

RIKEN Clone ID : W01A035J17

Vector : pENTR/D-TOPO

Gene	RGN	
Accession No.	BC073173.1	1560 bp
	<i>CDS</i>	900 bp
		1..1560
		185..1084

● Plasmid DNA purification

Date : 130115

Culture : LB (25 ug/ml Kanamycin) 5 ml -> at 37 deg C over night

Date : 130116

Purification : QIAGEN Miniprep kit -> dH₂O 100 ul

● Digestion by restriction enzyme/Concentration calibration

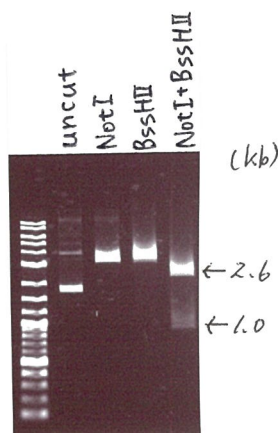
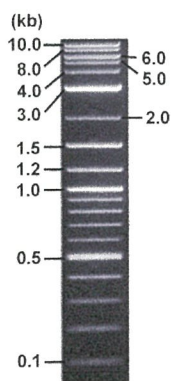
Date : 130116

DNA concentration (O.D.): 92 ng/ul

DNA	<u>0.3</u> ul
Enzyme (NotI + BssHII)	<u>0.5 + 0.5</u> ul
Buffer H	<u>1</u> ul
dH ₂ O	<u>7.7</u> ul
Total	<u>10</u> ul

Electrophoresis : 1% agarose gel, 1x TAE Buffer

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



<Expected size of fragment from BC073173.1 CDS>

2553, 1000bp

● Confirmation of the insertion sequence

Date : 13.1.17

Primer A	M13
Primer B	T7 long
Primer C	-
List of Sequencing Primers	http://dna.brc.riken.jp/en/GNPclone3en.html

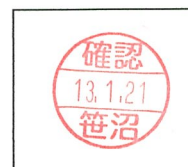
● Adjustment of DNA concentration

Shipping amount : 40 ul

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

Date : 13.1.21

DNA (<u>92</u> ng/ul)	<u>85.0</u> ul
10x TE	<u>31.3</u> ul
dH ₂ O	<u>196.5</u> ul
Total	<u>312.8</u> ul



APPROVED BY :

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

S/N G:38 A:22 T:26 C:19

KB.bcp

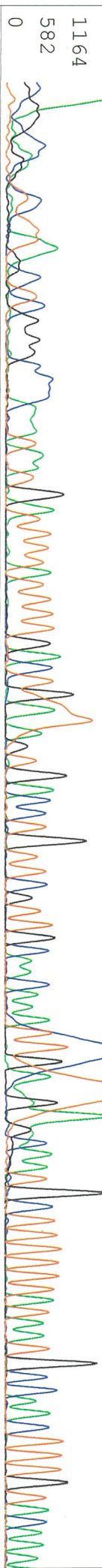
KB 1.4.1.8 Cap:2

Primer A : M13

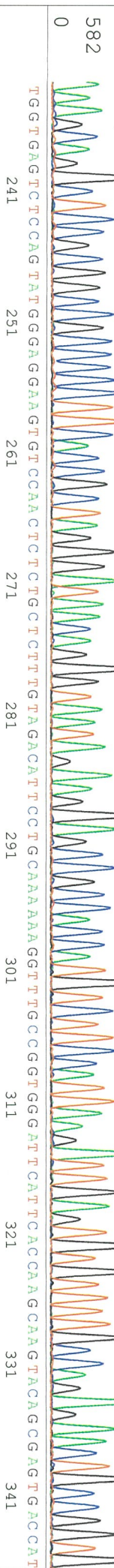
5' GTTTTCCAGTCCAGCAGCTTGTGA 3'

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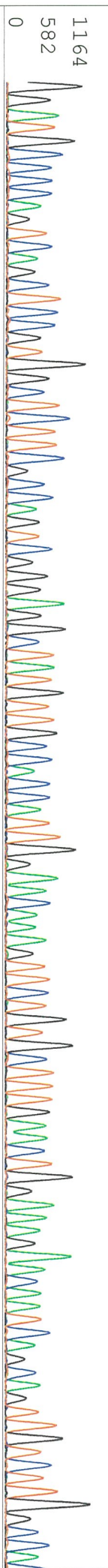
1 11 21 31 41 51 61 71 81 91 101 111



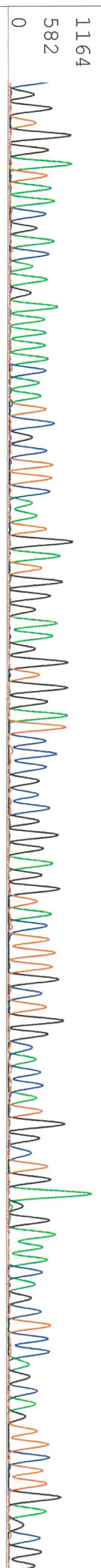
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121 131 141 151 161 171 181 191 201 211 221 231
attL1
NotI
Vector
Start
Insert



