

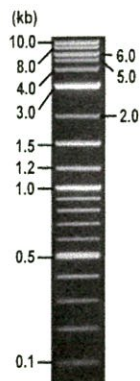


**RIKEN DNA BANK**

clone name : pMK409 (mAID-Nluc BSD)

- Clone ID : RDB \_ 16826
- Lot : 16826 \_ A8Lp
- DNA Concentration : 25 nanogram/microliter
- Volume : 40 microliter
- Form : DNA solution in TE buffer
- Host : DH5 alpha
- Culture : LB medium
- Antibiotics : 100 microgram/ml Ampicillin
- Purification : QIAGEN QIAprep Spin Miniprep kit
- Digestion by restriction enzyme

2-Log DNA Ladder  
(NEB#N3200L),  
250 ng/well



Electrophoresis : 100 nanogram DNA per lane ; 1% agarose gel , 1 x TAE Buffer

Restriction enzyme	Expected size of fragment
<b>NcoI</b>	<u>5.5</u> kbp
<b>NotI</b>	<u>5.5</u> kbp
<b>NcoI+NotI</b>	<u>4.6, 0.9</u> kbp
	kbp
	kbp

● Confirmation of the insertion sequence

Sequence name	Primer name	Sequence name	Primer name
Sequence - A	pUC-M13_-100	Sequence - E	-
Sequence - B	reverse	Sequence - F	-
Sequence - C	PGK_N_rev	Sequence - G	-
Sequence - D	PGKpro_F	Sequence - H	-

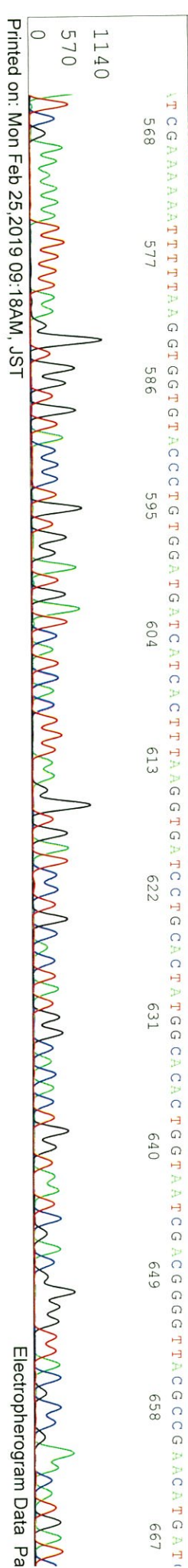
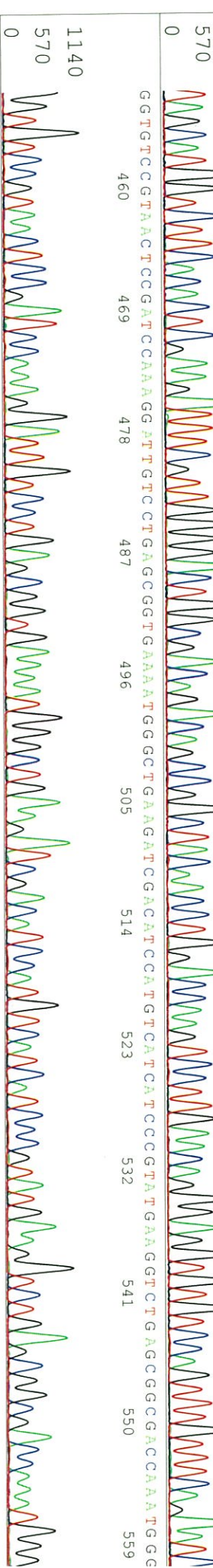
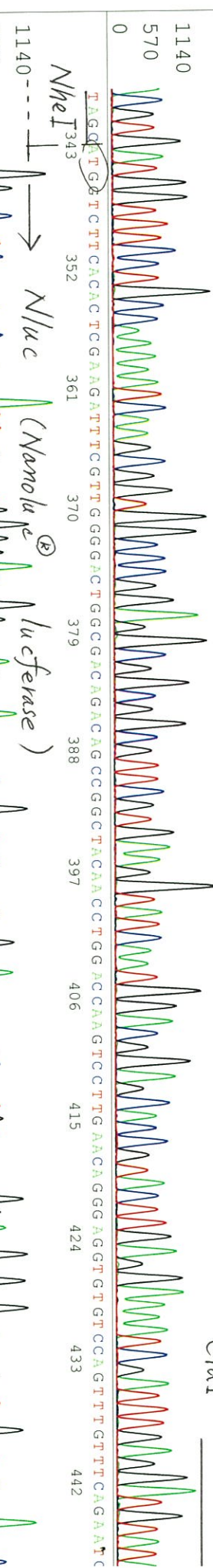
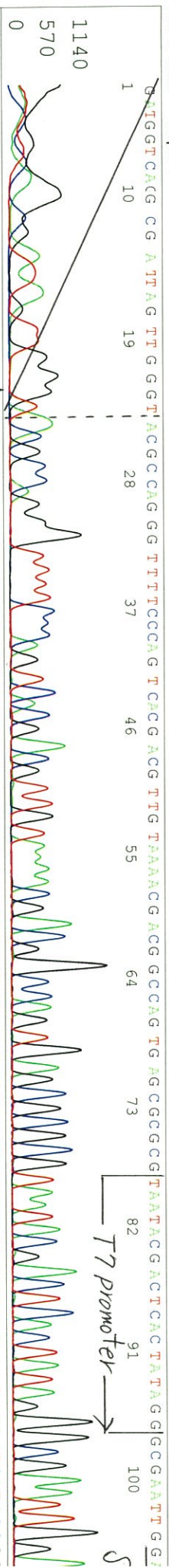
APPROVED BY :



