

RIKEN Clone ID : W01A006N04

Vector : pENTR/D-TOPO

Gene	SP2		
Accession No.	BC016680.2	2876 bp	1..2876
	<i>CDS</i>	1821 bp	52..1872

● Plasmid DNA purification

Date : 130225

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

Date : 130226

Purification : QIAGEN Miniprep kit -> dH₂O 100 ul

● Digestion by restriction enzyme/Concentration calibration

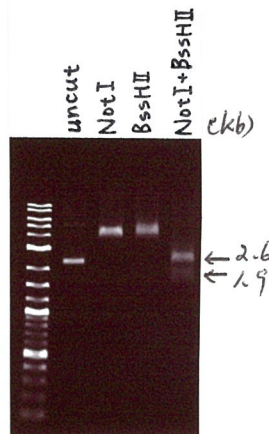
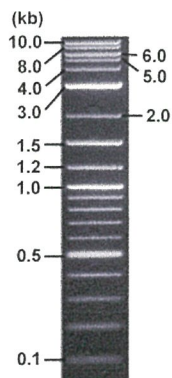
Date : 130301

DNA concentration (O.D.): 189 ng/ul

DNA	0.1	ul
Enzyme (NotI + BssHII)	0.5 + 0.5	ul
Buffer H	1	ul
dH ₂ O	7.9	ul
Total	10	ul

Erectrophoresis : 1% agarose gel, 1x TAE Buffer

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



<Expected size of fragment from BC016680.2 CDS>

2553, 1921 bp

● Confirmation of the insertion sequence

Date : 130227

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

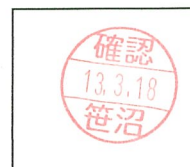
● Adjustment of DNA concentration

Shipping amount : 40 ul

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

Date : 130315

DNA (<u>189</u> ng/ul)	87	ul
10x TE	65.8	ul
dH ₂ O	504.9	ul
Total	657.7	ul



APPROVED BY :

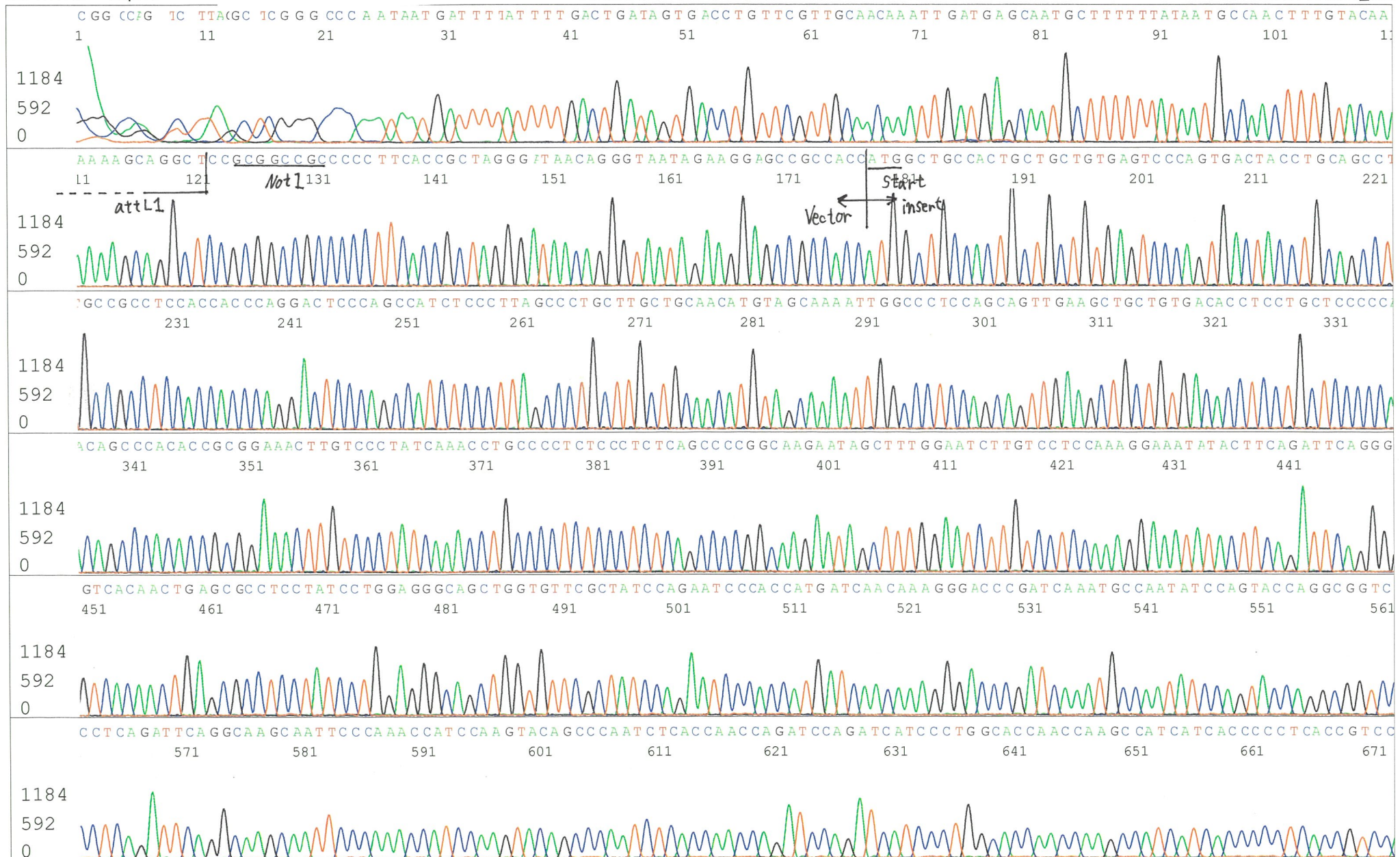
<http://dna.brc.riken.jp/index.html>

S/N G:216 A:163 T:148 C:18 Primer A : M13

KB.bcp

KB 1.4.1.8 Cap:13

5' GTTTTCCCAGTCACGACGTTGTA 3'



S/N G:18 A:9 T:11 C:11
KB.bcp
KB 1.4.1.8 Cap:14

Primer B : T7 long

5' CGCCAAGCTCTAATACGACTCACTATAGGG 3'

