

**RIKEN Clone ID : IRAK062H10**

Vector : pCMV-SPORT6

Gene	ICOS	
Accession No.	BC028210.1	2651 bp
	CDS	600 bp

● Plasmid DNA purification

Date : 22.10.11

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

Date : 22.10.12

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

● Digestion by restriction enzyme/Concentration calibration

Date : 22.10.12

DNA concentration (O.D.): 401 ng/ul

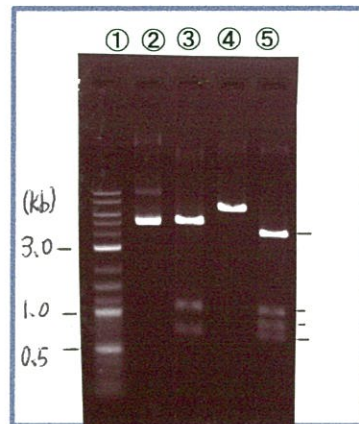
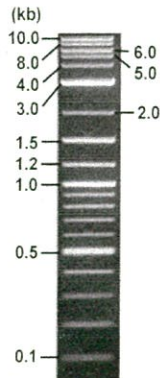
< Size of fragment expected from this clone >

DNA	0.25 ul
Enzyme (EcoRI + XhoI)	0.5 + 0.5 ul
Buffer H	1 ul
dH <sub>2</sub> O	7.75 ul
Total	10 ul

EcoRI	5.2, 1.1, 0.7 kb
XhoI	7.0 kb
EcoRI + XhoI	4.3kb (Vector) 1.1, 0.9, 0.7 kb

Electrophoresis : 1% agarose gel, 1x TAE Buffer

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



- ①:Marker
- ②:uncut
- ③:EcoRI
- ④:XhoI
- ⑤:EcoRI + XhoI

● Adjust plasmid DNA solution to 25 ng/ul

Date : 22.10.18

DNA ( 401 ng/ul)	86.0 ul
10x TE	137.9 ul
dH <sub>2</sub> O	1155.5 ul
Total	1379.4 ul

● Confirmation of the insertion sequence

Date : 22.10.13

Primer A	Reverse2
Primer B	M13
Primer C	poly(A) region primer #0
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:21 A:34 T:24 C:31