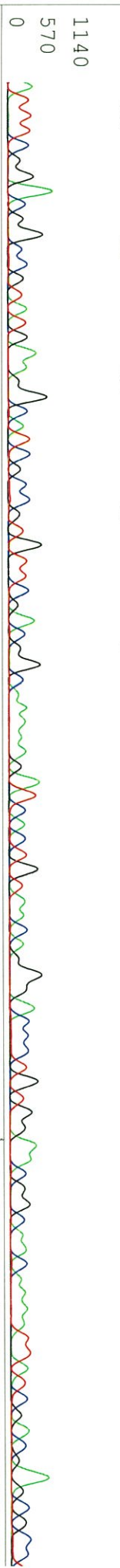
S/N G:363 A:281 T:215 C: primer name **A** : pUC-M13\_-100  
KB.bcp  
KB 1.4.1.8 Cap:15  
5'-ATTACGCCAGCTGGCGAAGG-3'

1 10 19 28 37 46 55 64 73 82 91 100

T7 promoter →



ATTTCCGGACCGCCCGTTATGAAAGCCATCCGCCGTGTTCGACGGCAAAAAGATCACCTGTAACAAGGGACCCTGTGGAAACGGCAACAAATTAATCCGACGAGCC



N/A

CAACCCCGACGGCTCCCTGCTGTTCAGTAACTCAACGGAGTGACCGGCTGGCCGCTGTCGAAACGGCAATTCGGCGTAAAGCGGCTCC

