

RIKEN Clone ID : IRAL024P08

Vector : pOTB7

Gene	PRMT5	
Accession No.	BC025979.2	2373 bp
	<i>CDS</i>	1914 bp
		1..2373
		13..1926

● Plasmid DNA purification

Date : 130304

Culture : LB (25 ug/ml Chloramphenicol) 10 ml -> at 37 deg C over night

Date : 130306

Purification : QIAGEN Miniprep kit -> dH₂O 100 ul

● Digestion by restriction enzyme/Concentration calibration

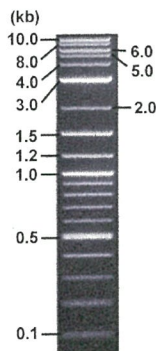
Date : 130307

DNA concentration (O.D.): 270. ng/ul

DNA	0.2	ul
Enzyme (BamHI + BglII)	0.5 + 0.5	ul
Buffer H		1 ul
dH ₂ O	7.8	ul
Total		10 ul

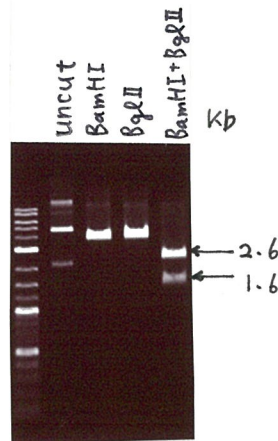
Electrophoresis : 1% agarose gel, 1x TAE Buffer

Marker : 2-Log DNA Ladder (NEB#N3200L)



<Expected size of fragment from BC025979.2 >

2559, 1615 bp.....



● Confirmation of the insertion sequence

Date : 130307

Primer A	pOTB7_F
Primer B	pOTB7_R
Primer C	Poly(A) region primer #0
List of Sequencing Primers	http://dna.brc.riken.jp/en/GNPclone3en.html

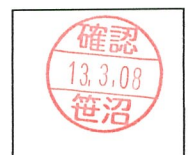
● Adjustment of DNA concentration

Shipping amount : 40 ul

Concentration at the time of preparation of plasmid DNA : 25 ng/ul

Date : 130308

DNA (<u>270</u> ng/ul)	96.0 ul
10x TE	94.1 ul
dH ₂ O	750.7 ul
Total	940.8 ul



APPROVED BY :

<http://dna.brc.riken.jp/index.html>

S/N G:50 A:30 T:35 C:38

KB bcp

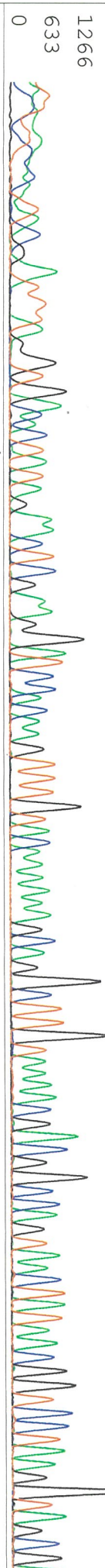
KB 1.4.1.8 Cap:8

Primer A: pOTB7-F

5' AACGGGGCTACAATTATACATAACC 3'

TCA TTCAAATA CCG A T T TA G G T G F C A C T A T A G A A C T C G A A G G A T C C A C A A G T T T G T T A C A A A A A A G C A G G C T T G T A A A A C G A C G G C C A G T A A C T A T T A A C G G T C C T A A G G T A C G C

1 11 21 31 41 51 61 **attB1** 71 81 91 101 111



AGGCC TGGG TGGC GAA T T C G G C A C G A G G C C G A G A G A A A A G A T G G C C G C C A T G G C G G T C C G G G G T G C T G G T G G G A G C C G C G T G T C C A G C C G G T T C C A T C C G G C T T C A G A G G A G T T C A T T C A G G A A C C T G C T A A G A A T C C G C

121 131 141 151 161 171 181 191 201 211 221

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

231 241 251 261 271 281 291 301 311 321 331 341

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

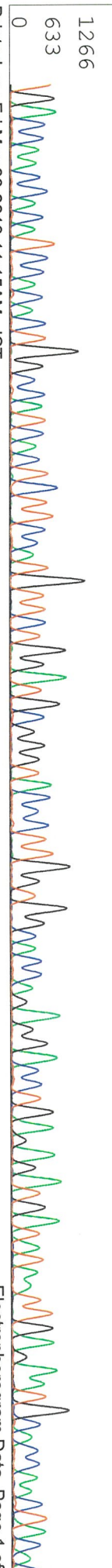
351 361 371 381 391 401 411 421 431 441 451

AGGAAC T C C G A G G C G G C C A T G T T A C A G G A G C T G A A T T T T G G T G C A T A T T T G G G T C C A G C T T C C T G C C T G C C C C T T A A T C A G G A A G P T A A C A C C A A C C T G G C C A G A G T T T

