

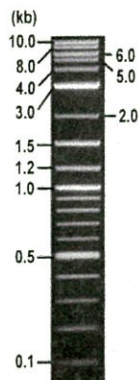


RIKEN DNA BANK

clone name : pGL4-JF1\_886-1349

- Clone ID : RDB \_ 15329
- Lot : 15329 \_ A7Hm
- 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),  
125 ng/well



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

Restriction enzyme	Expected size of fragment
<u>Sall</u>	<u>4.7</u> kbp
<u>PstI</u>	<u>4.7</u> kbp
<u>Sall + PstI</u>	<u>3.6, 1.1</u> kbp
	kbp
	kbp

● Confirmation of the insertion sequence

Sequence name	Primer name	Sequence name	Primer name
Sequence - A	pGL4-4174F	Sequence - E	-
Sequence - B	pGL4-136R	Sequence - F	-
Sequence - C	pAxCALNLF1	Sequence - G	-
Sequence - D	-	Sequence - H	-

APPROVED BY :





CATGAAAGCCGCC TACCGCCCTGGGTGCCCCGGCACCATCGCCCTTTACCGAGCCACATATCGAGGTGGACATTTACCCTAACGCCCGAGTACTTCCGAGATTGAGCCGCTCGGCTGGC

1172  
586

AGAAAGCTATGAAAGCCGC TATGGCCCTGGGTGAGCCCTGAAATACAAACCAATCGGATCGGTGGTGCAGCGAG AATAAGCTTGCAATTGCTTCAATGCCCGTGTTGGGTGCCCTGGT

1172  
586

CATCGGTGTGGCTGTGGCCCTCAGCTAACGACATCTACAA CGAGCGGAGCTGCTGAA CAGCATGGGCATCAGCCAG CCCAACCTCCG TAT

1172  
586

TCGTGAGCAAGAAA GGGGCTGCTAAAAGATCCC TCAAAGCTCAATACAAAAGATCAATCAATTTCT

Luciferase

TGGGAA TAGC AAG AAGGCACTGCAACCTTA CTA GGCCTTT CGG AAAAGCAAAGCTAATG TAACTA CCAA C ATTCGGGTTG

1172  
586

AACCTTTCT  
1108

1172  
586

S/N G:140 A:158 T:120 C:138

primer name  $\beta$  : pGL4-136R

KB.bcp KB\_3500\_POP7\_BDTV3.mob

Dec 12,2017 02:58PM, JST

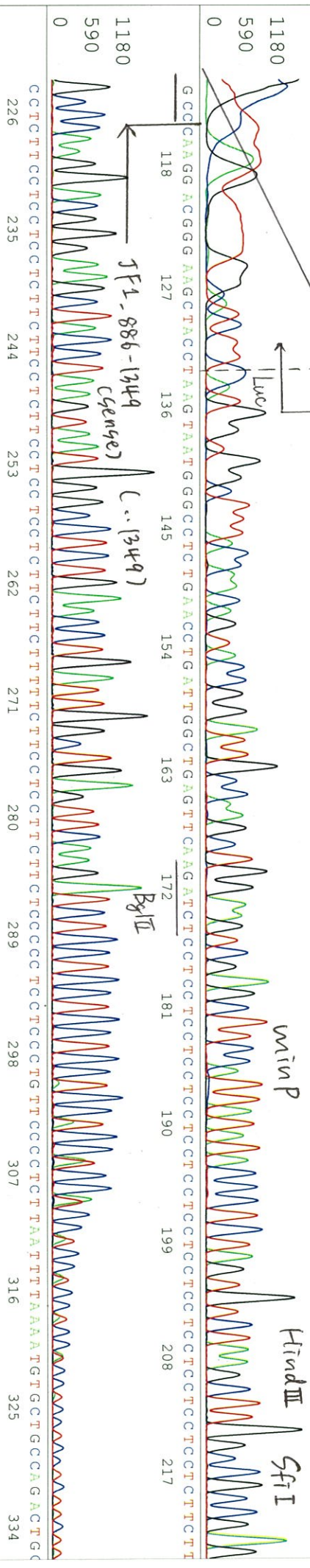
KB 1.4.1.8 Cap:8

5-CTTCGAGTGGGTAGAATGGC-3'

Pis 1502 to 12667 PK1 Loc:1479

Dec 12,2017 03:38PM, JST

GT CTTTAA GC TTT TG GCATC TTTCA TTS GTGG CCTTTC CCAACAG TAACCG GAT TGCCAA GCTGG AAGTCC GGC TTTCCA TATATA C CCTC TAA GTGTCT AAGCTT G G C C C C G A G



1180 : AACCTCAAGCCTCATTTTAAGATTGAACCTCCTACACAAATTTCTTTCAGGATCATGACACA AAAACAACAAA CACAGATAATTGAAACGGTTGGGCATGTCCTGTTCTATT  
590 343 352 361 370 379 388 397 406 415 424 433 442

1180 CCAAAAGTTTCCATCCTGAGTAACCTCCCGTCCCTTTTGCAACC TCAACATAGCTAACTTATGCCAAGGTAATAATAAGTCCCAATTATAACAGTCTGTAACCTATCCAACCTTA  
590 451 460 469 478 487 496 505 514 523 532 541 550 559

3 AAATTGGCCTTCAAGGGGCGGCCAGTTAGGCCAGAGAAATGTTCTGGCCACC TGCACTTGCCACTGGGGACA GGCCTATTTTGGCTA GTTGT TTTGTTTCTTGA TGG  
568 577 586 595 604 613 622 631 640 649 658 667

JF1-886-1349 (Csense) ← SfiI ← pause site

AGAGCGTTATGTTAGTACTTATCGATTTCACCAAAAAAACCAACCAACAGATGTAATGAAAAATAAAGATAATTATTGCGGCCCTGTCCAATACTCCCGTACCCTAATAAT

676 685 694 703 712 721 730 739 748 757 766 775

1180 pause site ←

Clal

poly(A) signal

TACTTACTTATCTCTTGAGAGACGTACTAGTAAACCCTGATAAATGCTTCAATAA TATTGAAAAAGACGATATGAGCATCCACACATTTTCCGTGTCGCCACTC

784 793 802 811 820 829 838 847 856 865 874

SpeI

→ Amp R

ATTCCTTCTTTGCCGCCATTTTCTTGCCCTGTTTGGCACACCCCGAAACGGCTGGTGAAGATAAAGATGCTGAA GATCAA C TGGG TGC AA G

883 892 901 910 919 928 937 946 955 964

1180  
590

AGTGGGGCTAATAATCGAATCGGGATCTCAA TAG CEGGCAA GATCC TTTGAGTCTTTCCCGCCCGAA GACGAATTGAGCATCCACACATTTTCCGTGTCGCCACTC

973 982 991 1000 1009 1018 1027 1036 1045

← Amp R

TGAAAGCAC TTTCAAGTAACCTGGCCATGTGA CCGCG ATGTGGTCCCGTAA TAGGACGACGACCG

154 1063 1072 1081 1090 1099 1108

1180  
590

GCTAAGGAAAGC

1117

1180  
590