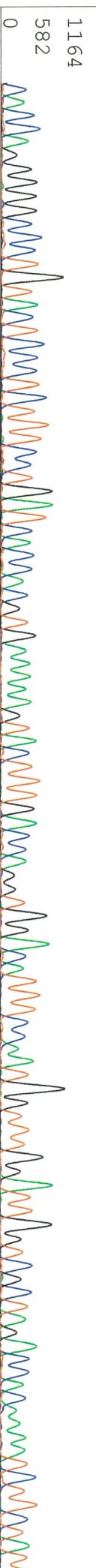
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241 251 261 271 281 291 301 311 321 331 341
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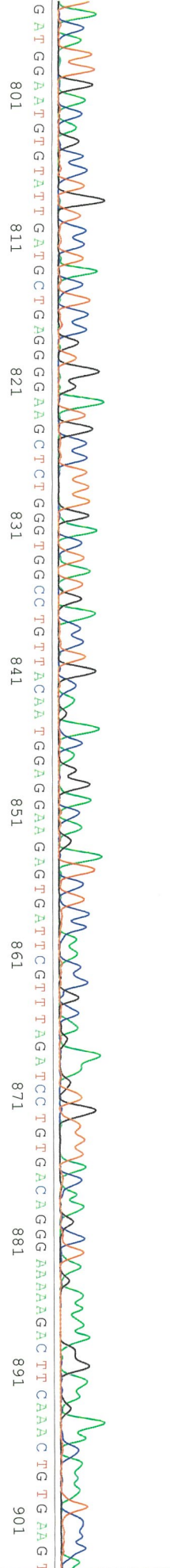
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461 471 481 491 501 511 521 531 541 551 561 571



CACCAAGGGGCGCTGTACTCCCTCTTCCCTGATCACCACGTGA AAAA G TACTTTGACCAAGGTTGACA TTTCCAA TGGTTTGGATTGGTCCCTAGACCAACAATACTTCTAT
581 591 601 611 621 631 641 651 661 671 681



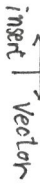
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GATGGAATGTTATGATGCTGAGGGGAAAGCTCTGGGTGGCTTGTTACAA TGGAGGAA GAGTGATTCGTT TAGATCC TGTGACAGGG AAAAAAGACTTCAAACTG TGAAGT
801 811 821 831 841 851 861 871 881 891 901

TGCCCTGTGATTAACA A CTT CATGCTGCTTTTGAGGG AAGAA T TACTCTGA AAA TGTATGTGA CCTGCG CCCGGG ATGGG ATGG ACCCGAGGG TCTTTTGA GGC
911 921 931 941 951 961 971 981 991 1001 1010

AA CCTGFA GC TGG TGGAA TTTT C A GATTA C TGG TTCT GGGGG TC A A GG AA TT GC TCC C TAC TCC T A TG C GGG A TG A G T T CC TGGT G AAC AA TTG
111 1021 1031 1041 1051 1061 1071 1081 1091 1101

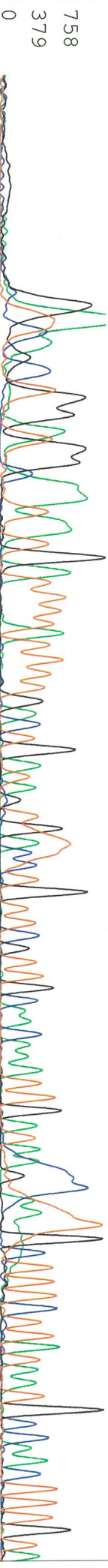


CTCCCTTC TC TA G TA GCCAA A GGGTGTGGG C GCG C G A C C A GCC T T CT T G TA C A A G T GG CCA TT ATT A G A A G
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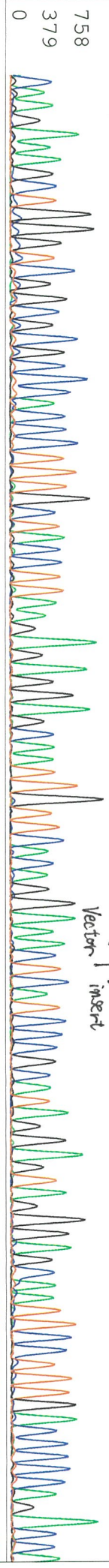
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1181 1191