461 471 481 491 501 511 521 531 541 551 561

TGACC AAC C A C A C A T C C C A C A C T G G C C A C T C T T C C A T G T T C T G G A T G C G G G T A C C C T T G G T G G C A C C A G A G G A C C T G A G A G A T G A T A T A A T T G A G A A T G C A C C A A C T A C A C

571 581 591 601 611 621 631 641 651 661 671



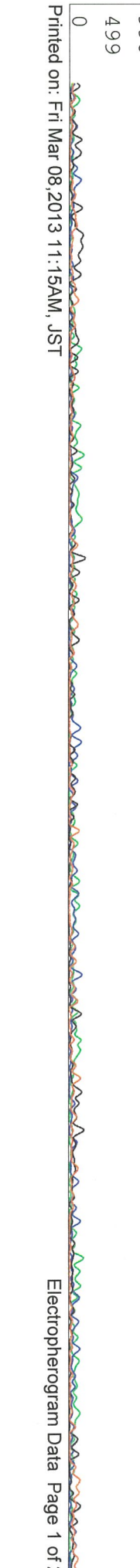
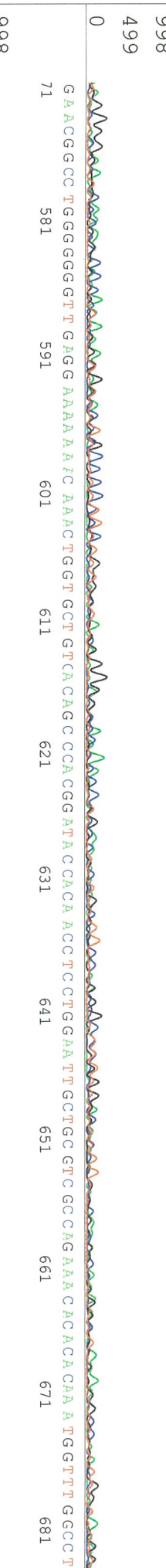
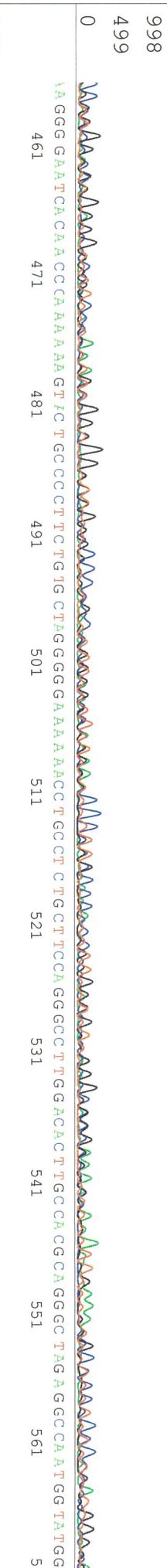
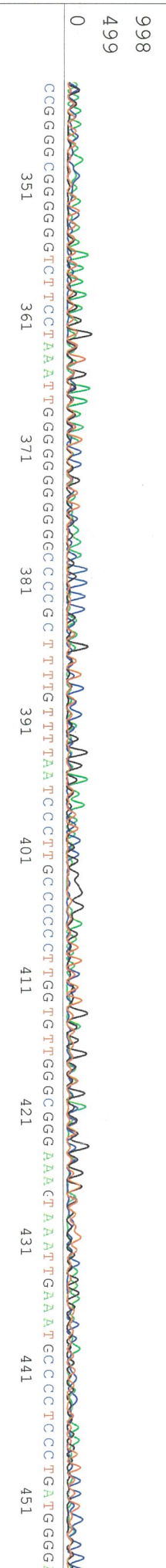
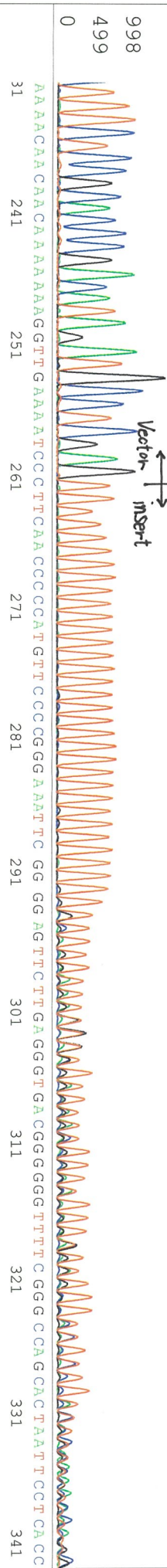
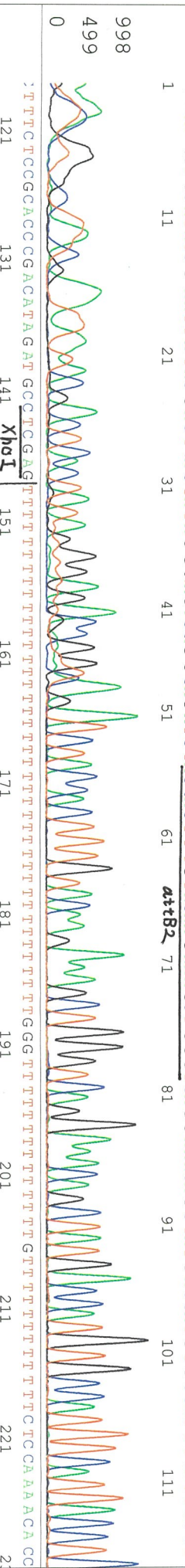
S/N G:33 A:23 T:31 C:24
 KB.bcp
 KB 1.4.1.8 Cap:11