CTAGATAACTGTGATCATTAATCAGCCATACCAACATTTGTAGAGGTTTAACTTGCTTTAAAAAACTCCCAACAATCCCTCCCTTGAAA

AAACATAAAAATGAAATGCAAATTTGTTGTTAACTTTGGTTTAACTTGAAGCCTTAATTAATGGTTTACAAAAATTTAAACAAAA

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T TG TTT G

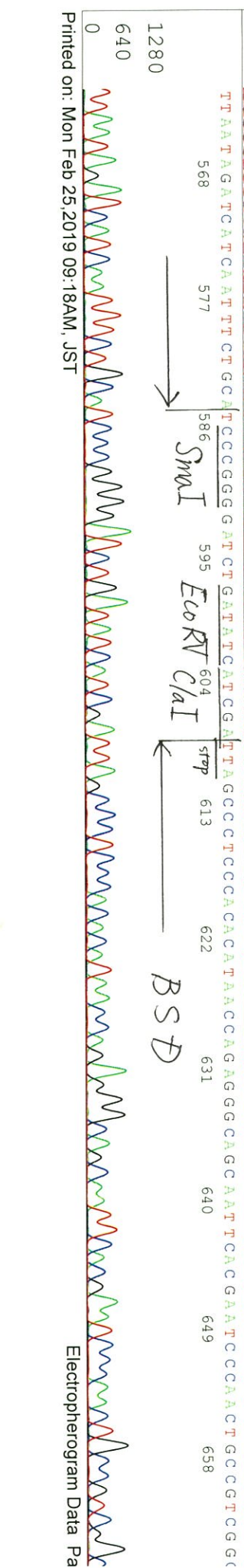
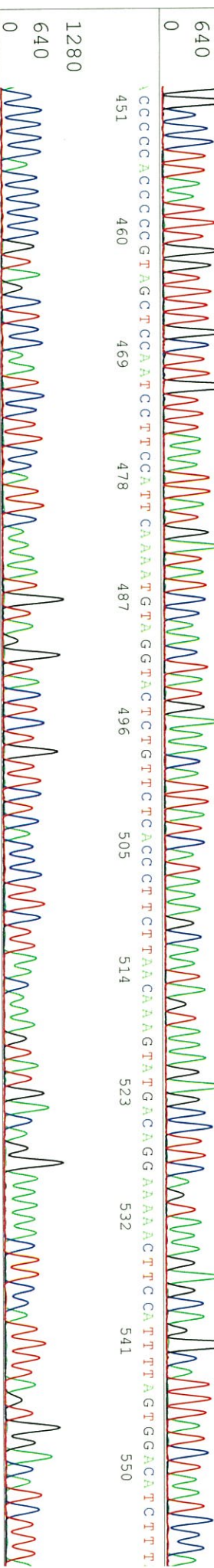
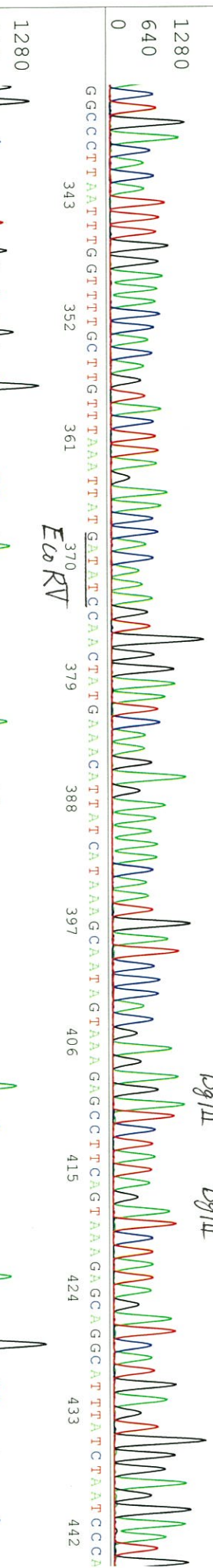
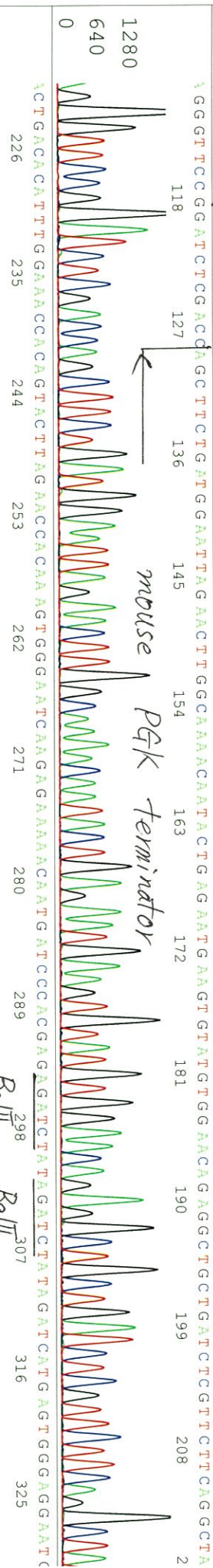
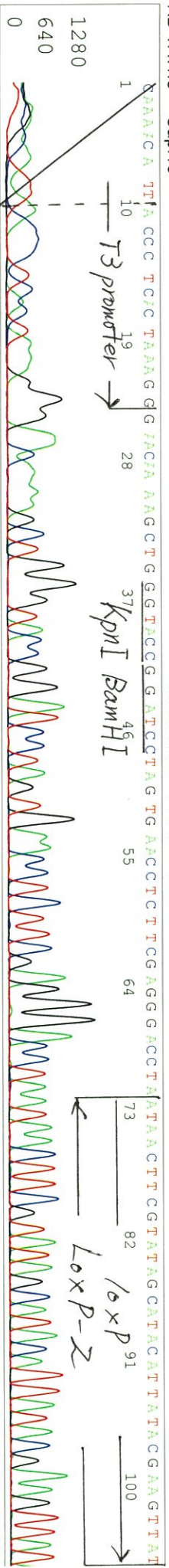
1140 570 0