Primer A : Reverse2

7893\_IRAK062H10\_B2lr\_1\_Reverse2

Oct 13,2022 01:50PM, JST

KB.bcp

KB\_3500\_POP7\_BDTv3.mob

Gray line indicates quality value

Oct 13,2022 02:13PM, JST

KB 1.4.1.8 Cap:1

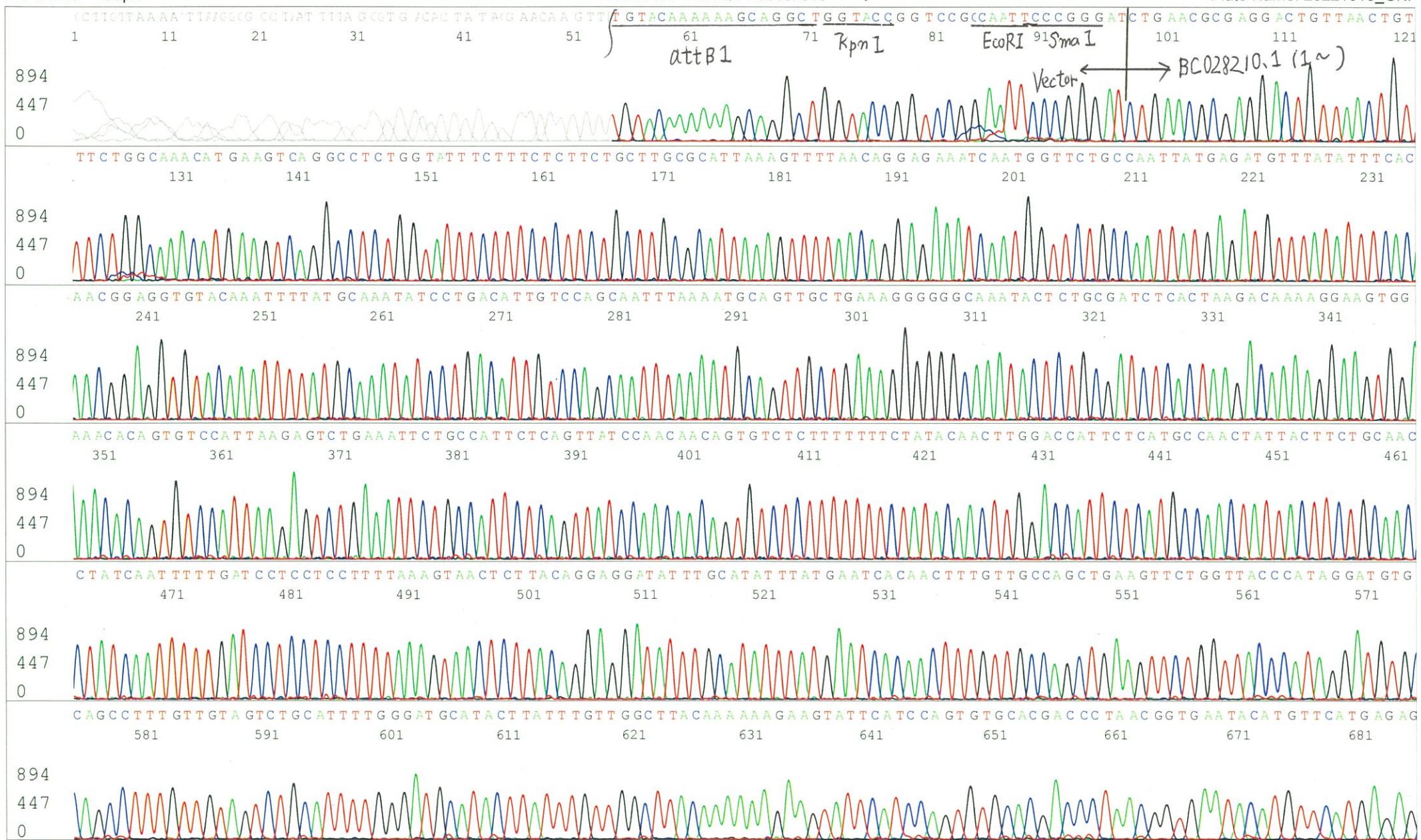
5' GCGGATAACAATTTTCACACAGG 3'

Pts 1539 to 12212 Pk1 Loc:1522

Version 6.0 HiSQV Bases: 813 (QV) < 40.

Spacing:13.41 Pts/Panel1350

Plate Name: 20221013\_GNP





S/N G:19 A:28 T:22 C:25

Primer B : M13

7893\_IRAK062H10\_B2lr\_1\_M13(-40)

Oct 13,2022 01:50PM, JST

KB.bcp

KB\_3500\_POP7\_BDTv3.mob

Gray line indicates quality value

Oct 13,2022 02:13PM, JST

KB 1.4.1.8 Cap:2

5' GTTTTCCCAGTCACGACGTTGTA 3'

