

Profiling of basal and ligand-dependent GPCR activities by means of a polyvalent cell-based high-throughput platform

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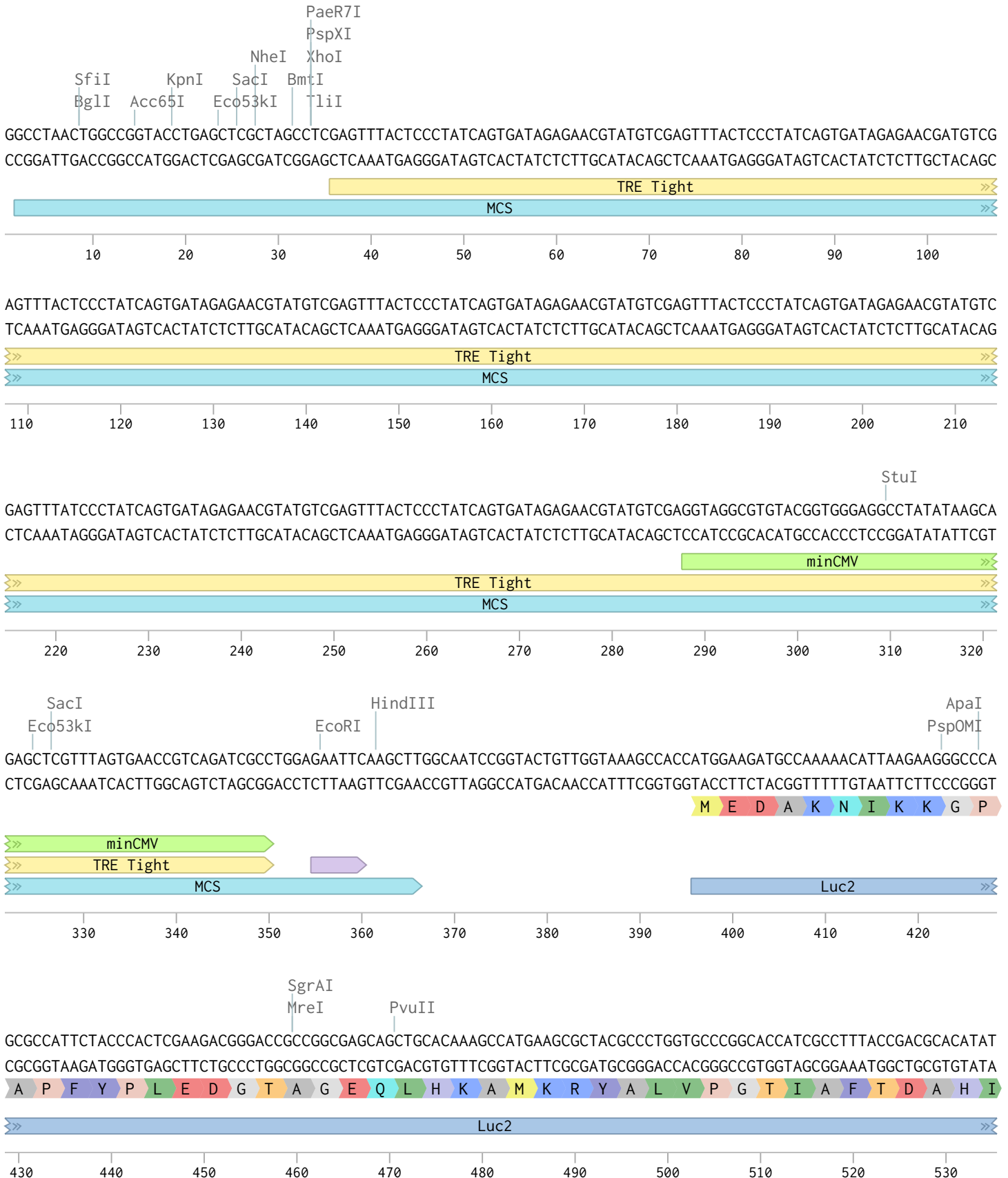
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² Brain and Mind Research Institute, University of Ottawa, Ottawa, ON, K1H8M5, Canada

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Supplementary File 2. Sequences of Tango-Trio constructs stably expressed in HTTL, HTTL-B1, HTTL-B2, and HTTL-F cell lines. Information regarding the cloning of the plasmids is provided in the Methods.

Tight-pNLCol1-Luc2 (6844 bp)



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GCTCCACCTGTAATGGATGCGGCTCATGAAGCTCTACTCGAAGCCGACCGTCTTCGATACTTCGCGATACCCGACTTATGTTTGGTAGCCTAGCACCACACGTCGC
E V D I T Y A E Y F E M S V R L A E A M K R Y G L N T N H R I V V C S

» Luc2 »

540 550 560 570 580 590 600 610 620 630 640

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E N S L Q F F M P V L G A L F I G V A V A P A N D I Y N E R E L L N S M

» Luc2 »

650 660 670 680 690 700 710 720 730 740

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G I S Q P T V V F V S K K G L Q K I L N V Q K K L P I I Q K I I I M D S

» Luc2 »

750 760 770 780 790 800 810 820 830 840 850

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K T D Y Q G F Q S M Y T F V T S H L P P G F N E Y D F V P E S F D R D

» Luc2 »

860 870 880 890 900 910 920 930 940 950 960

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K T I A L I M N S S G S T G L P K G V A L P H R T A C V R F S H A R D P

» Luc2 »

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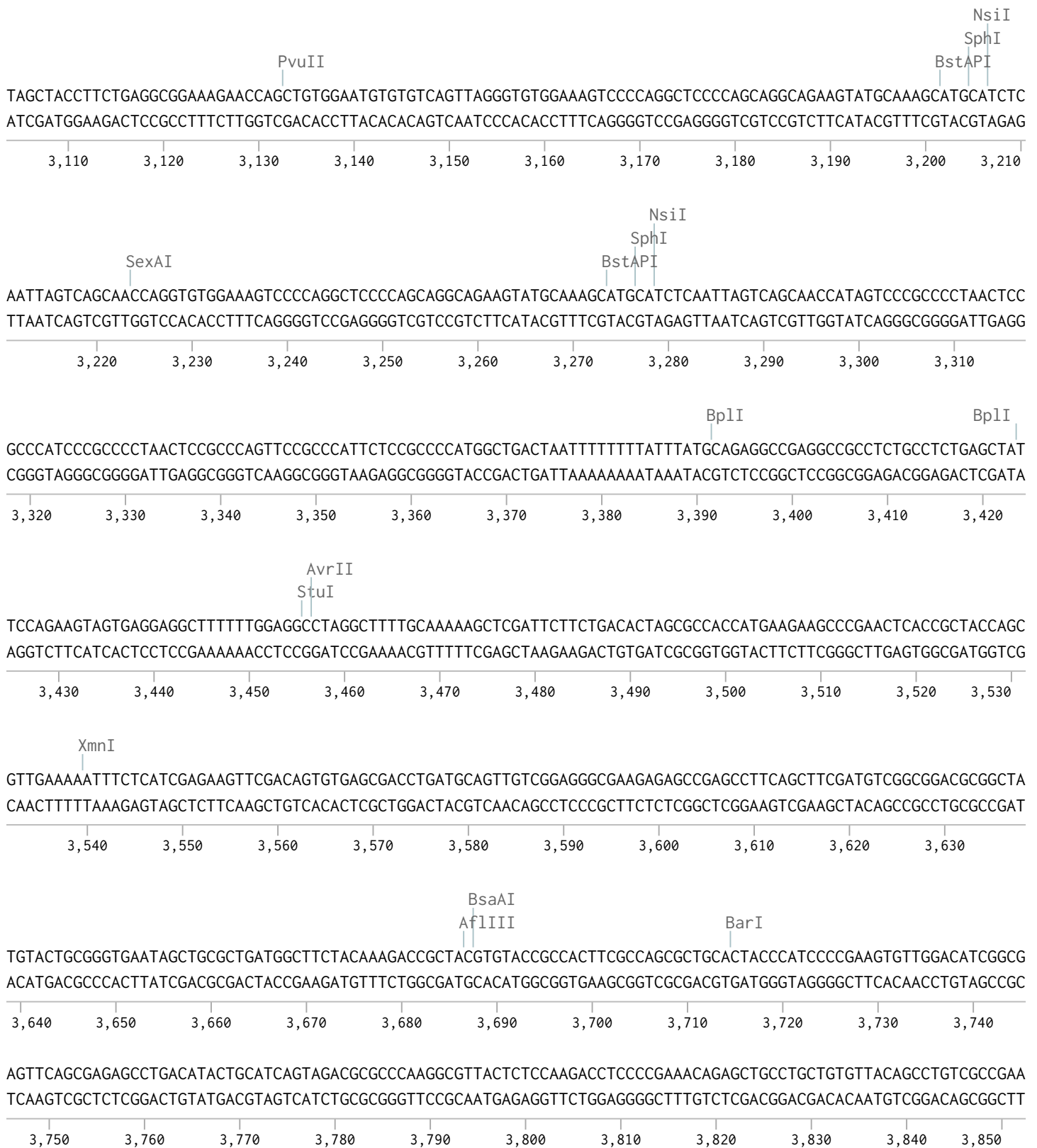
» Luc2 »

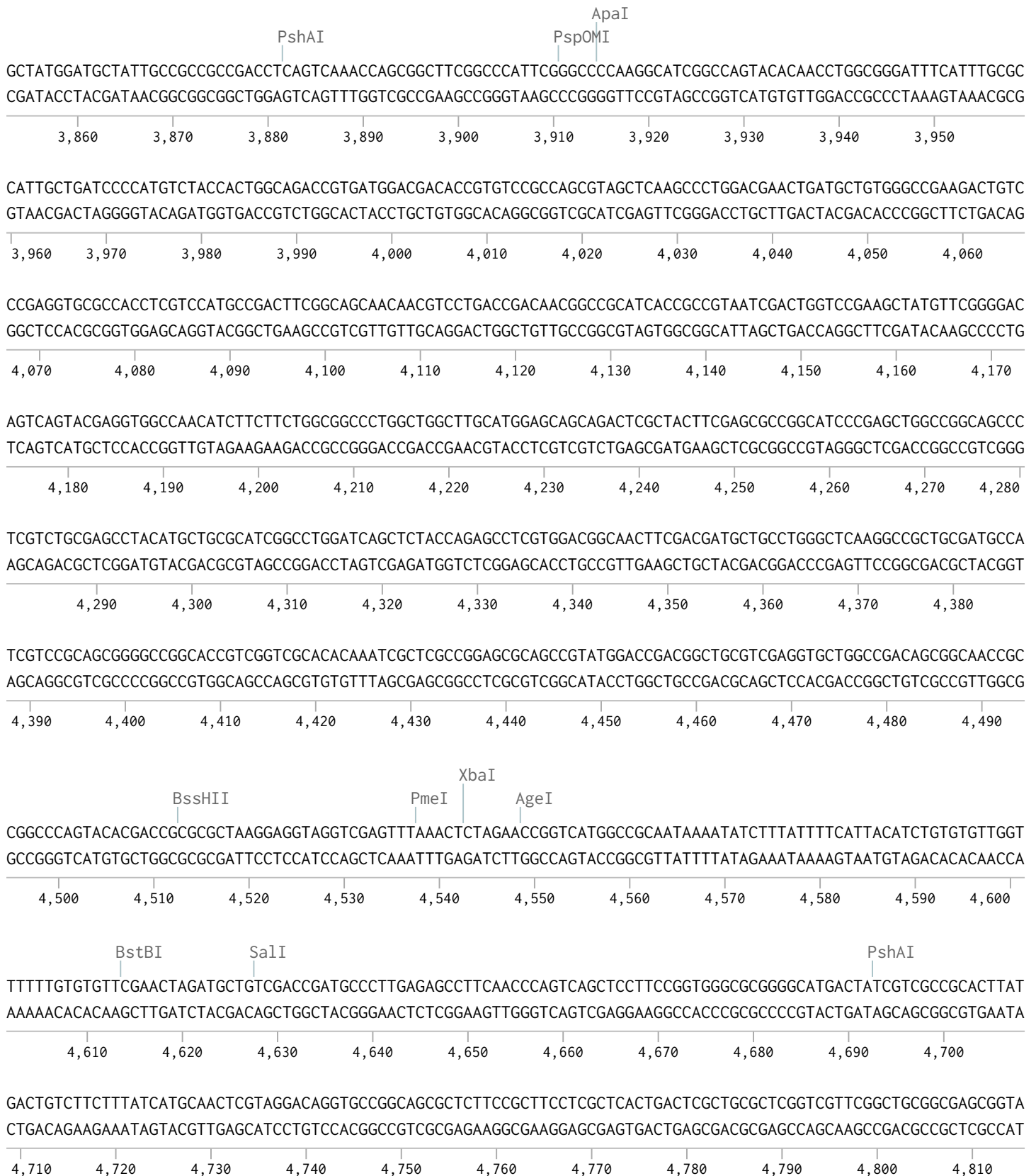
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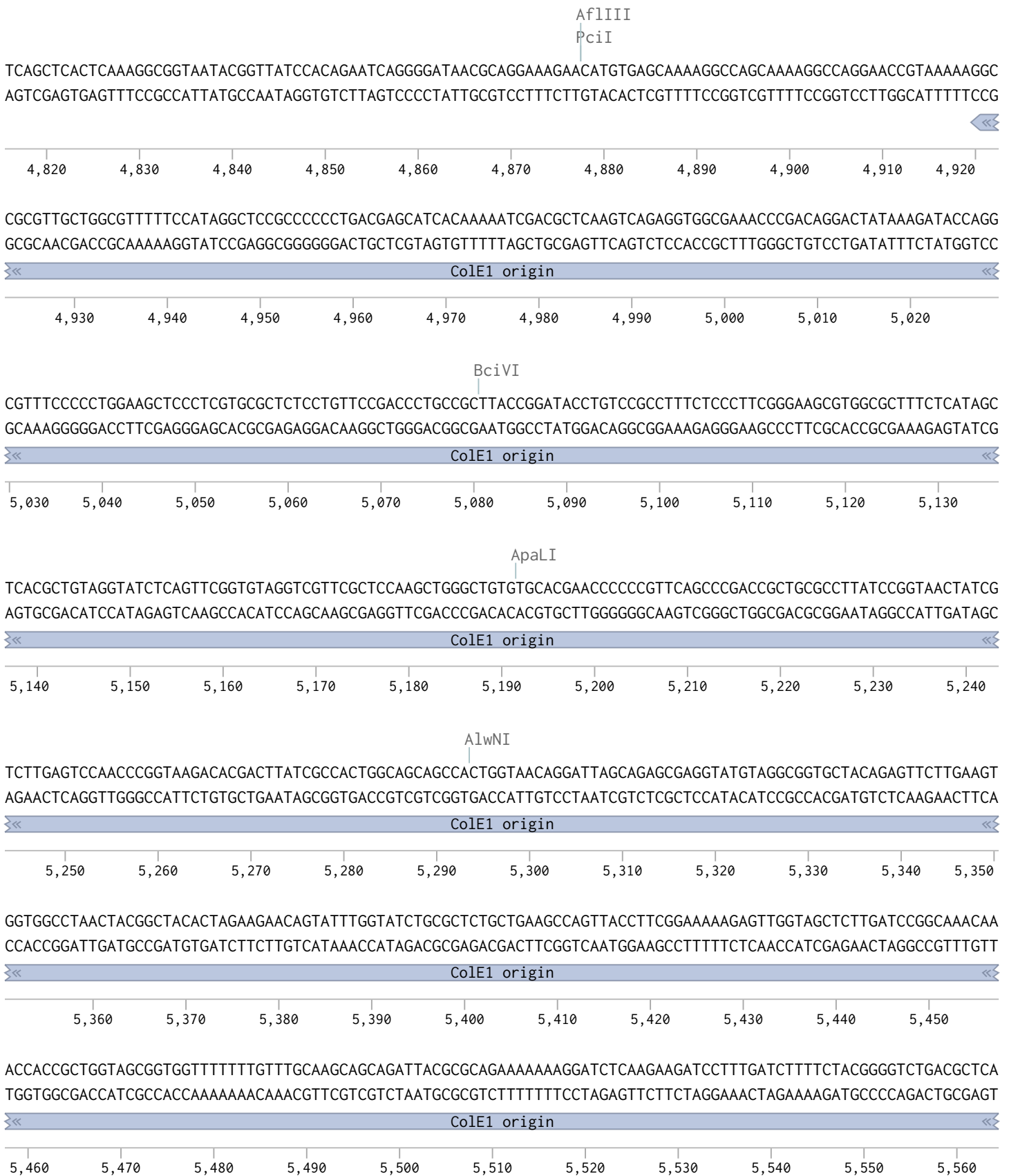