Printed on: Mon Feb 25, 2019 09:18AM, JST

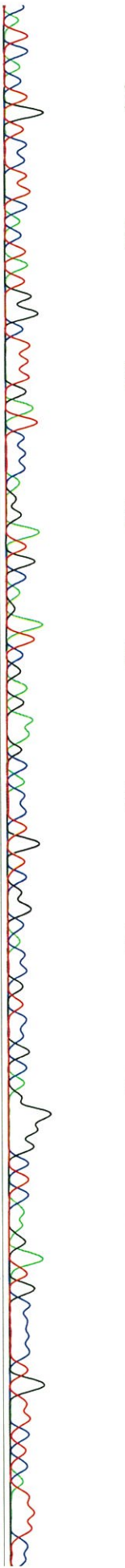
S/N G:182 A:237 T:186 C:194 primer name *B* : reverse  
 KB.bcp 5'-GGAACAGCTATGACCATG-3'  
 KB 1.4.1.8 Cap:18

D06157C4\_A8Lp\_1\_reverse  
 KB\_3500\_POP7\_BDTV3.mob  
 Pts 1557 to 12907 PK1 Loc:1534  
 Version 6.0 HISQV Bases: 949

Feb 22, 2019 02:35F  
 Feb 22, 2019 03:14F  
 Spacing:1.53 Pts/Pai  
 Plate Name: 2019022



C A T C A C T G T C C C T T C A C T A T G G C C T T T G A T C C C C A G G A T G C A G A T C C G A G A A G C A C C T G T C G G C C A C C G T C C G C A G G G C C T C A A G A T G C C C C T G T T C T C A T T T C C



BSD



775 784 793 802 811 820 829 838 847 856 865  
A C A C C A G T G A A G A T G C C G C C G C T A G A G A G A G C T G C G C G A C G C T G T A G T C T T C A G A G A T G G G G G A T G C T G T T G A T T G T

964 973 982 991 1000 1009 1018 1027  
G T T G C T C T T T C A A T G A G G T G G A T T C T T C T T T G A G A C A A A G G C T T T G G C C C A T G G T T G G C T C G A G A A T C T A G A

1036 1045 1054 1063 1072  
A T G G C A A G T C G A A C G C C A G G A G A T G T G G G A G P A G G A G A A C A G C C G C C G G

81  
G A A C G G T G C





S/N G:245 A:262 T:227 C:233

D06157C4\_A8Lp\_1\_PGK\_N\_rev

Feb 22, 2019 02:35P

KB.bcp

KB\_3500\_POP7\_BDTV3.mob

Feb 22, 2019 03:14P

KB 1.4.1.8 Cap:21

Pts 1555 to 13038 Pk1 Loc:1532

Spacing:1.62 Pts/Par

ACCTTAAATAATTTTTCGATCTGGCCCATTTTGTCGCCCGCTCAAGACCTTCATACGGGATGATGACATGGATGTCGATCTTCAGCCCATTTTCACTCCAG

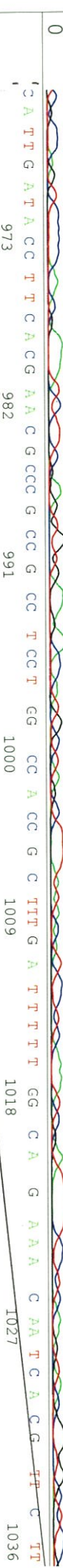


ATCCTTTGGATCGAGTTTACGGACACCCCGAGATTCTGAACAACCTGGACACACCCTCCCTGTTCPAAGGACTTGGTCCAGGTGTAGCCGGCTGT



CGCCAGTCCCCAA CGAATCTTCGAGTGTGAAGCCATGCTAGCTTTATA CATCC TCAAA TCGA TTTTCC TCAA G TAC EG TGC TCC

*Nhe* ← *Nhe I* → *MAlF clal*



JATTGATA CC T T C A C G A A C G C C G C C G C C T C C T C G C C A C C G C T T T G A T T T T G G C A G A A A C A A T C A C G T T



GG TATGGATCTTCACCGGGTGGTCAATCCACACCTTG TGCC TTA VA T G A G



GGCCCTGGG

S/N G:380 A:366 T:276 primer name **fD** : PGKpro\_F

D06157C4\_A8Lp\_1\_Pgkpro\_F

Feb 22, 2019 02:35F

KB: bcp 5'- ATTCTGCACCGCTTCAAAAAG-3'

