

**RIKEN Clone ID : ARi60G12**

Vector : pGCAP10

Gene	COL4A1	
RefSeq_mRNA	NM_001845.6	6540 bp

● Plasmid DNA purification

Date : 190205

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

Date : 190206

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

● Digestion by restriction enzyme/Concentration calibration

Date : 190206

DNA concentration (O.D.): 292 ng/ul

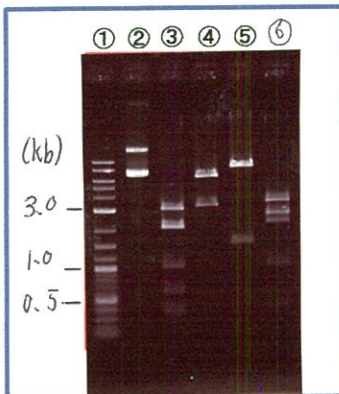
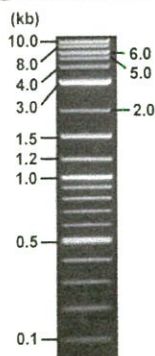
< Size of fragment expected from this clone >

DNA	<u>0.3</u> ul
Enzyme (BamHI)	1 ul
Buffer H	1 ul
dH <sub>2</sub> O	<u>7.7</u> ul
<b>Total</b>	<b>10 ul</b>

BamHI 3.1kb (vector), 2.1, 2.1, 1.0, 0.6, 0.3, 0.3, 0.1 kb (unsure)

Erectrophoresis : 1% agarose gel, 1x TAE Buffer

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



- ①:Marker
- ②:uncut
- ③:BamHI
- ④: SwaI + Not I
- ⑤: SwaI + EcoRV
- ⑥: NheI + EcoRI

● Confirmation of the insertion sequence

Date : 190207

Primer A	pGCAP-F
Primer B	pGCAP-R
Primer C	Poly(A) region
List of Sequencing Primers	<a href="http://dna.brc.riken.jp/en/NRCDhum2en.html">http://dna.brc.riken.jp/en/NRCDhum2en.html</a>

● Adjust plasmid DNA solution to 25 ng/ul

Date : 190212

DNA ( <u>292</u> ng/ul)	<u>84.0</u> ul
10x TE	<u>98.1</u> ul
dH <sub>2</sub> O	<u>799.0</u> ul
<b>Total</b>	<b>981.1 ul</b>

● Shipping



Conc. : 25 ng/μl, Volume : 40 μl



~~Conc. :     ng/μl, Volume :     μl~~

APPROVED BY :