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ColE1 origin

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BstEII
BstXI
PstI
NotI
AclI
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Bsu36I
PvuI
SacII
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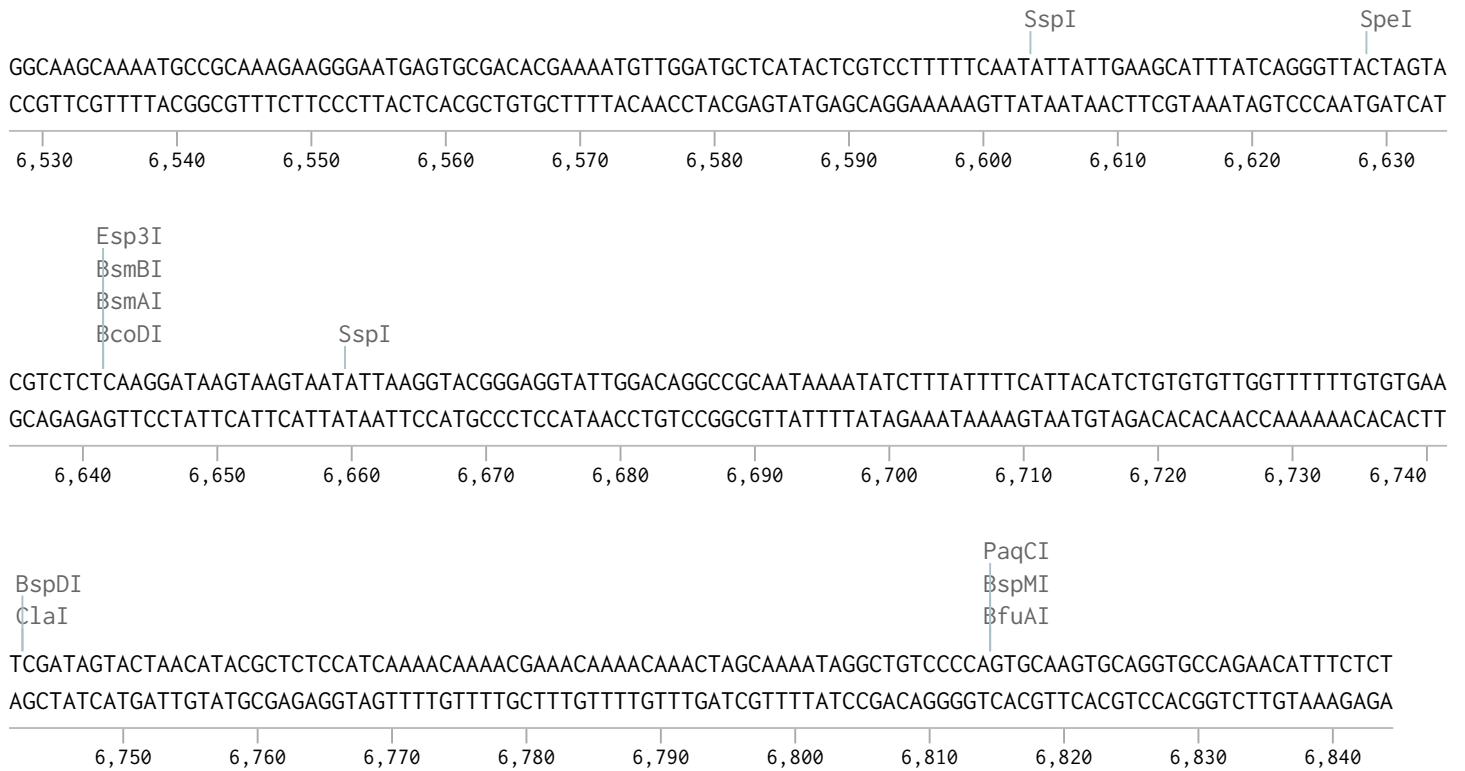
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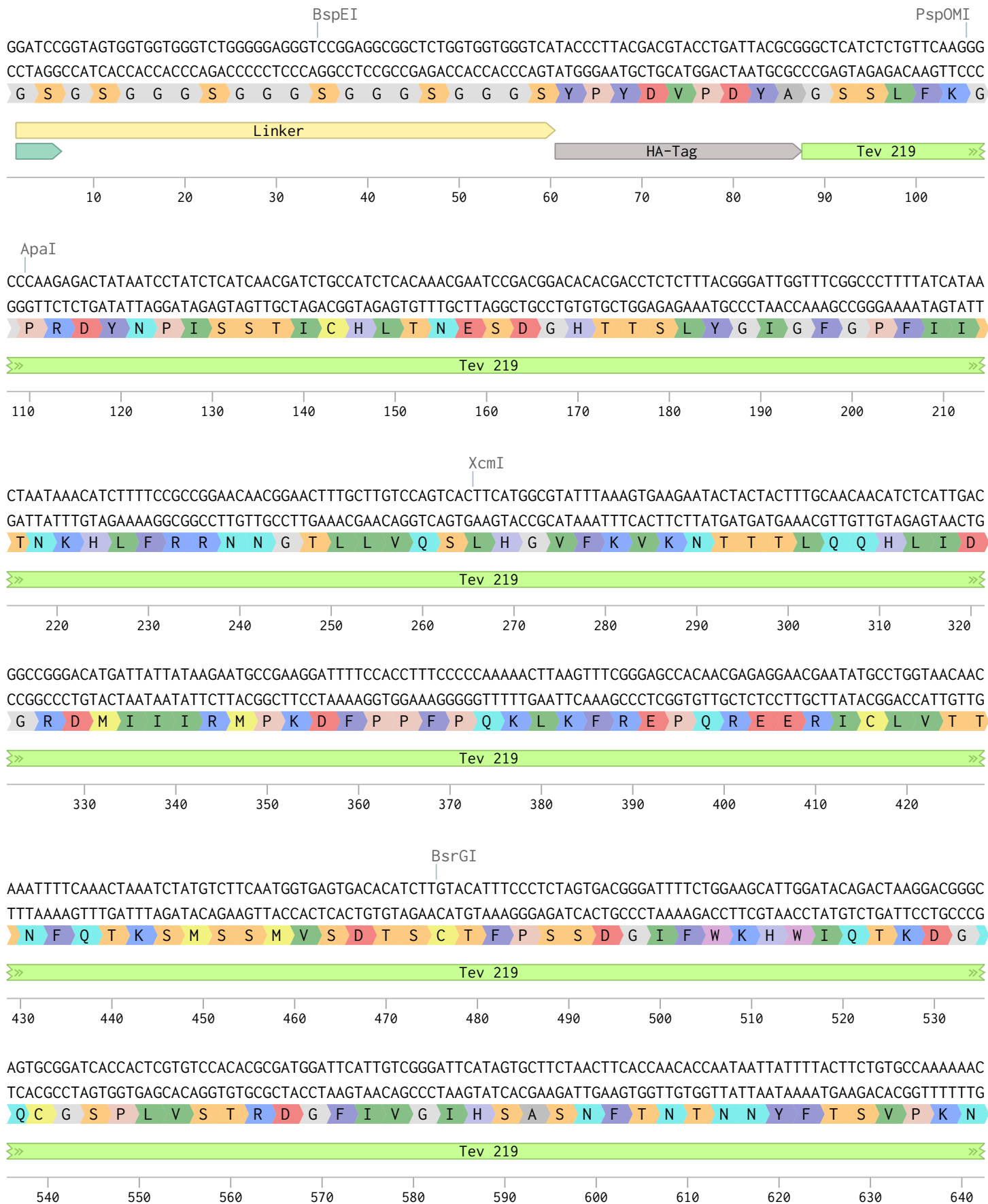
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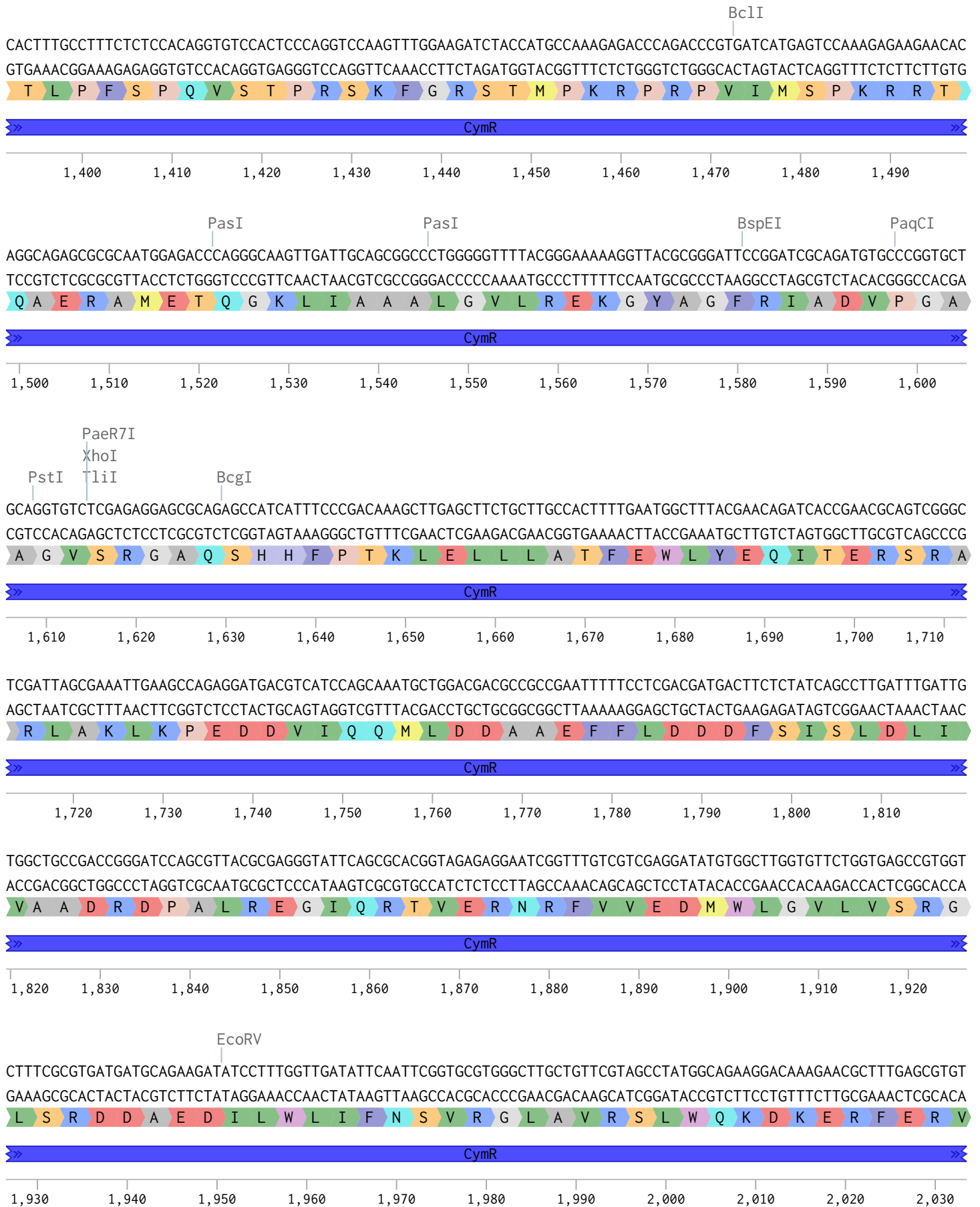
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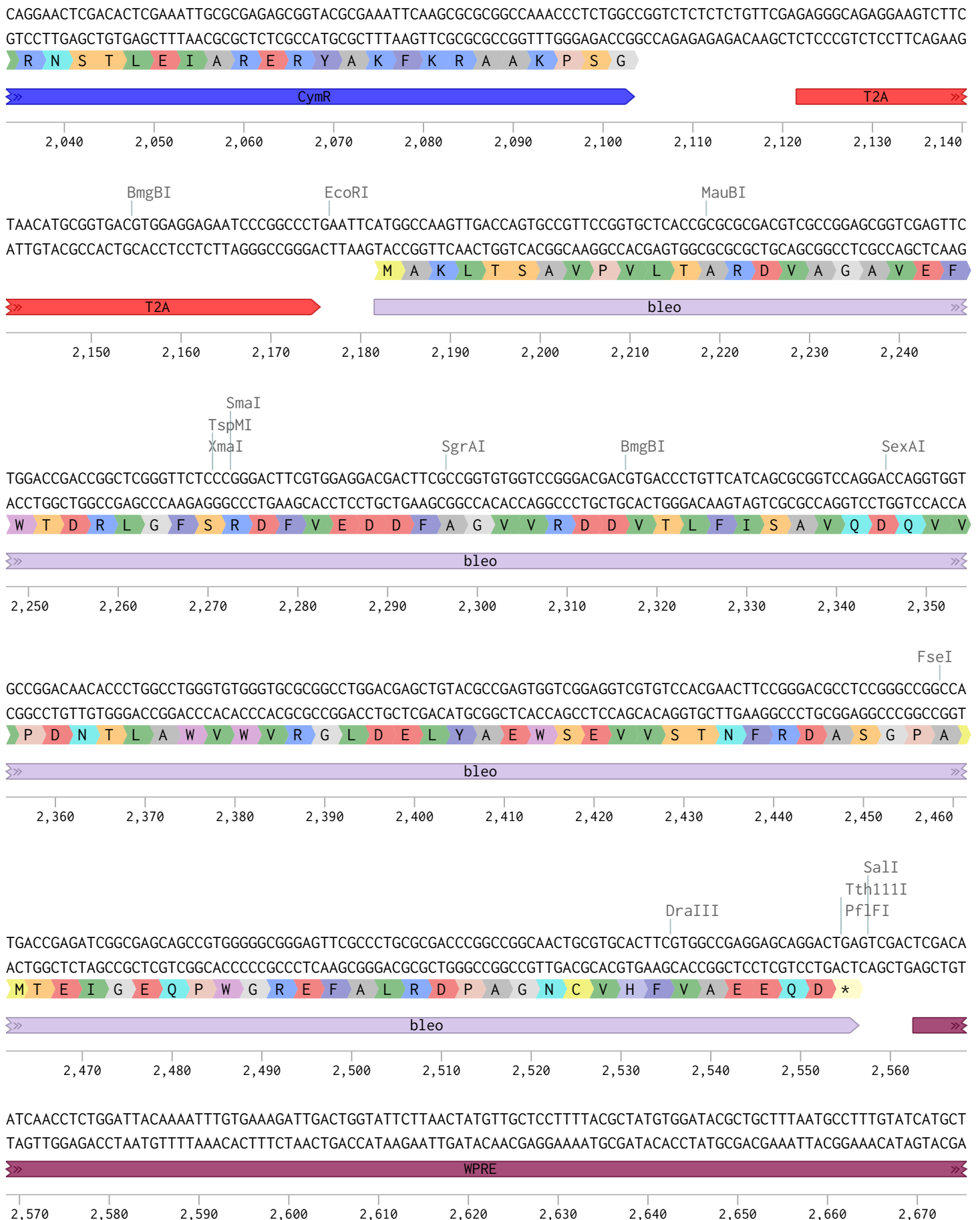
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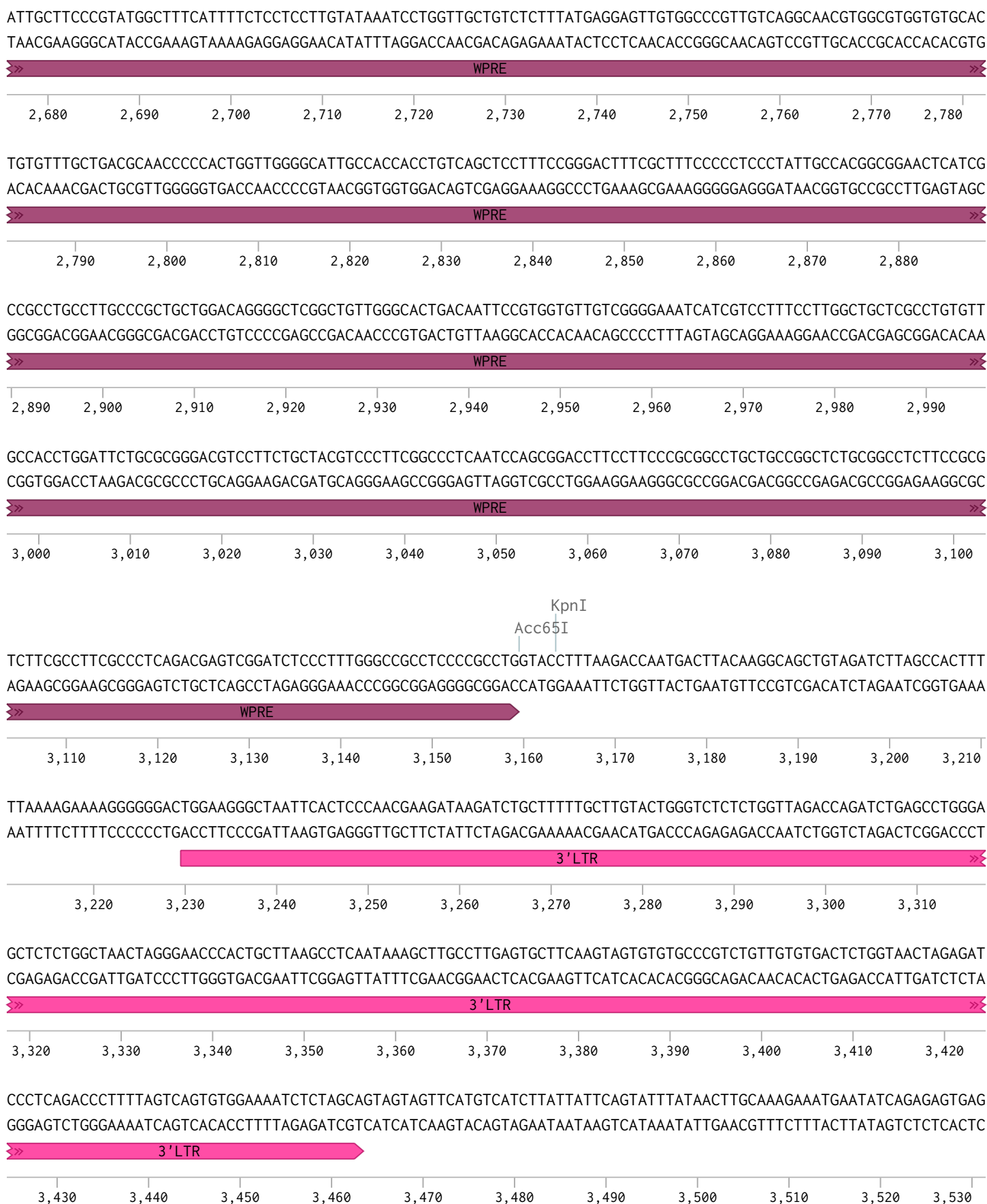


