

**RIKEN Clone ID : W01A021O15**

Vector : pENTR-TOPO

Gene	BRRN1	
Accession No.	BC024211.2	2802 bp
	<i>CDS</i>	2226 bp
		1..2802
		44..2269

● Plasmid DNA purification

Date : 141028

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

Date : 141029

Purification : QIAGEN Miniprep kit -> dH<sub>2</sub>O 100 ul

● Digestion by restriction enzyme/Concentration calibration

Date : 141029

DNA concentration (O.D.): 67 ng/ul

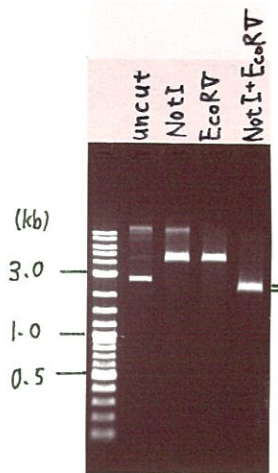
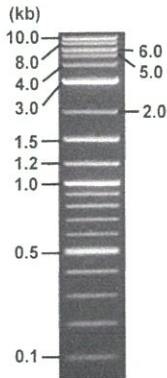
<Size of fragment expected from BC024211.2 >

DNA	0.4	ul
Enzyme (NotI + EcoRV)	0.5 + 0.5	ul
Buffer H	1	ul
dH <sub>2</sub> O	7.6	ul
Total	10	ul

NotI	4998 bp
EcoRV	4998 bp
NotI + EcoRV	2553bp (Vector)      2445 bp

Electrophoresis : 1% agarose gel, 1x TAE Buffer

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



● Adjust plasmid DNA solution to 25 ng/ul

Date : 141030

● Confirmation of the insertion sequence

Date : 141030

DNA ( <u>67</u> ng/ul)	80.0	ul
10x TE	21.4	ul
dH <sub>2</sub> O	113.0	ul
Total	214.4	ul

Primer A	M13
Primer B	T7 long
Primer C	-
List of Sequencing Primers	<a href="http://dna.brc.riken.jp/en/GNPclone3en.html">http://dna.brc.riken.jp/en/GNPclone3en.html</a>

● Shipping

Conc. : 25 ng/ul, Volume : 40 ul

Conc. : ng/ul, Volume : ul

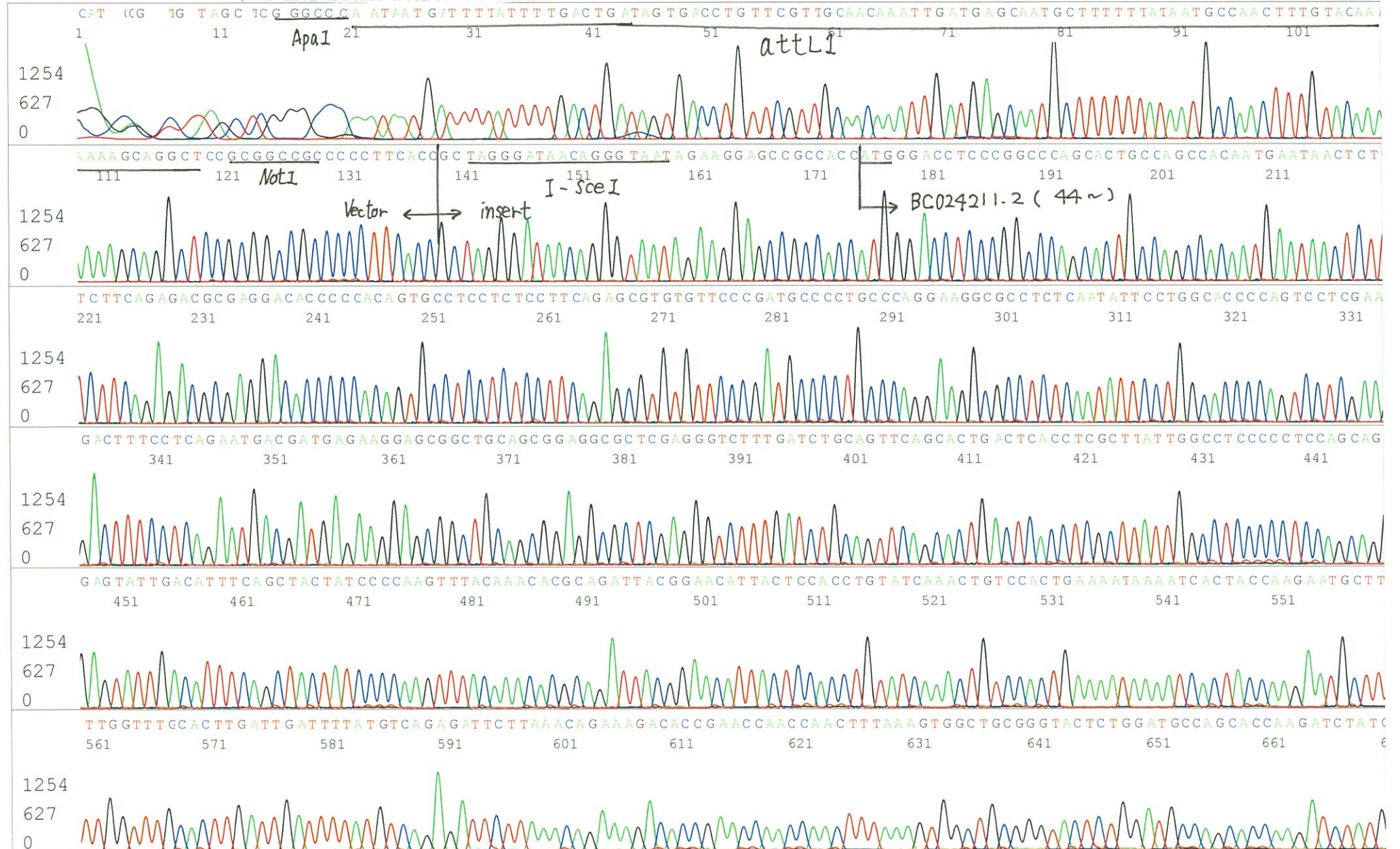


APPROVED BY : <http://dna.brc.riken.jp/index.html>

S/N G:417 A:503 T:362 C:458  
KB.bcp  
KB 1.4.1.8 Cap:21

Primer A : M13

5' GTTTTCCCAGTCACGACGTTGTA 3'



S/N G:417 A:503 T:362 C:458  
KB.bcp  
KB 1.4.1.8 Cap:21