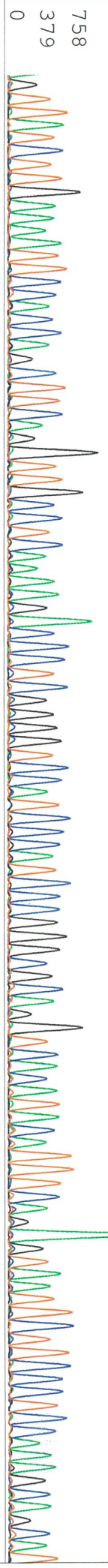
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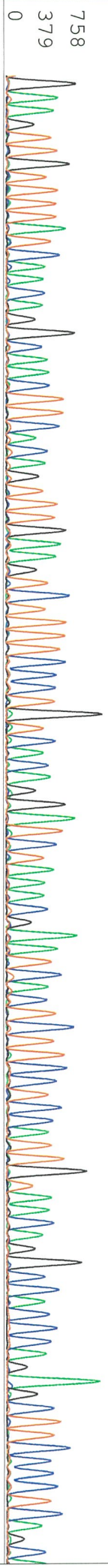
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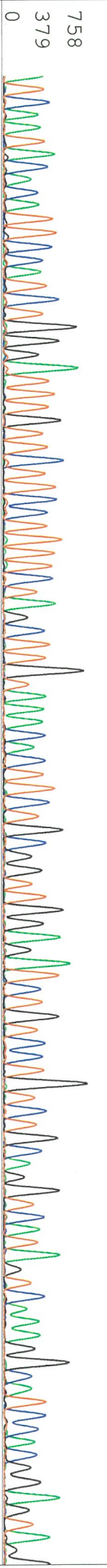
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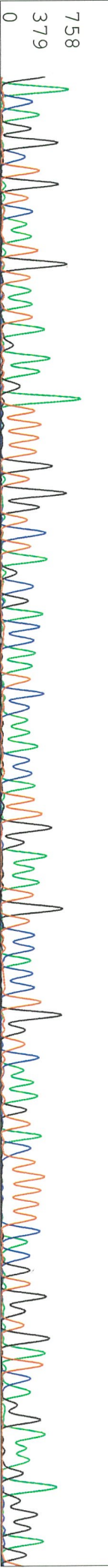
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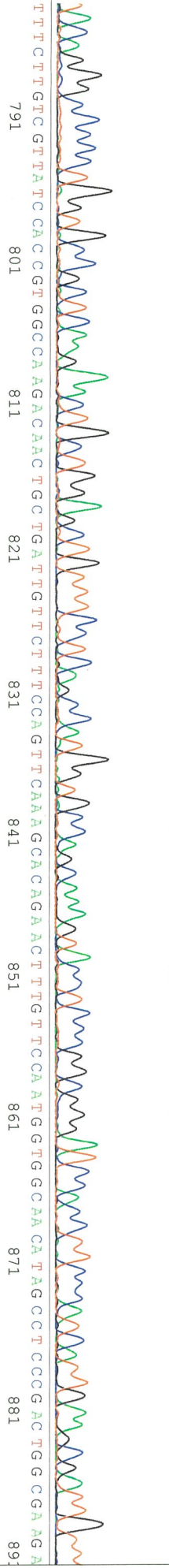
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3 A C A G G C T G T C A A T G T A T A T A G A A G A T T T G T G G T C T A G C G A C C A A T C C A A A C C A T T G G A A T G T C C A C C T G G T C A A A G T A C T T T T T C A C G T G G T G A T C C A G G A A A G A G G G A G



TACAGGGCC CCCC TGTGCCCC TCAA GAAC TGC TGGAGCCTGTGTTTCC TCA GCCCATGTGCCA GC AAA GTACCCTCCCGGGGGGATCCACCCTCCCA TCA TGAAGCCGAT TGGT



TTTCTTGTCCGTTATTC CACCGGTGCCAAGACCAAC TGC TGA TTTGTTCTTTCCAGTTCAAA GCA CAGAACTTTGTTCCCA ATGGTG GCAATAG CCTCCCGAC TGG CGA AGA
 791 801 811 821 831 841 851 861 871 881 891

GCCA(GGGAGC T GAC TGGGCA TCC ATG TCA C TCG TGTAC TTTG CTT GGTGA ATGAATCC CACCGG CA AA CCTTTT TTTT GCA GGA TGTCTACAAA G AGCAG AG
 901 911 921 931 941 951 961 971 981 991

TTGGGACAC T TCCCTCCCA TAC TGGAGAC TCA CCA CCA CC GGG CAG TTTCTCTGGG CAAAA CACCAC TCAATCTTTAA TGG A G ACATGG TTG CC GG C TCC TT
 1001 1011 1021 1031 1041 1051 1061 1071 1081 1091

CTA TTA C C T G TAA T C C TTA G C GG TG A GG G G C GG C C G C G AAG CC T G CC T T T T G TA CA A G T GG CAA T TA
 1101 1111 1121 1131 1141 1151 1161

AA A AAAAA G C C A TTT GT C (TC T)AGA TT CGA
 1171 1181 1191