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SV40 Polyadenylation signal

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SV40 Polyad...ion signal

SV40 Ori

3,640 3,650 3,660 3,670 3,680 3,690 3,700 3,710 3,720 3,730 3,740

SfiI

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SV40 Ori

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3,960 3,970 3,980 3,990 4,000 4,010 4,020 4,030 4,040 4,050 4,060

BspQI

SapI

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AflIII

PciI

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pUC ori

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pUC ori

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«« pUC ori »»

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«« pUC ori »»

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«« pUC ori »»

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«« pUC ori »»

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«« pUC ori »»

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»»

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AhdI

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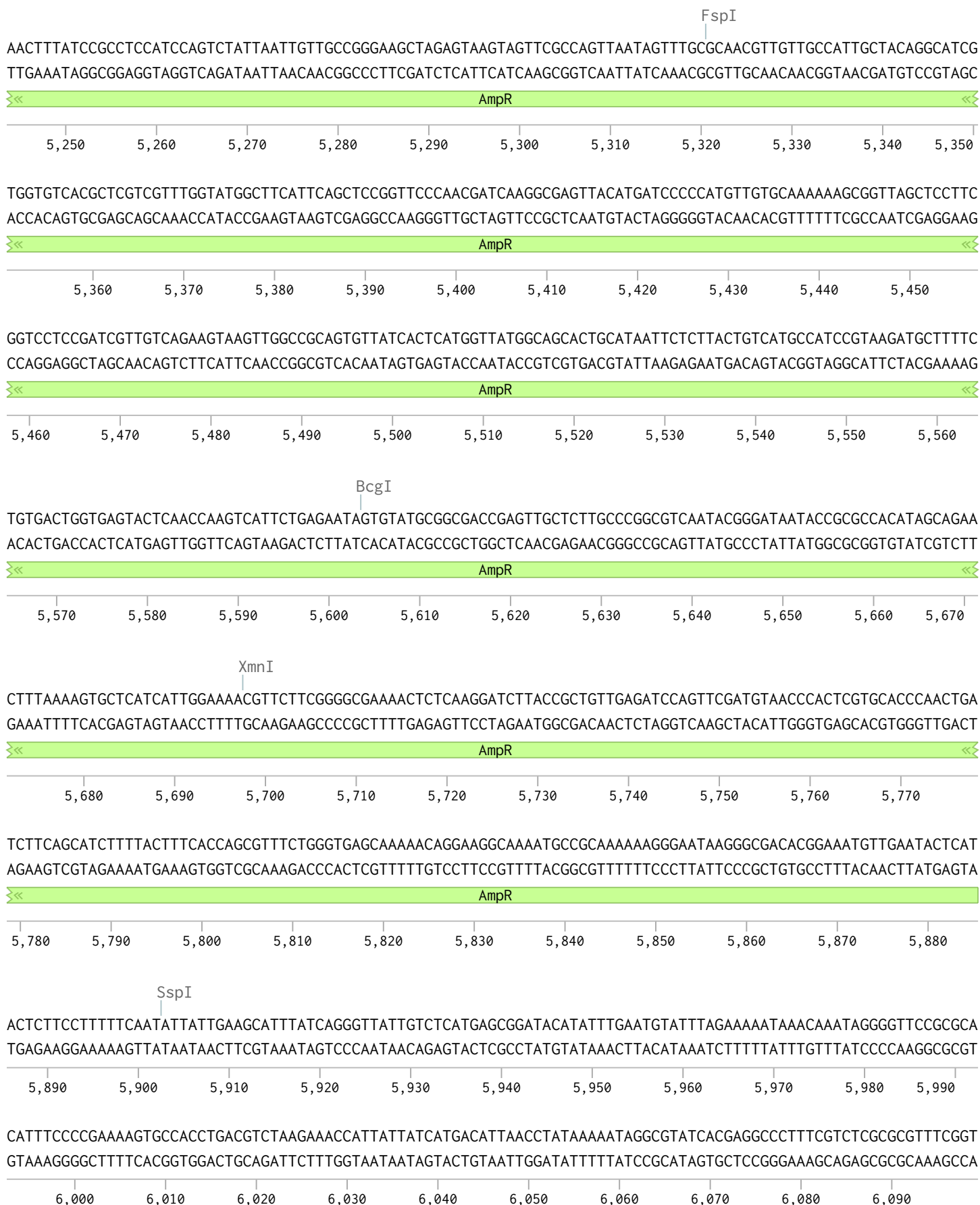
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«« AmpR »»

