

**RIKEN Clone ID : IRAK022A21**

Vector : pCMV-SPORT6

Gene	HAX1	
Accession No.	BC015209.1	1082 bp
	CDS	840 bp

● Plasmid DNA purification

Date : 22.04.26

Culture : LB (100 ug/ml Ampicillin) 5 ml -> at 37 deg C over night

Date : 22.04.27

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

● Digestion by restriction enzyme/Concentration calibration

Date : 22.04.27

DNA concentration (O.D.): 26 ng/ul

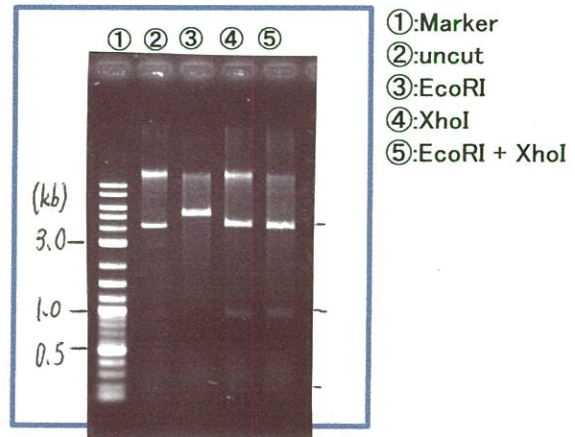
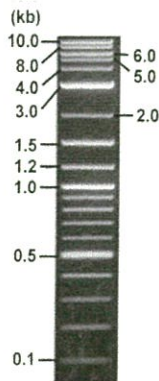
< Size of fragment expected from this clone >

DNA	3.8 ul
Enzyme (EcoRI + XhoI)	0.5 + 0.5 ul
Buffer H	1 ul
dH <sub>2</sub> O	4.2 ul
Total	10 ul

EcoRI	5.5 kb
XhoI	4.5, 1.0 kb
EcoRI + XhoI	4.3kb (Vector) 1.0, 0.2 kb

Electrophoresis : 1% agarose gel, 1x TAE Buffer

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



● Adjust plasmid DNA solution to 25 ng/ul

Date : 22.04.28

DNA ( <u>26</u> ng/ul)	<u>66.0</u> ul
10x TE	— ul
dH <sub>2</sub> O	— ul
Total	<u>66.0</u> ul

● Confirmation of the insertion sequence

Date : 22.04.27

Primer A	Reverse2
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>

● Shipping

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

Conc.: 26 ng/ul, Volume: 40 ul

APPROVED BY :

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



S/N G:16 A:14 T:8 C:11

Primer A : Reverse2

7893\_IRAK022A21\_B2D9\_Reverse2

Apr 27,2022 02:40PM, JST

KB.bcp

KB\_3500\_POP7\_BDTv3.mob

Apr 27,2022 02:57PM, JST

KB 1.4.1.8 Cap:7

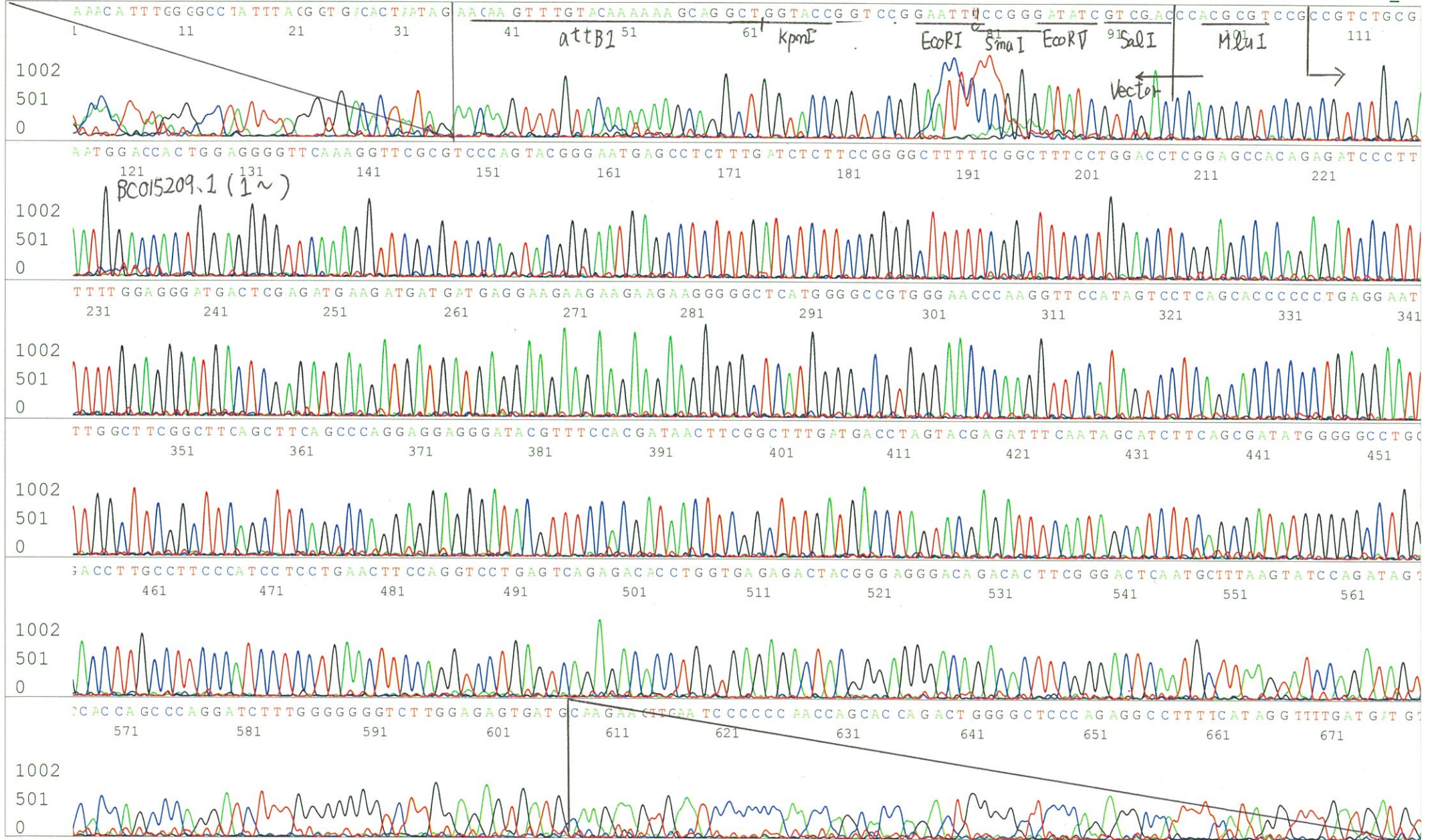
5' GCGGATAACAATTTTCACACAGG 3'

Pts 1628 to 8704 Pk1 Loc:1596

Spacing:10.86 Pts/Panel1350

Version 6.0 HiSQV Bases: 607

Plate Name: 20220427\_mix



S/N G:12 A:10 T:6 C:8

Primer B : M13

7893\_IRAK022A21\_B2D9\_M13(-40)

Apr 27, 2022 02:40PM, JST

KB.bcp

KB\_3500\_POP7\_BDTv3.mob

Apr 27, 2022 02:57PM, JST

KB 1.4.1.8 Cap:8

5' GTTTTCCCAGTCACGACGTTGTA 3'

Pts 1653 to 8704 Pk1 Loc:1621

Spacing:10.94 Pts/Panel1350

Version 6.0 HiSQV Bases: 527

Plate Name: 20220427\_mix

