

**RIKEN Clone ID : IRAK004G03**

Vector : pCMV-SPORT6

Gene	RAD18		
Accession No.	BC001302.1	1896 bp	1..1896
	<i>CDS</i>	1488 bp	58..1545

● Plasmid DNA purification

Date : 130213

Culture : LB (100 ug/ml Ampicillin) 5 ml → at 37 deg C over nigh

Date : 130214

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

● Digestion by restriction enzyme/Concentraron calibration

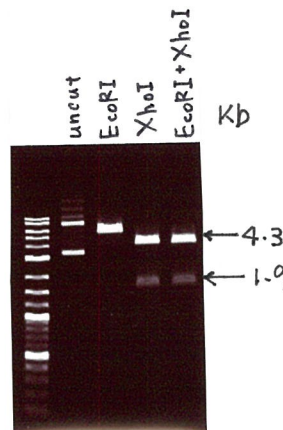
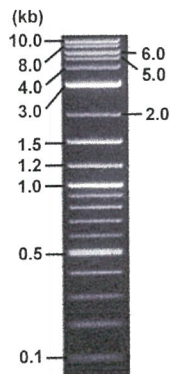
Date : 130214

DNA concentration (O.D.): 235 ng/ul

DNA	0.2 ul
Enzyme (EcoRI + XhoI)	0.5 + 0.5 ul
Buffer H	1 ul
dH <sub>2</sub> O	7.8 ul
<b>Total</b>	<b>10 ul</b>

Erectrophoresis : 1% agarose gel, 1x TAE Buffer

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



<Expected size of fragment from BC001302.1 >

4339, 1872, 67 bp

● Confirmation of the insertion sequence

Date : 130221

Primer A	SP6
Primer B	M13
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>

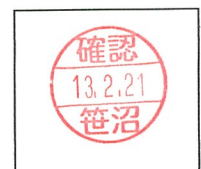
● Adjustment of DNA concentration

Shipping amount : 40 ul

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

Date : 130221

DNA ( <u>235</u> ng/ul )	85.0 ul
10x TE	80.0 ul
dH <sub>2</sub> O	634.0 ul
<b>Total</b>	<b>799.0 ul</b>



APPROVED BY :

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

S/N G:48 A:36 T:37 C:30  
KB.bcp  
KB 1.4:1.8 Cap:11

Primer A : SP6  
5' ATTTAGGTGACACTATAG 3'