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NdeI

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FspI

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AflIII
MluI

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SphI

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RSV

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BaeI

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RSV HIV LTR

6,640 6,650 6,660 6,670 6,680 6,690 6,700 6,710 6,720 6,730 6,740

HIV LTR

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HIV LTR

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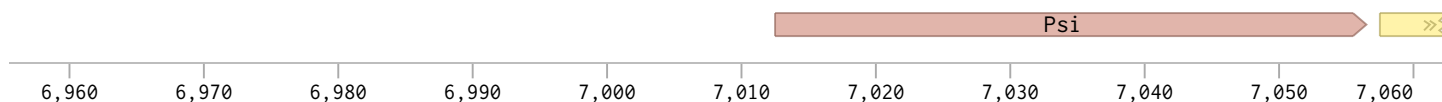
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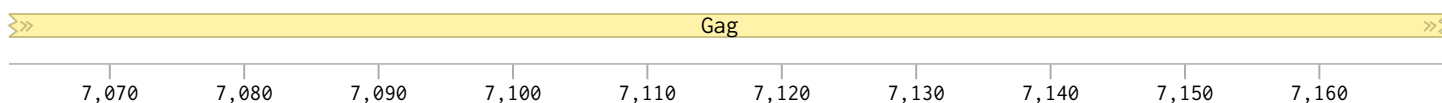
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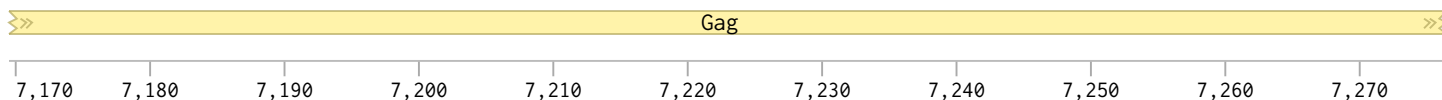
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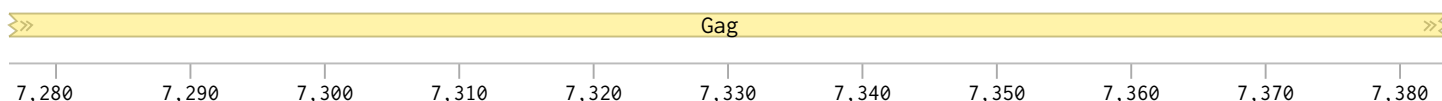
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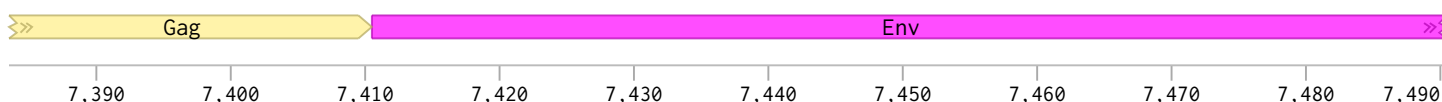
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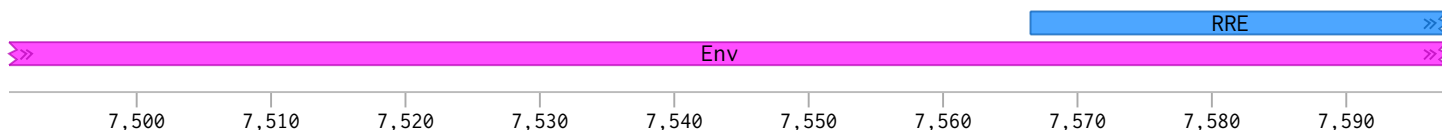
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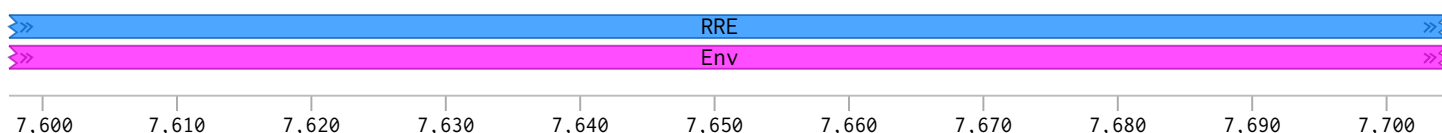
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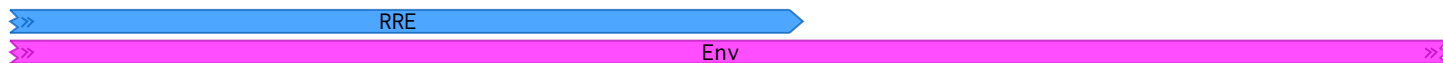


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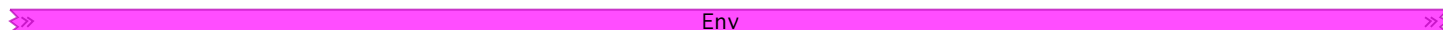
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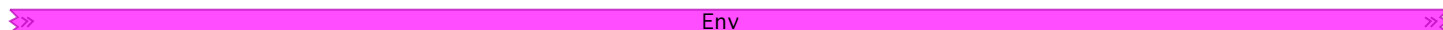
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7,920 7,930 7,940 7,950 7,960 7,970 7,980 7,990 8,000 8,010 8,020

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8,030 8,040 8,050 8,060 8,070 8,080 8,090 8,100 8,110 8,120 8,130

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8,140 8,150 8,160 8,170 8,180 8,190 8,200 8,210 8,220 8,230

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8,240 8,250 8,260 8,270 8,280 8,290 8,300 8,310 8,320 8,330 8,340

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8,350 8,360 8,370 8,380 8,390 8,400 8,410 8,420 8,430 8,440 8,450

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» CMV + CUO »

8,460 8,470 8,480 8,490 8,500 8,510 8,520 8,530 8,540 8,550 8,560

NdeI

GTTCCCATAGTAACGCCAATAGGGACTTTCCATTGACGTCAATGGGTGGAGTATTTACGGTAAACTGCCACTTGGCAGTACATCAAGTGTATCATATGCCAAGTCC
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» CMV + CUO »

8,570 8,580 8,590 8,600 8,610 8,620 8,630 8,640 8,650 8,660

SnaBI

GCCCCCTATTGACGTCAATGACGGTAAATGGCCCGCCTGGCATTATGCCCAGTACATGACCTTACGGGACTTTCCTACTTGGCAGTACATCTACGTATTAGTCATCG
CGGGGATAACTGCAGTTACTGCCATTTACCGGGCGGACCGTAATACGGGTCTGTAAGTGAATGCCCTGAAAGGATGAACCGTCATGTAGATGCATAATCAGTAGC

» CMV + CUO »

8,670 8,680 8,690 8,700 8,710 8,720 8,730 8,740 8,750 8,760 8,770

CspCI

CTATTACCATGGTGATGCGGTTTTGGCAGTACACCAATGGGCGTGGATAGCGGTTTGACTCACGGGGATTTCCAAGTCTCCACCCATTGACGTCAATGGGAGTTTG
GATAATGGTACCACTACGCCAAAACCGTCATGTGGTTACCCGCACCTATCGCCAAACTGAGTGCCCTAAAGGTTTCAGAGGTGGGGTAAGTGCAGTTACCCTCAAAC

» CMV + CUO »

8,780 8,790 8,800 8,810 8,820 8,830 8,840 8,850 8,860 8,870 8,880

TTTTGGACCAAAATCAACGGGACTTTCCAAAATGTCGTAATACCCCCGCCCCGTTGACGCAAAATGGGCAAGCTTGCCGGGTCGAGGTAGGCGGTGACGGTGGGAGG
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» CMV + CUO » CMV + CUO »

8,890 8,900 8,910 8,920 8,930 8,940 8,950 8,960 8,970 8,980

AgeI

AgeI

AscI

CCTATATAAGCAACCGGTATAATCAAACAGACCAGATTGTCTGTTTGTACCGGTGTTTAGTGAACCGGGCGCGCCTCATATCGCCTGGAGACGCCATCCACGCTGT
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» CMV + CUO »

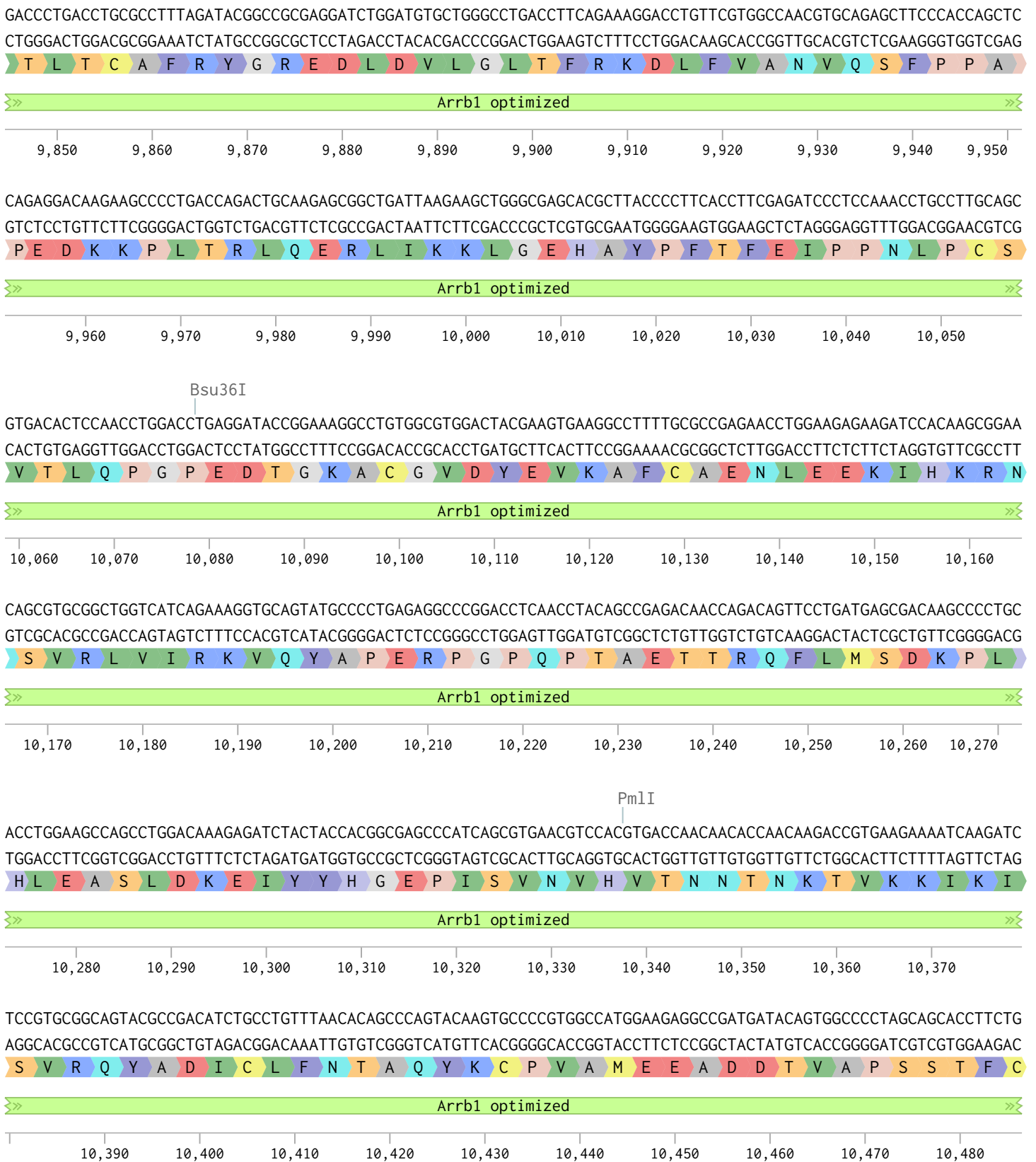
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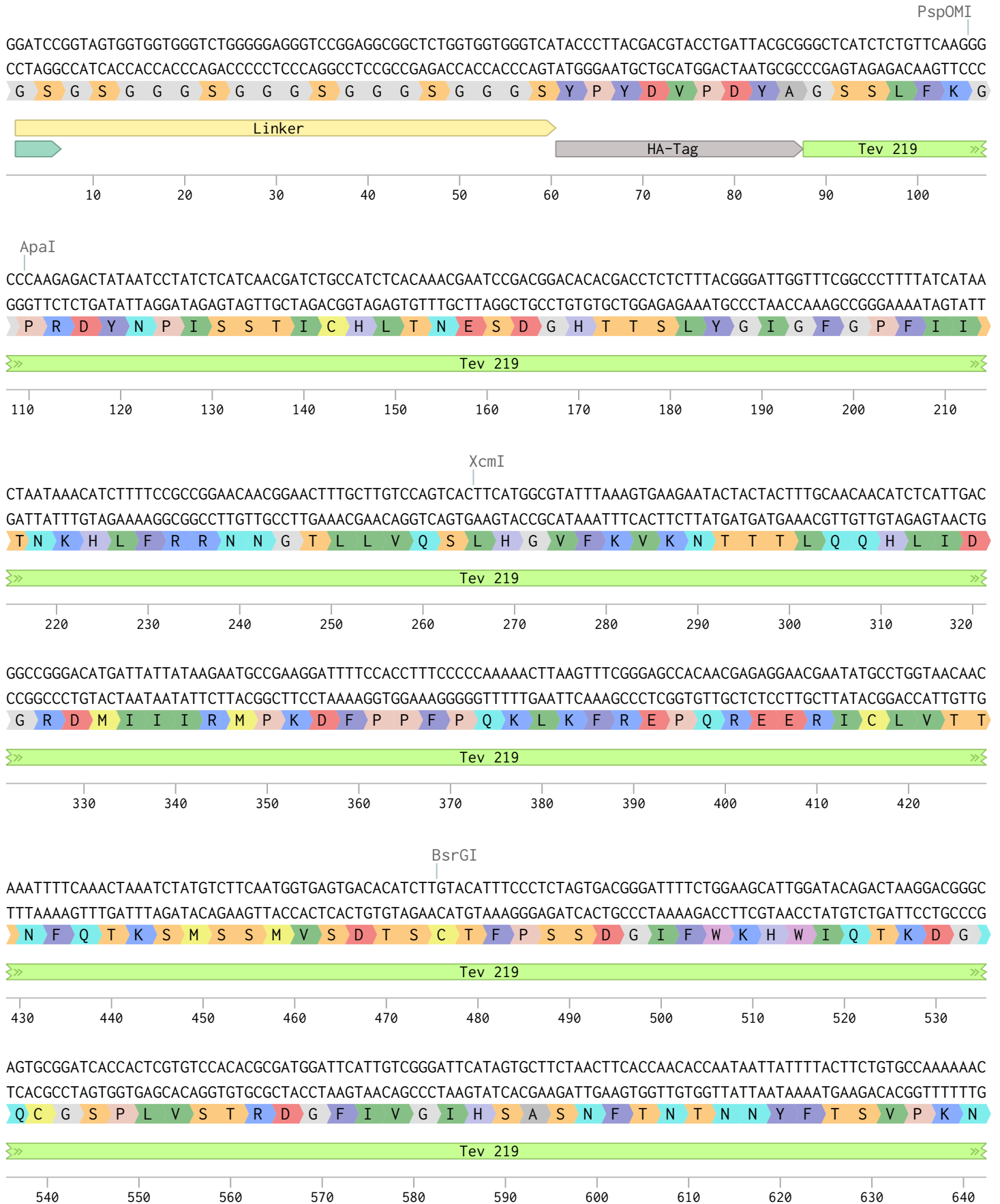
» CMV + CUO »