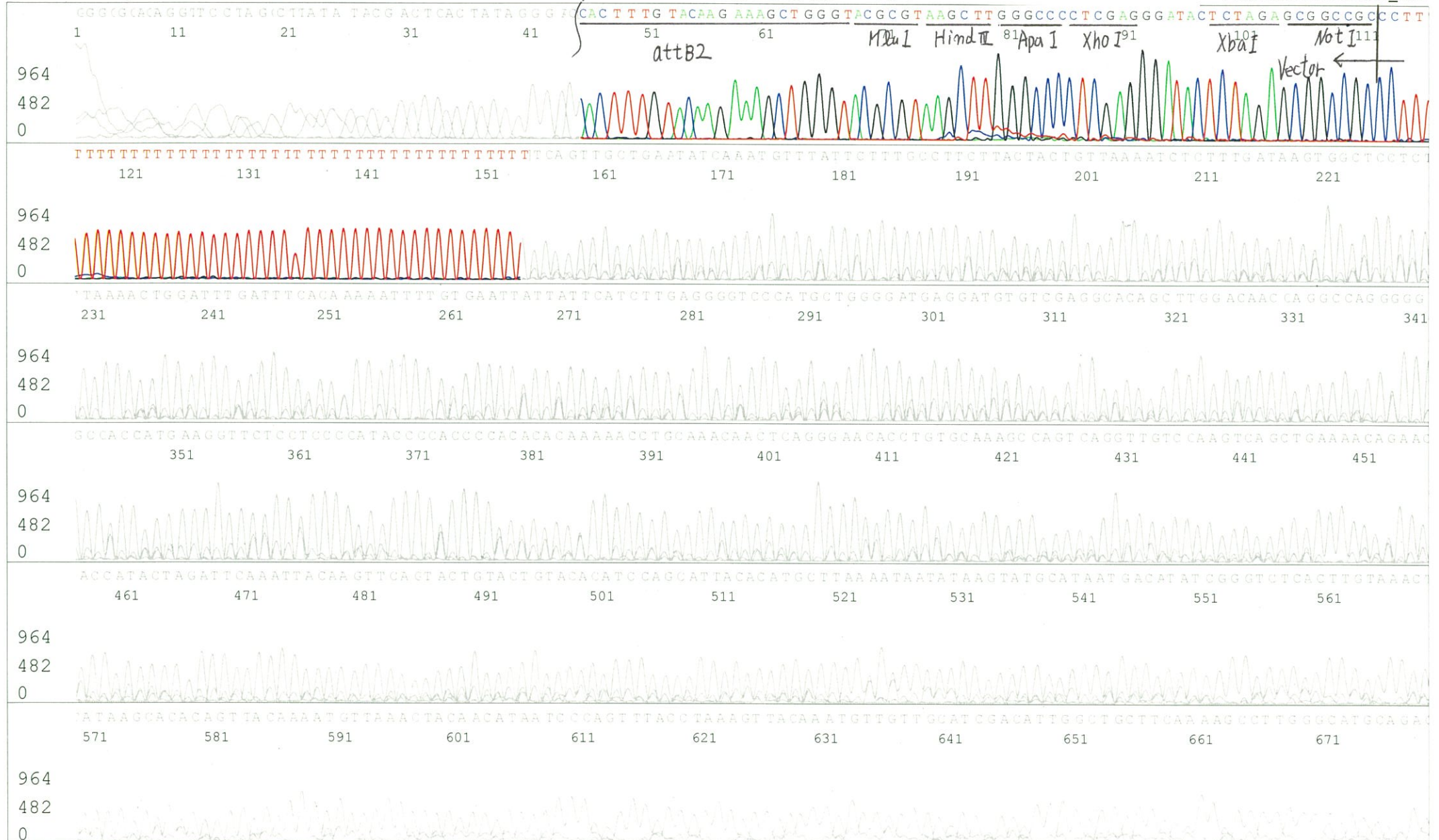
Pts 1538 to 12059 Pk1 Loc:1521

Version 6.0 HiSQV Bases: 539

(QV) < 40.

Spacing:13.35 Pts/Panel1350

Plate Name: 20221013\_GNP





S/N G:6 A:9 T:7 C:8

Primer C : Poly(A) region primer #0

7893\_IRAK062H10\_B2lr\_1\_GNPmixture

Oct 13,2022 01:50PM, JST

KB.bcp

KB\_3500\_POP7\_BDTv3.mob

Gray line indicates quality value

Oct 13,2022 02:13PM, JST

KB 1.4.1.8 Cap:3

5' TTTTTTTTTTTTTTTTTTTTTTVNN 3'

Pts 1555 to 12058 Pk1 Loc:1538

(QV) < 40.

Spacing:13.23 Pts/Panel1350

Version 6.0 HiSQV Bases: 530

Plate Name: 20221013\_GNP

