecl2
EcoK	EcoN1	EcoRV	FCatB	FCatL	FCatR	FCatP	ScFRT	Fse1	FspA1	Hind3	I_Ceu	loxP	Not1
Nru1	Pac1	Pme1	Pml1	Pvu1	R4atB	R4atL	R4atP	R4atR	Sac1	SanD1	Sbf1	Sca1	Sgf1
SgrA1	Spe1	Srf1	Swal	T3RNA	T7RNA	T7Ter	PISce	Xcm1	Xho1	Xmn1			

Excluded by site complexity:

Acc1	Acil	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	Hinfl	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													