

**RIKEN Clone ID : W01A094E05**

Vector : pENTR-TOPO

Gene	WNT3A	
Accession ID of Origin	AK056278.1	2932 bp
	<i>CDS</i>	1059 bp

● Plasmid DNA purification

Date : 171109

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

Date : 171114

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

● Digestion by restriction enzyme/Concentration calibration

Date : 171114

DNA concentration (O.D.): 64 ng/ul

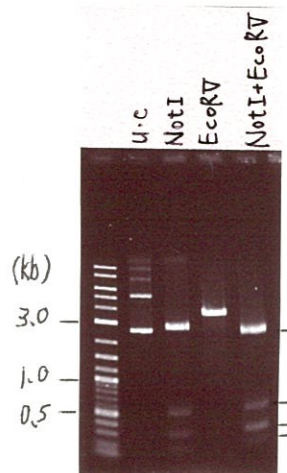
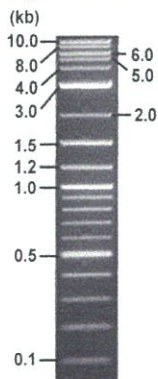
< Size of fragment expected from this clone >

DNA	1.5	ul
Enzyme (NotI + EcoRV)	0.5 + 0.5	ul
Buffer H	1	ul
dH <sub>2</sub> O	6.5	ul
Total	10	ul

NotI	2.7, 0.5, 0.3, 0.2 kb		
EcoRV	3.8 kb		
NotI + EcoRV	2.5kb (Vector)	0.5, 0.3, 0.3, 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 : 171117

DNA ( <u>64</u> ng/ul )	<u>70.0</u>	ul
10x TE	<u>17.9</u>	ul
dH <sub>2</sub> O	<u>91.3</u>	ul
Total	<u>179.2</u>	ul

● Confirmation of the insertion sequence

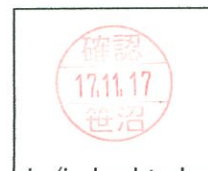
Date : 171116

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:172 A:119 T:109 C:184

Primer A : M13

W01A094E05\_A7K9\_M13(-40)

KB\_3500\_POP7\_BDTv3.mob

Pts 1776 to 8707 Pk1 Loc:1744

Version 6.0 HiSQV Bases: 710

Gray line indicates quality value

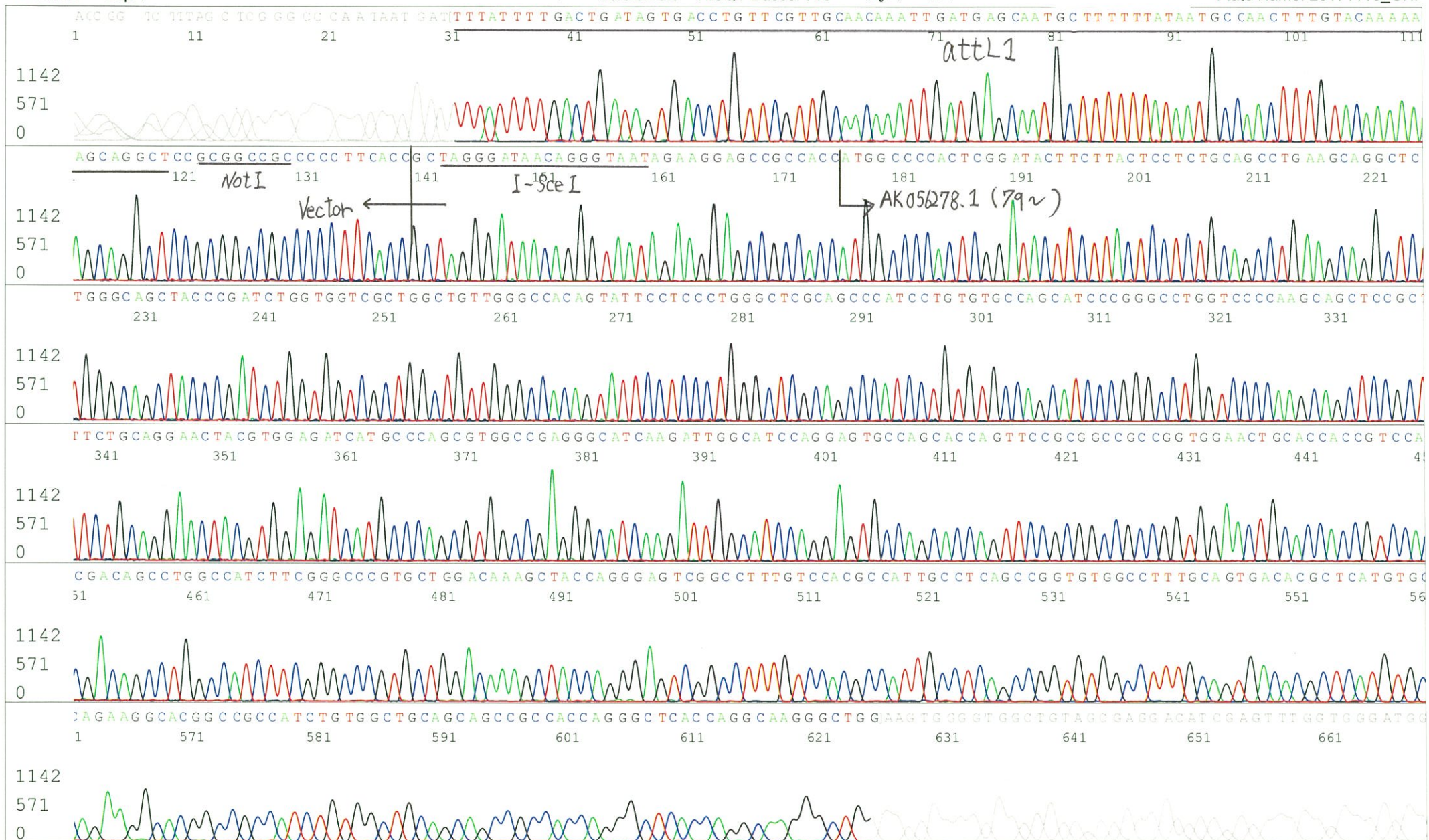
(QV) < 40.

Nov 16,2017 10:15AM, JST

Nov 16,2017 10:43AM, JST

Spacing:10.7 Pts/Panel1350

Plate Name: 20171116\_GNP





S/N G:222 A:189 T:169 C:285  
KB.bcp  
KB 1.4.1.8 Cap:4

Primer B : T7 long

W01A094E05\_A7K9\_T7long  
KB\_3500\_POP7\_BDTv3.mob  
Pts 1775 to 8707 Pk1 Loc:1743  
Version 6.0 HiSQV Bases: 707

Gray line indicates quality value  
(QV) < 40.

Nov 16,2017 10:15AM, JST  
Nov 16,2017 10:43AM, JST  
Spacing:10.54 Pts/Panel1350  
Plate Name: 20171116\_GNP