9,100 9,110 9,120 9,130 9,140 9,150 9,160 9,170 9,180 9,190 9,200

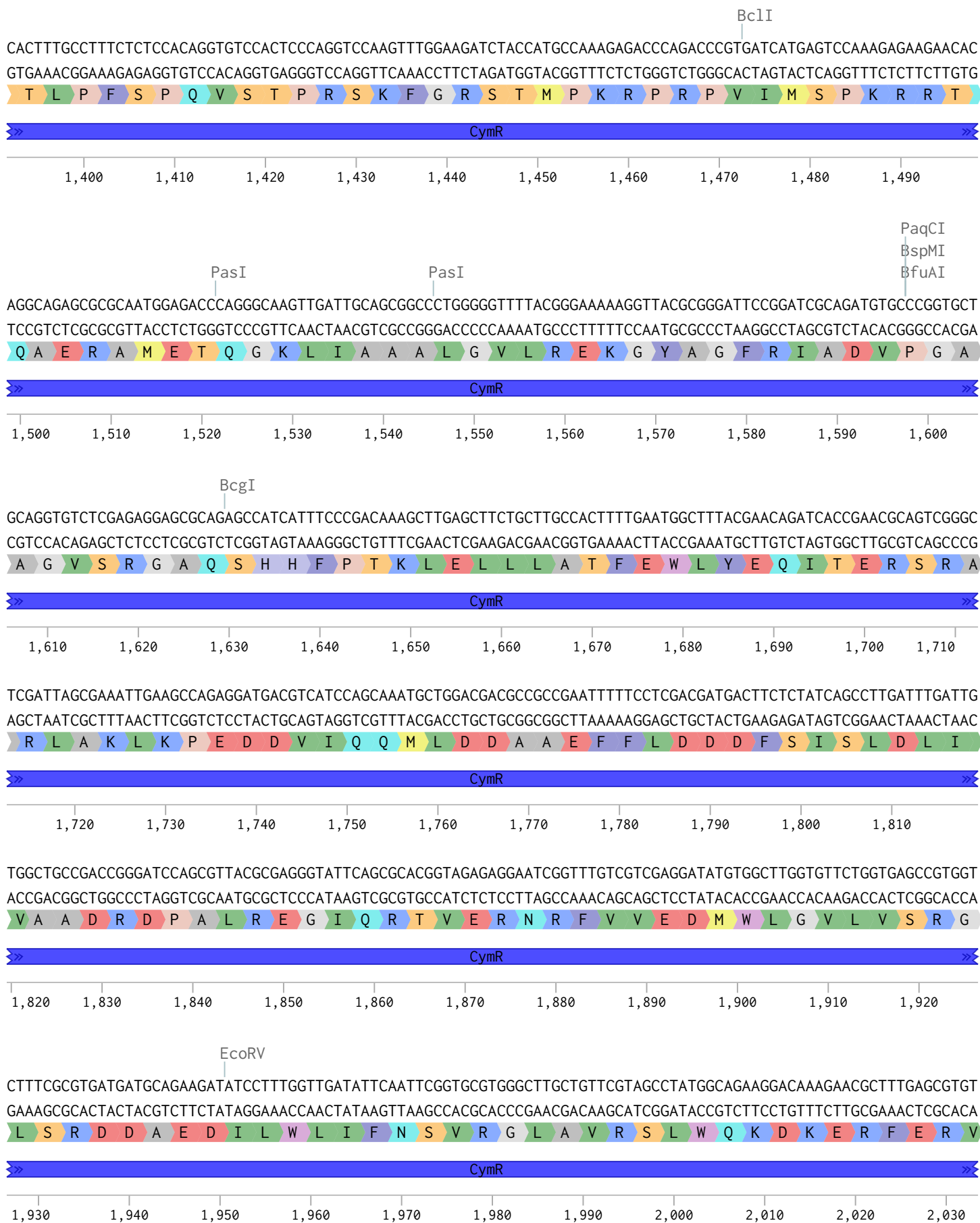


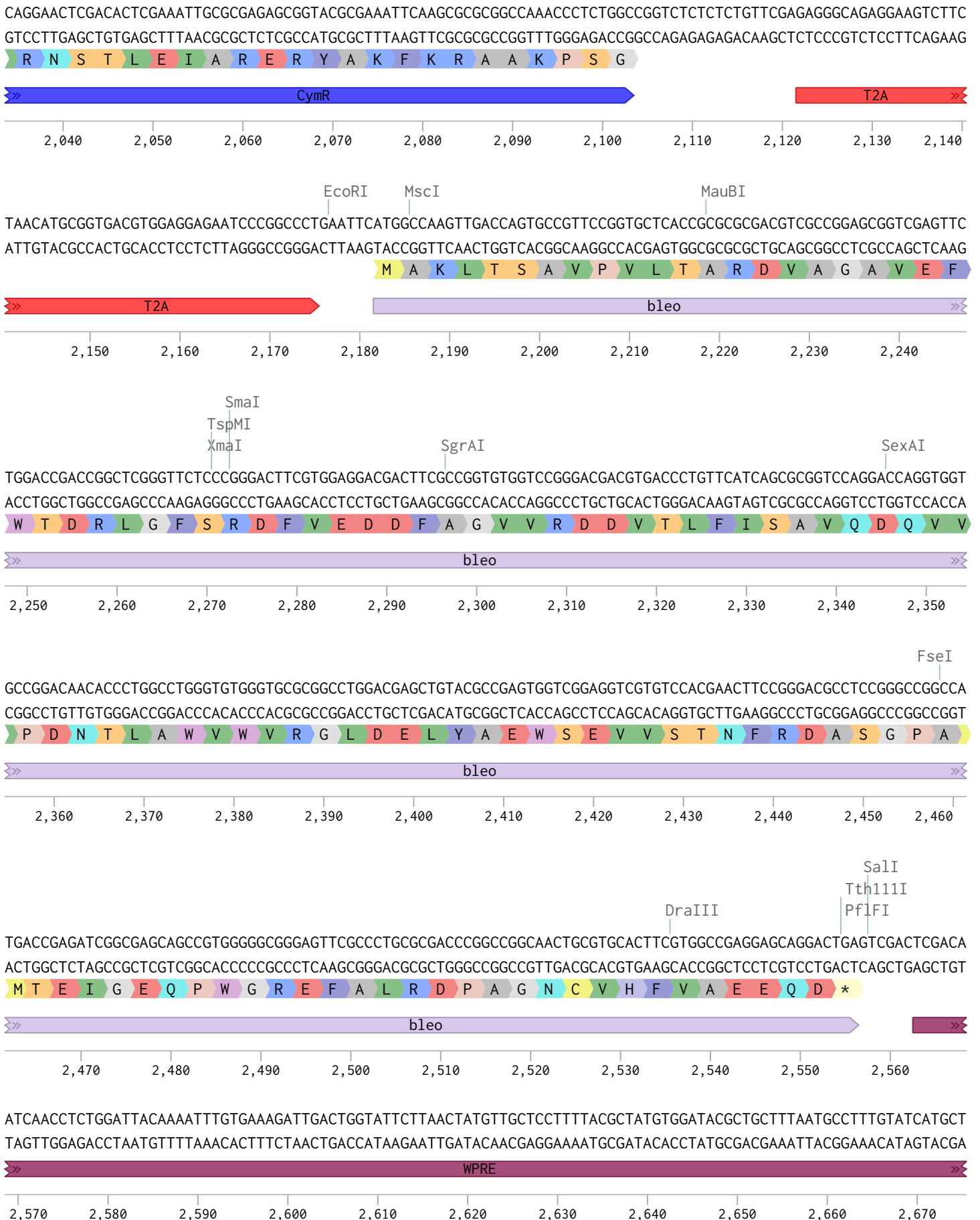




bArr2-HA-TEV219 pCDH-CuO-MCS-EF1 α -CymR-T2A-Bleo...









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SV40 Polyadenylation signal

3,540 3,550 3,560 3,570 3,580 3,590 3,600 3,610 3,620 3,630

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SV40 Polyad...ion signal

SV40 Ori

3,640 3,650 3,660 3,670 3,680 3,690 3,700 3,710 3,720 3,730 3,740

SfiI

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TGATTAATAAATAAATACGTCTCCGGCTCCGGCGGAGCCGAGACTCGATAAGGTCTTCATCACTCTCCGAAAAACCTCCGGATCTGAAAACGTCTCTGGTTT

SV40 Ori

3,750 3,760 3,770 3,780 3,790 3,800 3,810 3,820 3,830 3,840 3,850

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3,860 3,870 3,880 3,890 3,900 3,910 3,920 3,930 3,940 3,950

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3,960 3,970 3,980 3,990 4,000 4,010 4,020 4,030 4,040 4,050 4,060

BspQI

SapI

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4,070 4,080 4,090 4,100 4,110 4,120 4,130 4,140 4,150 4,160 4,170

AflIII

PciI

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pUC ori

4,180 4,190 4,200 4,210 4,220 4,230 4,240 4,250 4,260 4,270 4,280

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pUC ori

4,290 4,300 4,310 4,320 4,330 4,340 4,350 4,360 4,370 4,380

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« pUC ori »

4,390 4,400 4,410 4,420 4,430 4,440 4,450 4,460 4,470 4,480 4,490

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« pUC ori »

4,500 4,510 4,520 4,530 4,540 4,550 4,560 4,570 4,580 4,590 4,600

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« pUC ori »

4,610 4,620 4,630 4,640 4,650 4,660 4,670 4,680 4,690 4,700

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« pUC ori »

4,710 4,720 4,730 4,740 4,750 4,760 4,770 4,780 4,790 4,800 4,810

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« pUC ori »

4,820 4,830 4,840 4,850 4,860 4,870 4,880 4,890 4,900 4,910 4,920

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»

4,930 4,940 4,950 4,960 4,970 4,980 4,990 5,000 5,010 5,020

AhdI

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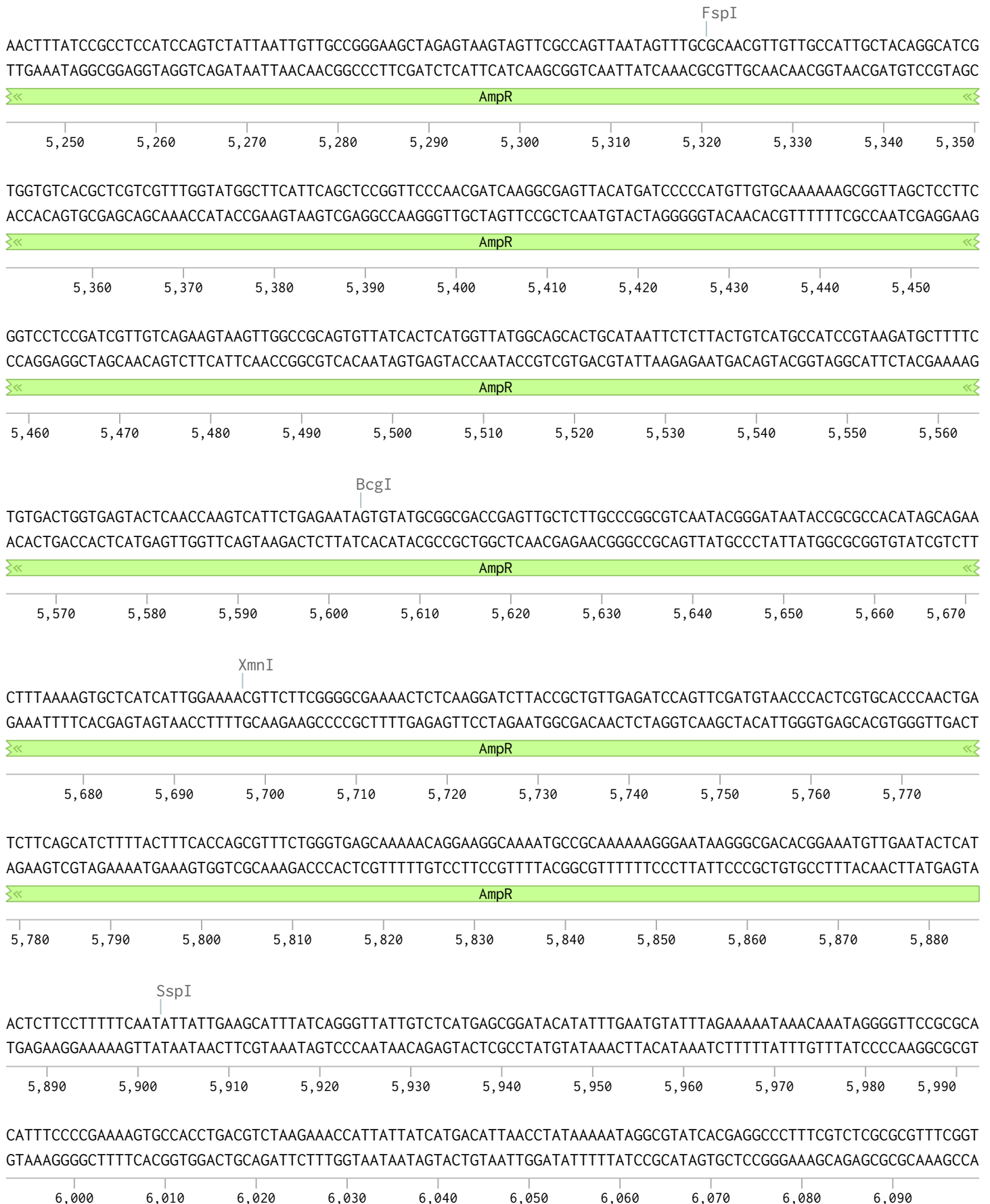
« AmpR »

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« AmpR »

5,140 5,150 5,160 5,170 5,180 5,190 5,200 5,210 5,220 5,230 5,240



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6,100 6,110 6,120 6,130 6,140 6,150 6,160 6,170 6,180 6,190 6,200

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6,210 6,220 6,230 6,240 6,250 6,260 6,270 6,280 6,290 6,300 6,310

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6,320 6,330 6,340 6,350 6,360 6,370 6,380 6,390 6,400 6,410 6,420