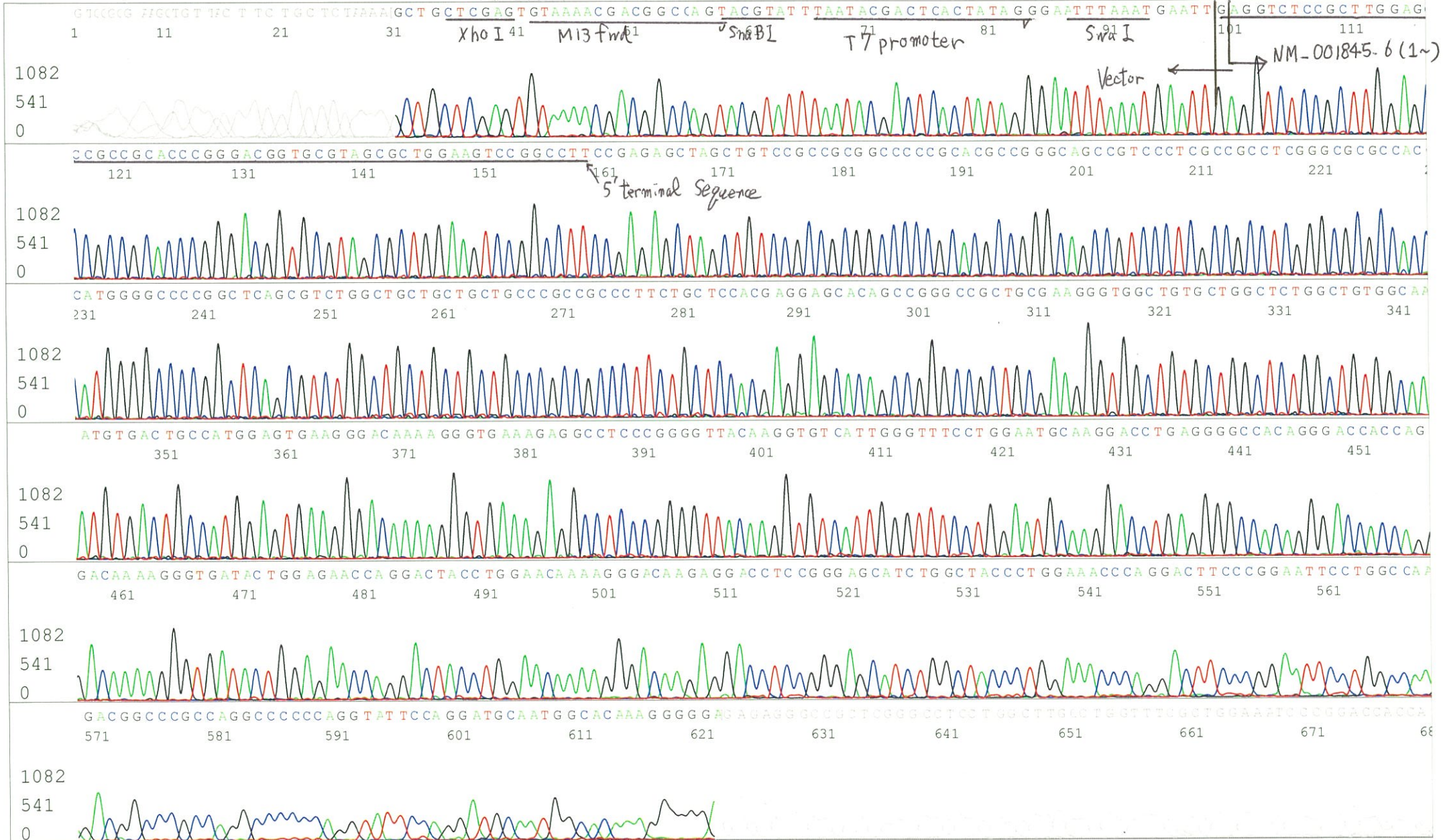
S/N G:56 A:43 T:43 C:70  
KB.bcp  
KB 1.4.1.8 Cap:3

Primer A : pGCAP-F  
5' CTCAGTGGATGTTGCCTTTAC 3'

ARI60G12\_A9B4\_III\_pGCAP\_F  
KB\_3500\_POP7\_BDTv3.mob  
Pts 1758 to 8703 Pk1 Loc:1726  
Version 6.0 HiSQV Bases: 683

Gray line indicates quality value  
(QV) < 40.

Feb 07,2019 10:21AM, JST  
Feb 07,2019 10:37AM, JST  
Spacing:11.4 Pts/Panel1350  
Plate Name: 20190207\_GNP





S/N G:21 A:29 T:20 C:22

Primer C : Poly(A) region primer #0

ARI60G12\_A9B4\_III\_GNP Mixture

KB\_3500\_POP7\_BDTv3.mob

Pts 1778 to 8704 Pk1 Loc:1746

Version 6.0 HiSQV Bases: 651

Gray line indicates quality value (QV) < 40.

Feb 08,2019 09:37AM, JST

Feb 08,2019 10:07AM, JST

Spacing:11.59 Pts/Panel1350

Plate Name: 20190208\_GNP

KB 1.4.1.8 Cap:2

5' TTTTTTTTTTTTTTTTTTTTTTNNN 3'