Primer B: pOTB7-R
 5' G T A C T G C A G C C C G A T T C A T T A A T G C 3'

IRAL024P08_A3C4_pOTB7_R_D11_11
 IRAL024P08_A3C4_pOTB7_R
 KB_3500_POP7_BDTV3.mob
 Pts 1261 to 13168 Pk1 Loc:1238
 Version 5.4 HISQV Bases: 416

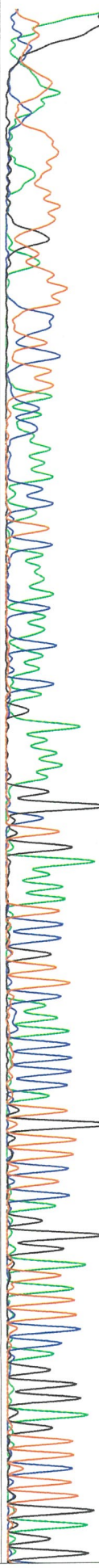
Inst Model/Name 3500/3500 Instrument

Mar 07.2013 09:44PM, JST
 Mar 07.2013 10:11PM, JST
 Spacing: 11.86 Pts/Panel1350
 Plate Name: 20130307_GNP



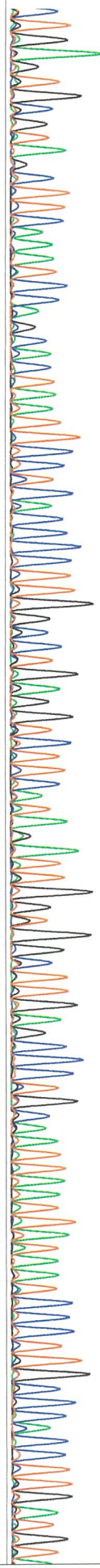
CTTCCATFACCCTT TTTT(GTATTTCC TCTTACC AAAACCA TCAAAACA)ACAACA GAAAAAGGCT GAAAAATCCGTTCAAA CCCCATTGTTCTCA GGGATATCCCA GGGAGTCTTGAGG

1 10 19 28 37 46 55 64 73 82 91 100 109 118

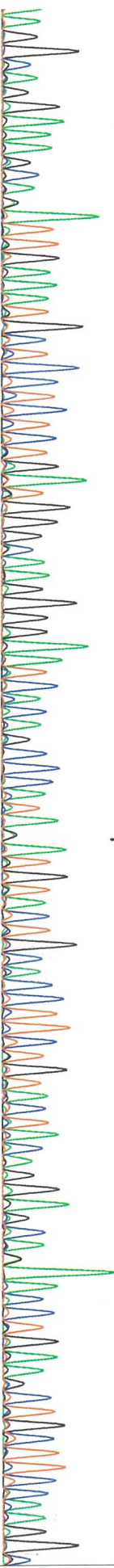


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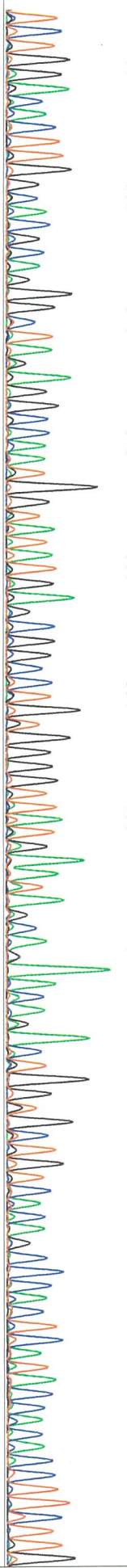
127 136 145 154 163 172 181 190 199 208 217 226



235 244 253 262 271 280 289 298 307 316 325 334 343



352 361 370 379 388 397 406 415 424 433 442 451 460



469 478 487 496 505 514 523 532 541 550 559 568



577 586 595 604 613 622 631 640 649 658 667 676 685