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RSV

6,430 6,440 6,450 6,460 6,470 6,480 6,490 6,500 6,510 6,520

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RSV

6,530 6,540 6,550 6,560 6,570 6,580 6,590 6,600 6,610 6,620 6,630

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RSV HIV LTR

6,640 6,650 6,660 6,670 6,680 6,690 6,700 6,710 6,720 6,730 6,740

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HIV LTR

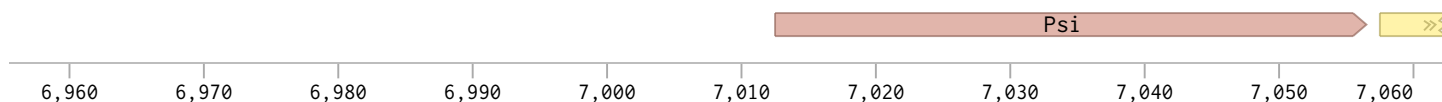
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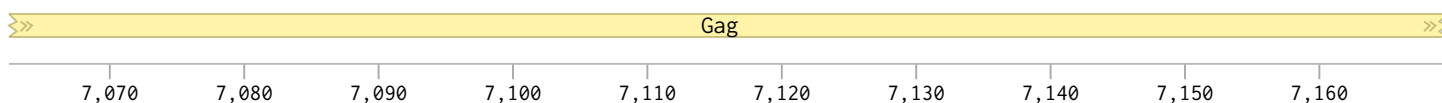
HIV LTR

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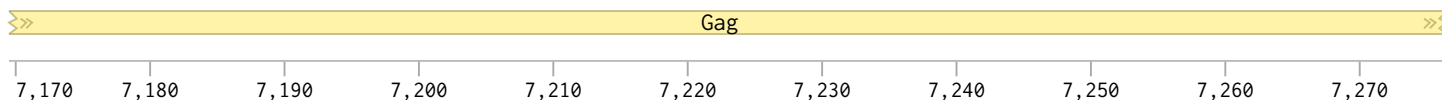
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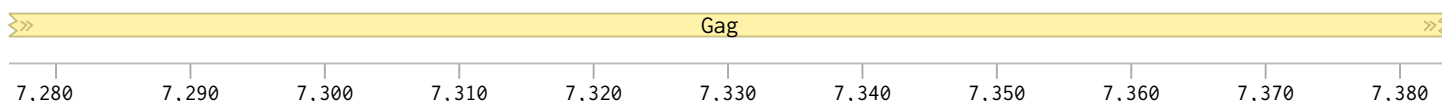
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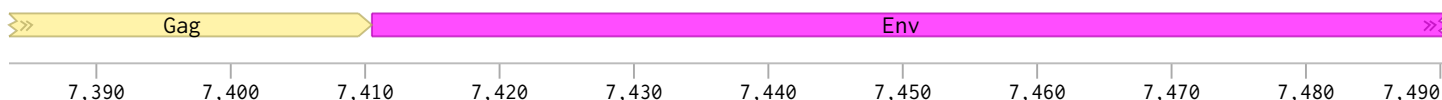
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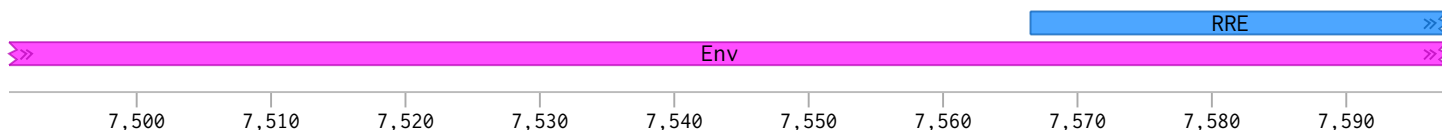
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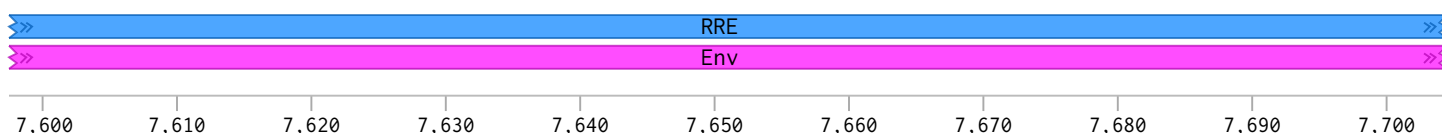
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» CMV + CUO »

8,460 8,470 8,480 8,490 8,500 8,510 8,520 8,530 8,540 8,550 8,560

NdeI

GTTCCCATAGTAACGCCAATAGGGACTTTCCATTGACGTCAATGGGTGGAGTATTTACGGTAAACTGCCACTTGGCAGTACATCAAGTGTATCATATGCCAAGTCC
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» CMV + CUO »

8,570 8,580 8,590 8,600 8,610 8,620 8,630 8,640 8,650 8,660

SnaBI

BsaAI

GCCCCCTATTGACGTCAATGACGGTAAATGGCCGCGCTGGCATTATGCCAGTACATGACCTTACGGGACTTTCCTACTTGGCAGTACATCTACGTATTAGTCATCG
CGGGGATAACTGCAGTTACTGCCATTTACGGGGCGGACCGTAATACGGGTCTGTAAGTGAATGCCCTGAAAGGATGAACCGTCATGTAGATGCATAATCAGTAGC

» CMV + CUO »

8,670 8,680 8,690 8,700 8,710 8,720 8,730 8,740 8,750 8,760 8,770

CspCI

CTATTACCATGGTGATGCGGTTTTGGCAGTACACCAATGGGCGTGGATAGCGGTTTGACTCACGGGGATTTCCAAGTCTCCACCCATTGACGTCAATGGGAGTTTG
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» CMV + CUO »

8,780 8,790 8,800 8,810 8,820 8,830 8,840 8,850 8,860 8,870 8,880

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» CMV + CUO » CMV + CUO »

8,890 8,900 8,910 8,920 8,930 8,940 8,950 8,960 8,970 8,980

AscI

CCTATATAAGCAACCGGTATAATCAAACAGACCAGATTGTCTGTTTGTACCGGTGTTTGTAGTGAACCGGGCGCGCCTCATATCGCCTGGAGACGCCATCCACGCTGT
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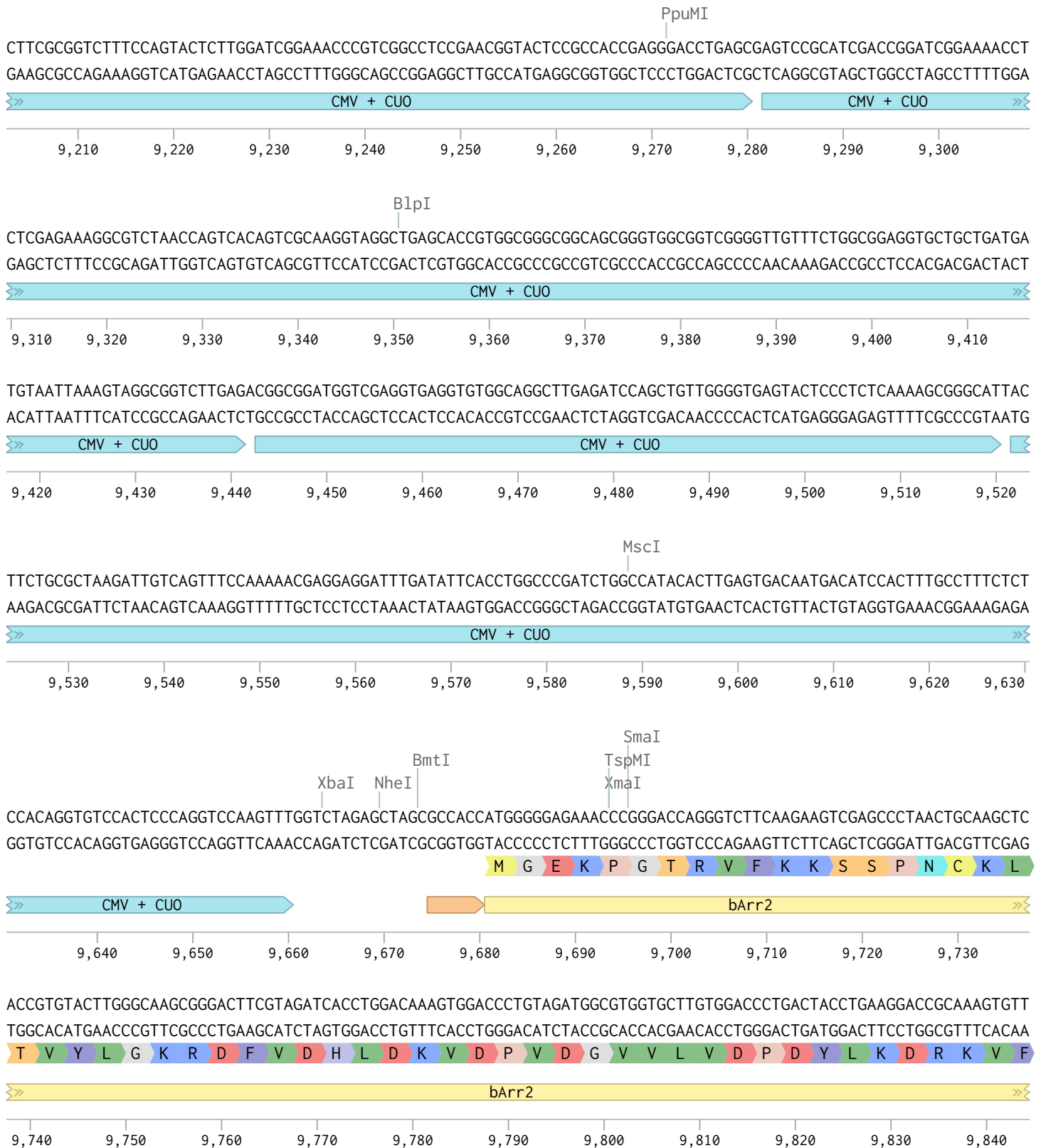
» CMV + CUO »

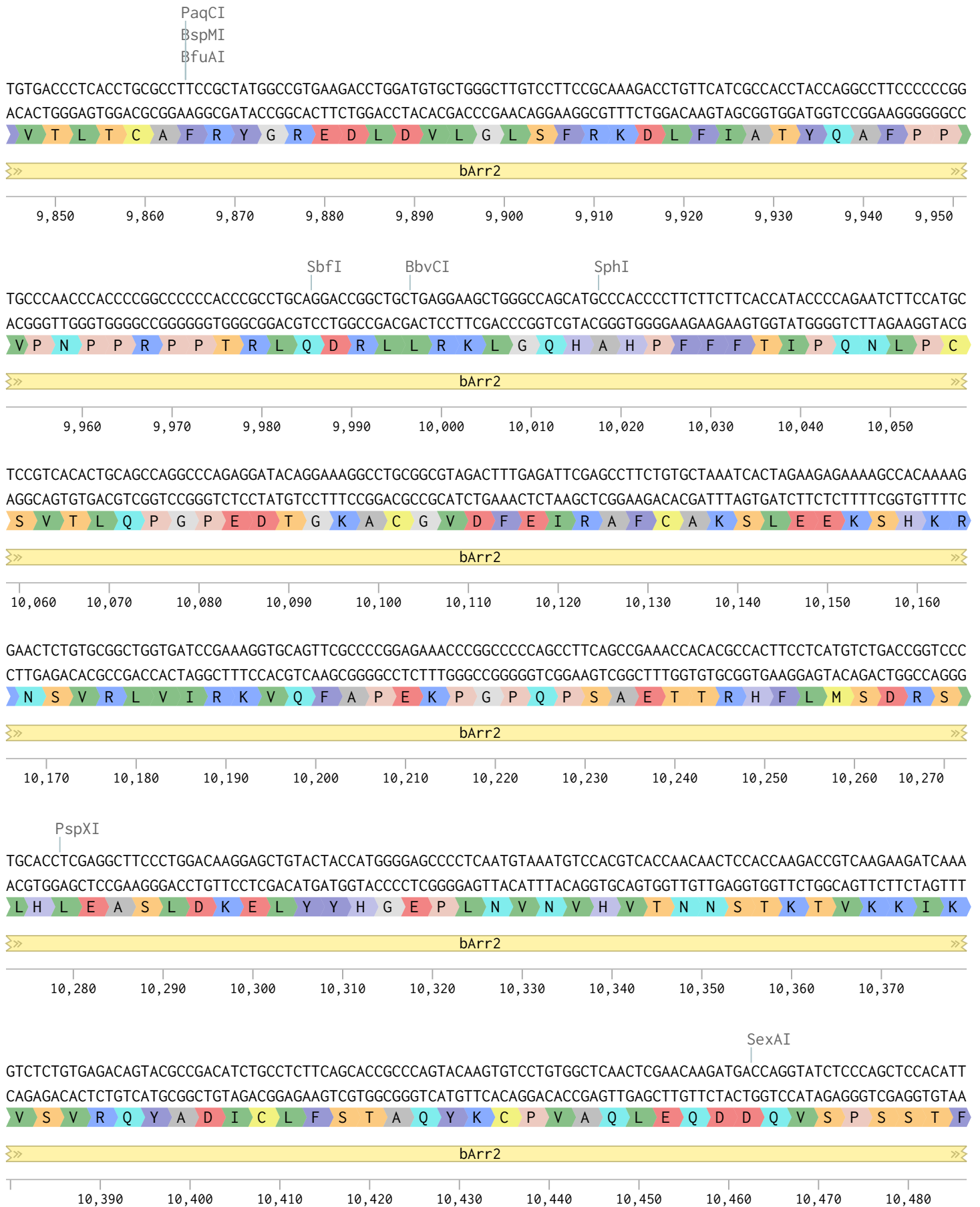
8,990 9,000 9,010 9,020 9,030 9,040 9,050 9,060 9,070 9,080 9,090