KB\_3500\_POP7\_BDTV3.mob

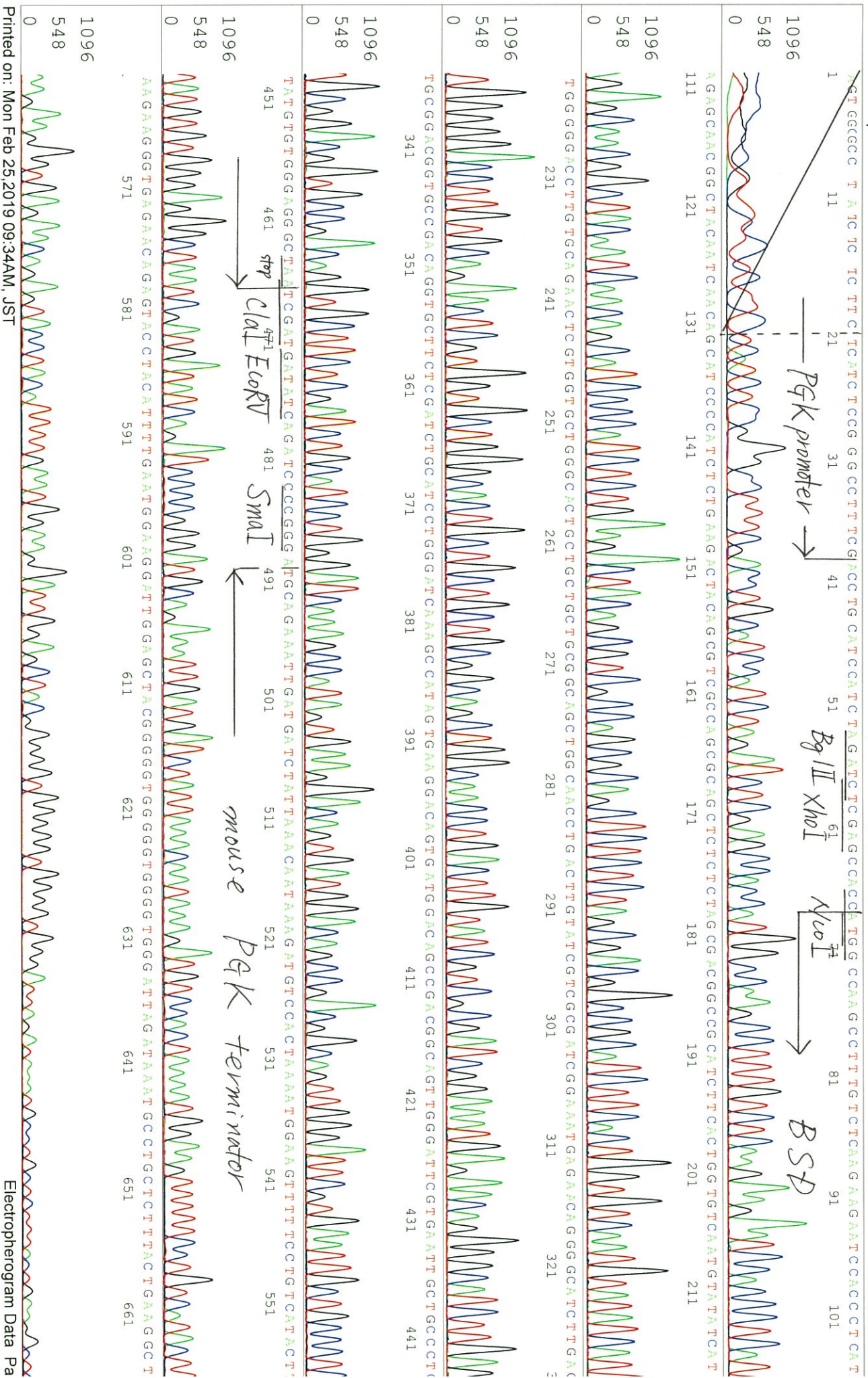
Feb 22, 2019 03:14F

KB 1.4.1.8 Cap:24

Pls 1561 to 10000 PK1 Loc: 1538

Spacing: 1.59 Pts/Pai

Plate Name: 2019022





S/N G:380 A:366 T:276 C:362

KB:bcp

KB 1.4.1.8 Cap:24

D06157C4\_A8Lp\_1\_PGKpro\_F\_H09\_24\_ABI24

D06157C4\_A8Lp\_1\_PGKpro\_F

KB\_3500\_POP7\_BDTV3.mob

Pts 1561 to 10000 PK1 Loc:1538

Version 6.0 HiSQV Bases: 733

Inst Model/Name 3500/3500 Ins

Feb 22, 2019 02:35F

Feb 22, 2019 03:14F

Spacing: 11.59 Pts/Pair

Plate Name: 2019022