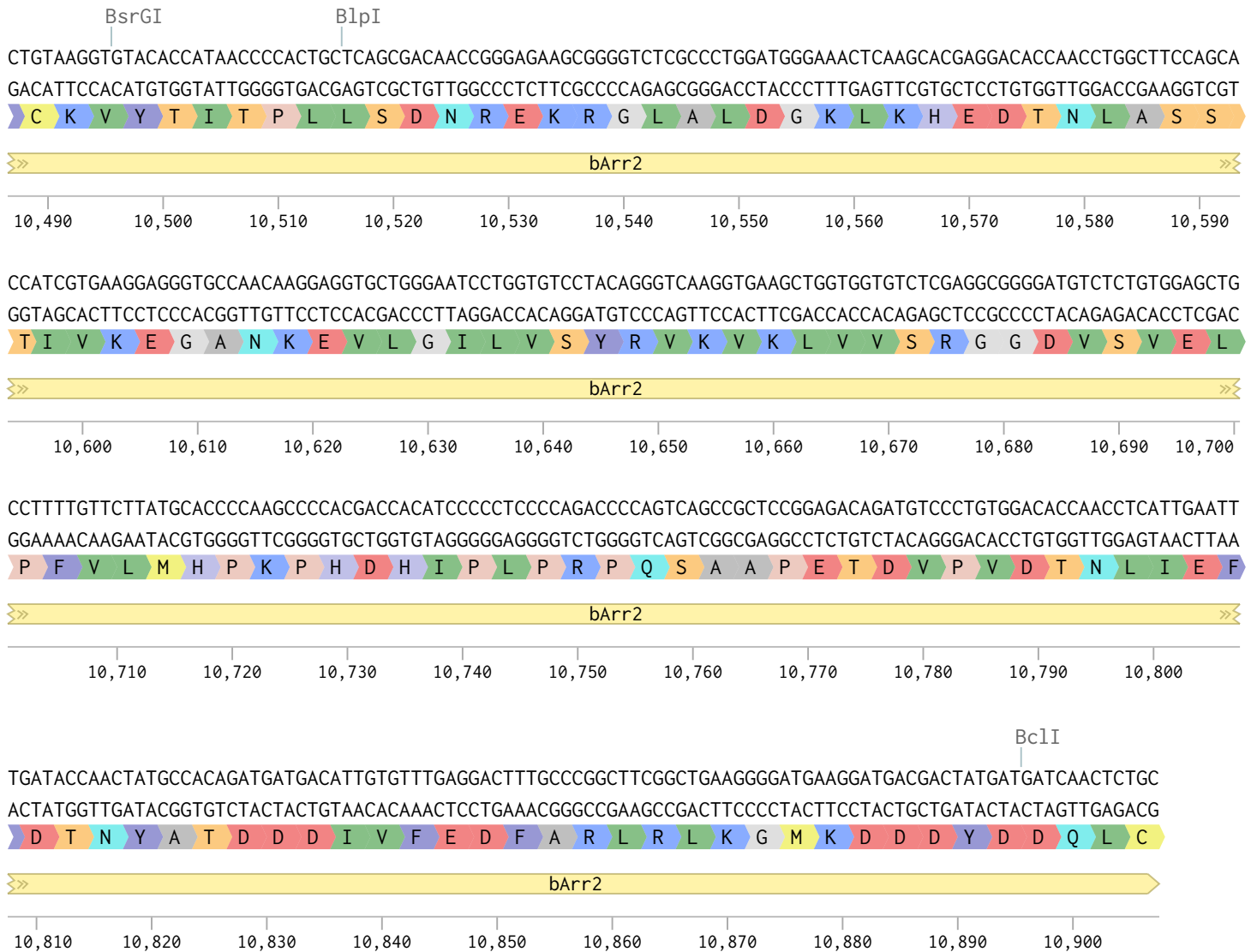
TTTGACCTCCATAGAAGACACCGGGACCGATCCAGCCTCCGCGGTCACTCTTCCGCATCGCTGTCTGCGAGGGCCAGCTGTTGGGCTCGCGGTTGAGGACAAACT
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» CMV + CUO »

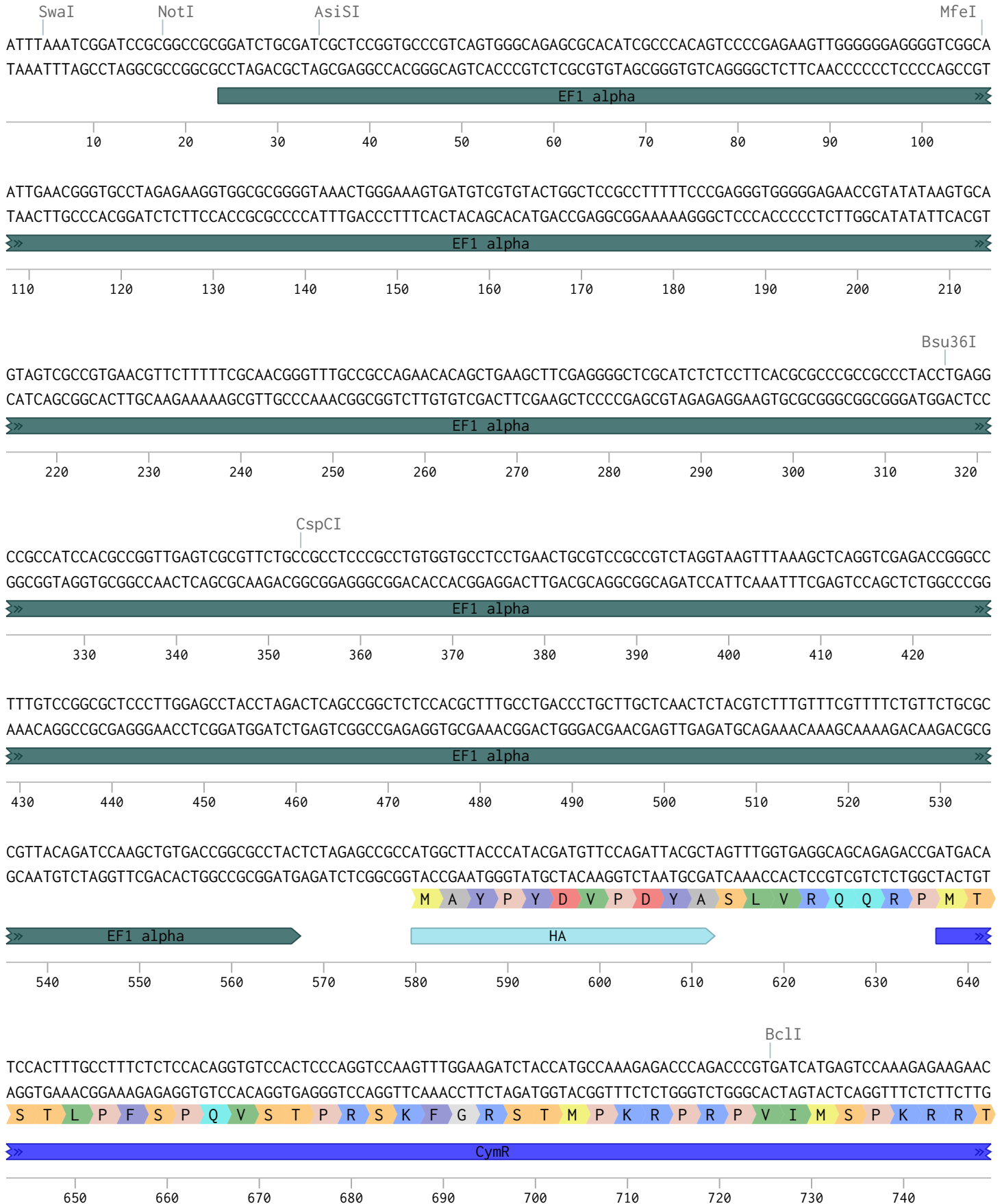
9,100 9,110 9,120 9,130 9,140 9,150 9,160 9,170 9,180 9,190 9,200

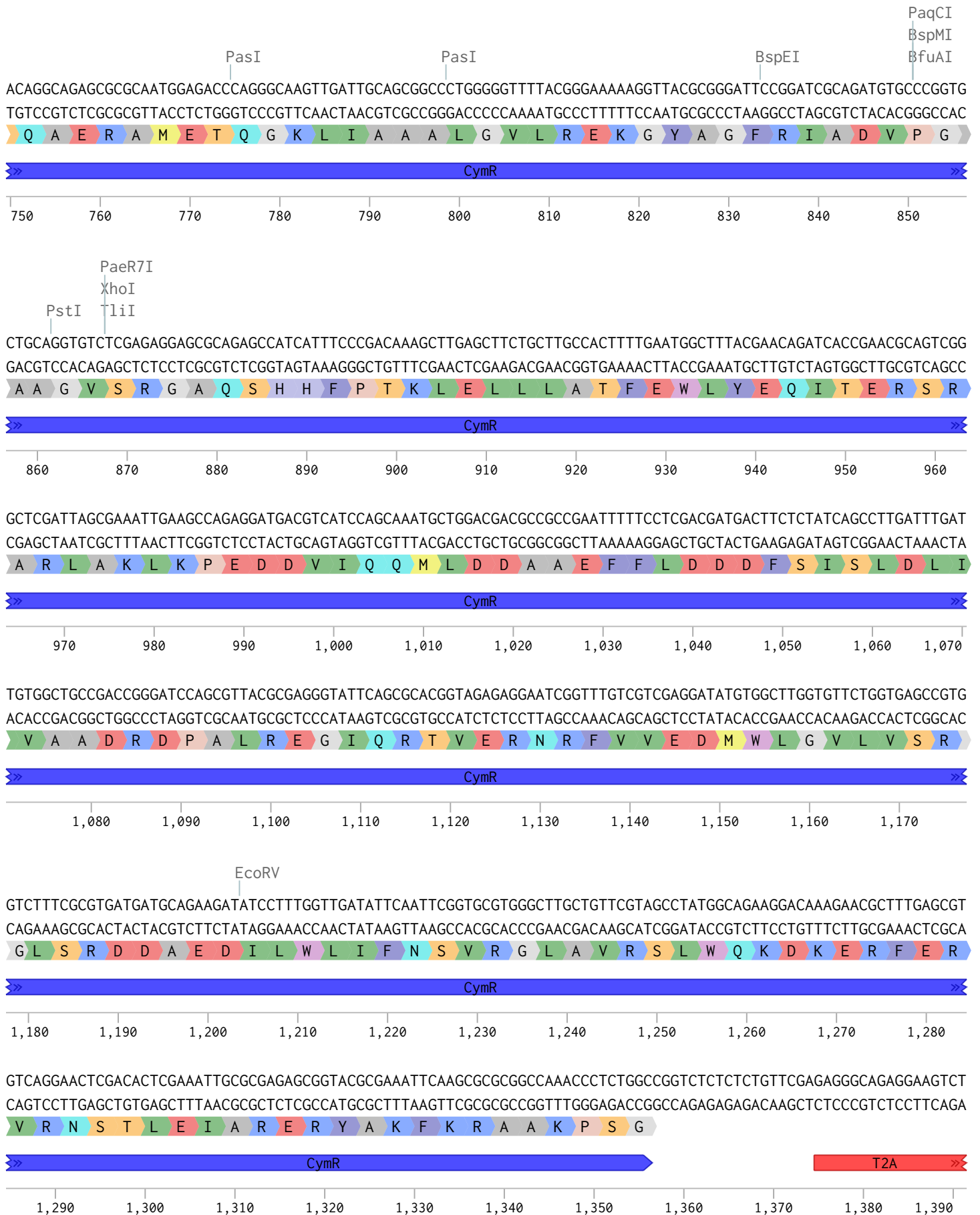


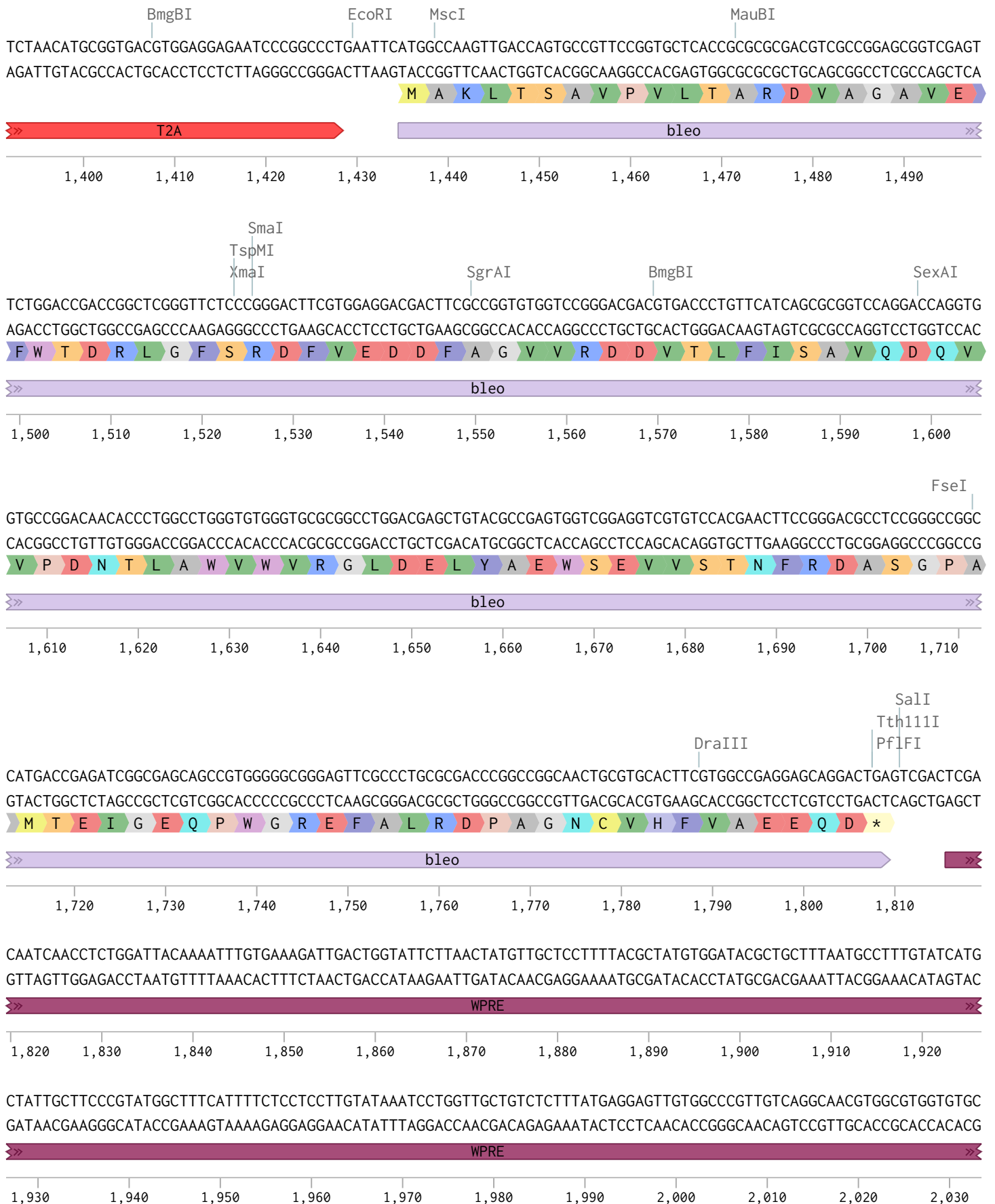




Fyve-HA-TEV219 pCDH-CuO-MCS-EF1 α -CymR-T2A-Bleo3...







ACTGTGTTTGTCTGACGAACCCCCACTGGTTGGGGCATTGCCACCACCTGTCAGCTCCTTCCGGGACTTTTCGCTTTCCCCCTCCCTATTGCCACGGCGGAACATCAT
TGACACAAACGACTGCGTTGGGGGTGACCAACCCCGTAACGGTGGTGGACAGTCGAGGAAAGGCCCTGAAAGCGAAAGGGGAGGGATAACGGTGCCGCTTGAGTA

» WPRE »

2,040 2,050 2,060 2,070 2,080 2,090 2,100 2,110 2,120 2,130 2,140

CGCCGCTGCCTTGCCCGCTGCTGGACAGGGGCTCGGCTGTTGGGCACTGACAATTCGTGGTGTGTCGGGAAATCATCGTCCTTTCTTGCTGCTGCCTGTG
GCGGCGGACGGAACGGCGACGACCTGTCCCGAGCCGACAACCCGTGACTGTAAAGGACCACAACAGCCCTTTAGTAGCAGGAAAGGAACCGACGAGCGGACAC

» WPRE »

2,150 2,160 2,170 2,180 2,190 2,200 2,210 2,220 2,230 2,240

TTGCCACCTGGATTCTGCGCGGACGTCCTTCTGCTACGTCCCTTCGGCCCTCAATCCAGCGGACCTTCCTTCCCGCGGCTGCTGCCGGCTCTGCGGCTCTTCCG
AACGGTGGACCTAAGACGCGCCTGCAGGAAGACGATGCAGGAAGCCGGGAGTTAGGTGCCTGGAAGGAAGGGCGCCGACGACGGCCGAGACGCCGAGAAGGC

» WPRE »

2,250 2,260 2,270 2,280 2,290 2,300 2,310 2,320 2,330 2,340 2,350

CGTCTTCGCCTTCGCCCTCAGACGAGTCGGATCTCCCTTTGGGCCGCTCCCGCCTGGTACCTTTAAGACCAATGACTTACAAGGCAGCTGTAGATCTTAGCCACT
GCAGAAGCGGAAGCGGGAGTCTGCTCAGCCTAGAGGAAACCCGGCGAGGGCGGACCATGGAATTTCTGGTTACTGAATGTTCCGTCGACATCTAGAATCGGTGA

» WPRE »

2,360 2,370 2,380 2,390 2,400 2,410 2,420 2,430 2,440 2,450 2,460

TTTTAAAGAAAAGGGGGGACTGGAAGGCTAATCACTCCCAACGAAGATAAGATCTGCTTTTTGCTTGTACTGGGTCTCTCTGGTTAGACCAGATCTGAGCCTGG
AAAATTTTCTTTCCCCCTGACCTTCCCGATTAAGTGAGGGTGTCTTCTATTCTAGACGAAAAACGAACATGACCCAGAGAGACCAATCTGGTCTAGACTCGGACC

3'LTR »

2,470 2,480 2,490 2,500 2,510 2,520 2,530 2,540 2,550 2,560

GAGCTCTCTGGCTAACTAGGGAACCCACTGCTTAAGCCTCAATAAAGCTTGCCTTGAGTGCTTCAAGTAGTGTGTGCCGCTGTTGTGTGACTCTGGTAACTAGAG
CTCGAGAGACCGATTGATCCCTTGGTGACGAATTCGGAGTTATTTGAAACGGAACCTACGAAGTTCATCACACACGGGCAGACAACACACTGAGACCATTGATCTC

» 3'LTR »

2,570 2,580 2,590 2,600 2,610 2,620 2,630 2,640 2,650 2,660 2,670

ATCCCTCAGACCCTTTTAGTCAGTGTGGAATCTCTAGCAGTAGTAGTTCATGTCATCTTATTATTCAGTATTTATAACTTGCAAAGAAATGAATATCAGAGAGTG
TAGGGAGTCTGGGAAATCAGTCACACCTTTAGAGATCGTCATCATCAAGTACAGTAGAATAATAAGTCATAAATATTGAACGTTTCTTTACTTATAGTCTCTCAC

» 3'LTR »

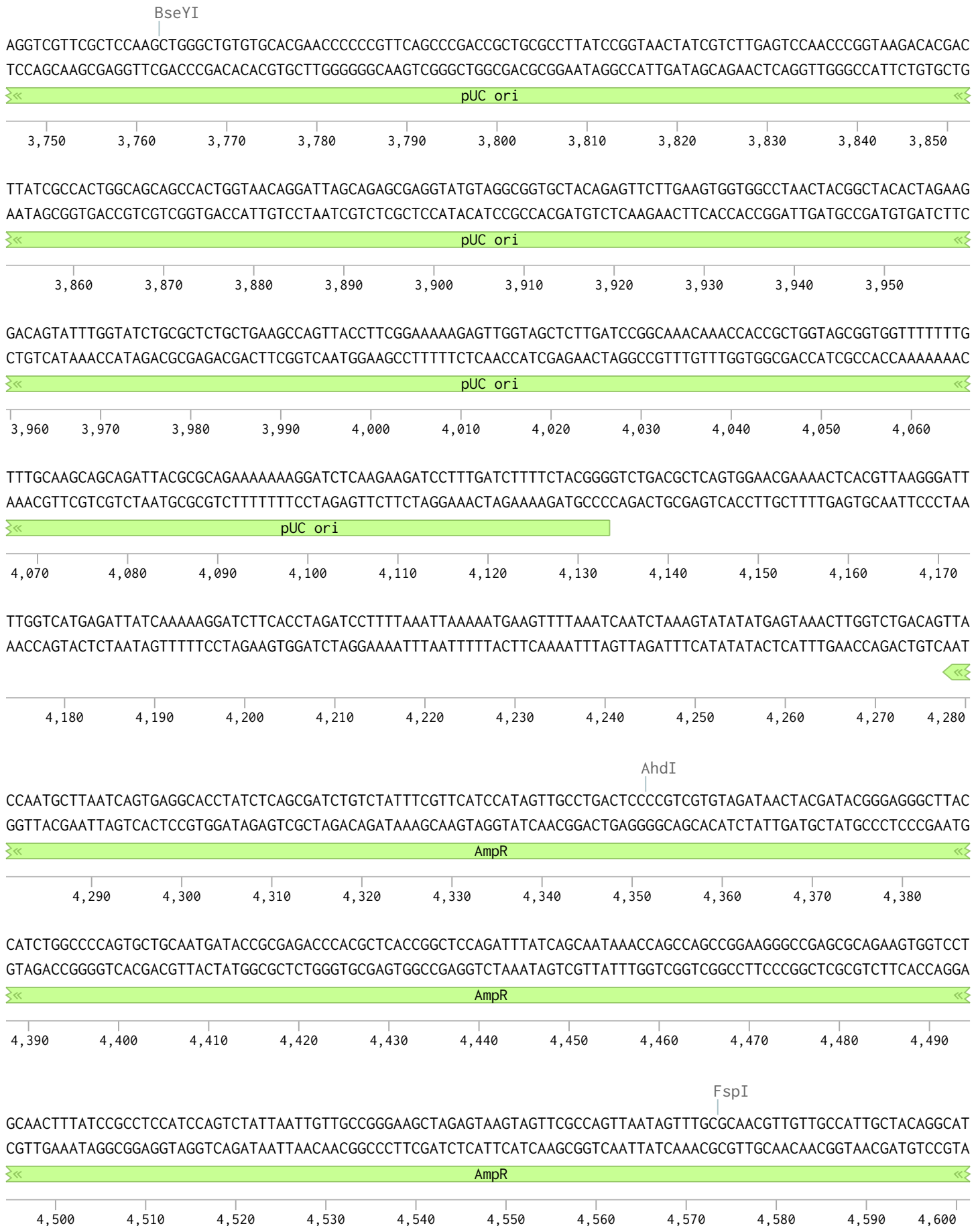
2,680 2,690 2,700 2,710 2,720 2,730 2,740 2,750 2,760 2,770 2,780

AGAGGAACCTGTTTATTGCAGCTTATAATGGTTACAAATAAAGCAATAGCATCACAATTTACAAATAAAGCATTTTTTCTACTGCATTCTAGTTGTGGTTGTCC
TCTCTTGAACAAATAACGTGCAATATTACCAATGTTTATTTCGTTATCGTAGTGTAAAGTGTATTTCGTAATAAAGTACGTAAGATCAACACCAACAGG

SV40 Polyadenylation signal »

2,790 2,800 2,810 2,820 2,830 2,840 2,850 2,860 2,870 2,880





CGTGGTGTCACGCTCGTCGTTTGGTATGGCTTCATTAGCTCCGGTCCCAACGATCAAGGCGAGTTACATGATCCCCATGTTGTGCAAAAAAGCGGTTAGCTCCT
GCACCACAGTGCGAGCAGCAAACCATACCGAAGTAAGTCGAGGCCAAGGGTGTCTAGTTCCGCTCAATGTACTAGGGGGTACAACACGTTTTTTCGCCAATCGAGGA

« AmpR »

4,610 4,620 4,630 4,640 4,650 4,660 4,670 4,680 4,690 4,700

TCGGTCTCCGATCGTTGTCAGAAGTAAGTTGGCCGAGTGTTATCACTCATGGTTATGGCAGCACTGCATAATTCTTTACTGTCATGCCATCCGTAAGATGCTTT
AGCCAGGAGGCTAGCAACAGTCTTCATTCAACCGCGCTCACAATAGTGAGTACCAATACCGTCGTGACGTATTAAGAGAATGACAGTACGGTAGGCATTCTACGAAA

« AmpR »

4,710 4,720 4,730 4,740 4,750 4,760 4,770 4,780 4,790 4,800 4,810

TCTGTGACTGGTGAGTACTCAACCAAGTCATTCTGAGAATAGTGTATGCGGCGACCGAGTTGCTCTTGCCCGCGGTCAATACGGGATAATACCGCGCCACATAGCAG
AGACACTGACCACTCATGAGTTGGTTCAGTAAGACTCTTATCACATACGCCGTGGTCAACGAGAACGGGCCGAGTTATGCCTATTATGGCGCGGTGTATCGTC

« AmpR »

4,820 4,830 4,840 4,850 4,860 4,870 4,880 4,890 4,900 4,910 4,920

XmnI

AACITTTAAAGTGCTCATCATTGAAAAACGTTCTTCGGGGCGAAAACTCTCAAGGATCTTACCGCTGTTGAGATCCAGTTTCATGTAACCCACTCGTGACCCAACT
TTGAAATTTTCACGAGTAGTAACCTTTTGAAGAAGCCCCGCTTTTGAGAGTTCCTAGAATGGCGACAACCTTAGGTCAAGCTACATTGGGTGAGCACGTGGGTTGA

« AmpR »

4,930 4,940 4,950 4,960 4,970 4,980 4,990 5,000 5,010 5,020

GATCTTCAGCATCTTTTACTTTACCAGCGTTTCTGGGTGAGCAAAAACAGGAAGGCAAAATGCCGCAAAAAGGGAATAAGGGCGACACGGAAATGTTGAATACTC
CTAGAAGTCGTAGAAAATGAAAGTGGTCGAAAGACCCACTCGTTTTTGTCTTCCGTTTTACGGCGTTTTTCCCTTATTCGCCGTGTGCCTTTACAACCTTATGAG

« AmpR »

5,030 5,040 5,050 5,060 5,070 5,080 5,090 5,100 5,110 5,120 5,130

SspI

ATACTCTTCCTTTTCAATATTATTGAAGCATTTATCAGGGTTATTGTCTCATGAGCGGATACATATTTGAATGTATTTAGAAAAATAACAAATAGGGGTTCCGCG
TATGAGAAGGAAAAAGTTATAATAACTTCGTAATAGTCCCAATAACAGAGTACTCGCCTATGTATAAACTTACATAAATCTTTTATTTGTTTATCCCAAGGCGC

»

5,140 5,150 5,160 5,170 5,180 5,190 5,200 5,210 5,220 5,230 5,240

CACATTTCCCGAAAAGTGCCACCTGACGTCTAAGAAACCATTTATTCATGACATTAACCTATAAAAATAGGCGTATCACGAGGCCCTTTCGTCTCGCGGTTTCG
GTGTAAAGGGGCTTTTCACGGTGGACTGCAGATTCTTTGGTAATAATAGTACTGTAAATTGGATATTTTATCCGCATAGTGCTCCGGGAAAGCAGAGCGCGCAAGC

5,250 5,260 5,270 5,280 5,290 5,300 5,310 5,320 5,330 5,340 5,350

GTGATGACGGTGAAAACCTCTGACACATGCAGCTCCCGGAGACGGTCACAGCTTGCTGTAAAGCGGATGCCGGGAGCAGACAAGCCCGTCAGGGCGCGTCAGCGGGT
CACTACTGCCACTTTTGAGACTGTGTACGTCGAGGGCCTCTGCCAGTGTGCAACAGACATTGCGCTACGGCCCTCGTCTGTTCCGGCAGTCCCGCGCAGTCGCCCA

5,360 5,370 5,380 5,390 5,400 5,410 5,420 5,430 5,440 5,450

