

**RIKEN Clone ID : W01A127B16**

Vector : pENTR-TOPO

Gene	NOTCH1	
Accession No.	AK000012.1	1487 bp
	<i>CDS</i>	426 bp
		1..1487
		674..1099

● Plasmid DNA purification

Date : 140319

Culture : LB (25 ug/ml Kanamycin) 10 ml -> at 37 deg C over night

Date : 140320

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

● Digestion by restriction enzyme/Concentration calibration

Date : 140320

DNA concentration (O.D.): 114 ng/ul

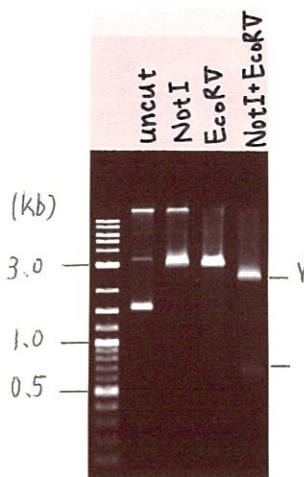
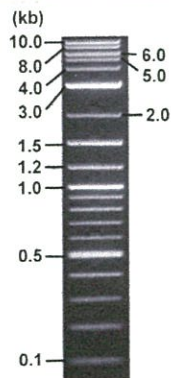
<Size of fragment expected from AK000012.1 >

DNA	0.4	ul
Enzyme (NotI + EcoRV)	0.5 + 0.5	ul
Buffer H	1	ul
dH <sub>2</sub> O	7.6	ul
Total	10	ul

NotI	3198 bp
EcoRV	3198 bp
NotI + EcoRV	2553bp (Vector)      645 bp

Electrophoresis : 1% agarose gel, 1x TAE Buffer

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



● Confirmation of the insertion sequence

Date : 140326

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>

● Adjustment of DNA concentration

Shipping amount : 40 ul

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

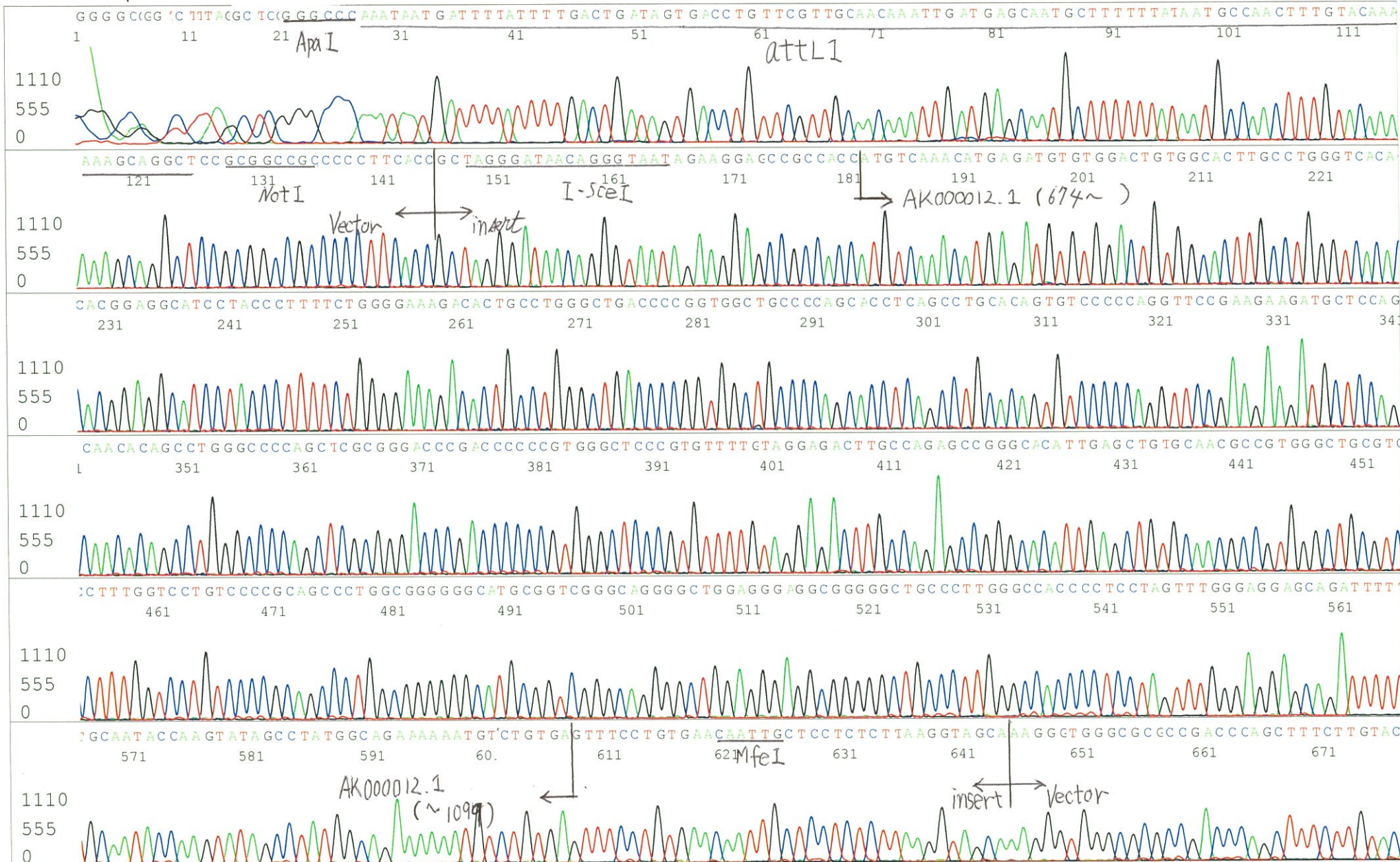
Date : 140326

DNA ( <u>114</u> ng/ul)	86.0	ul
10x TE	39.2	ul
dH <sub>2</sub> O	267.0	ul
Total	392.2	ul

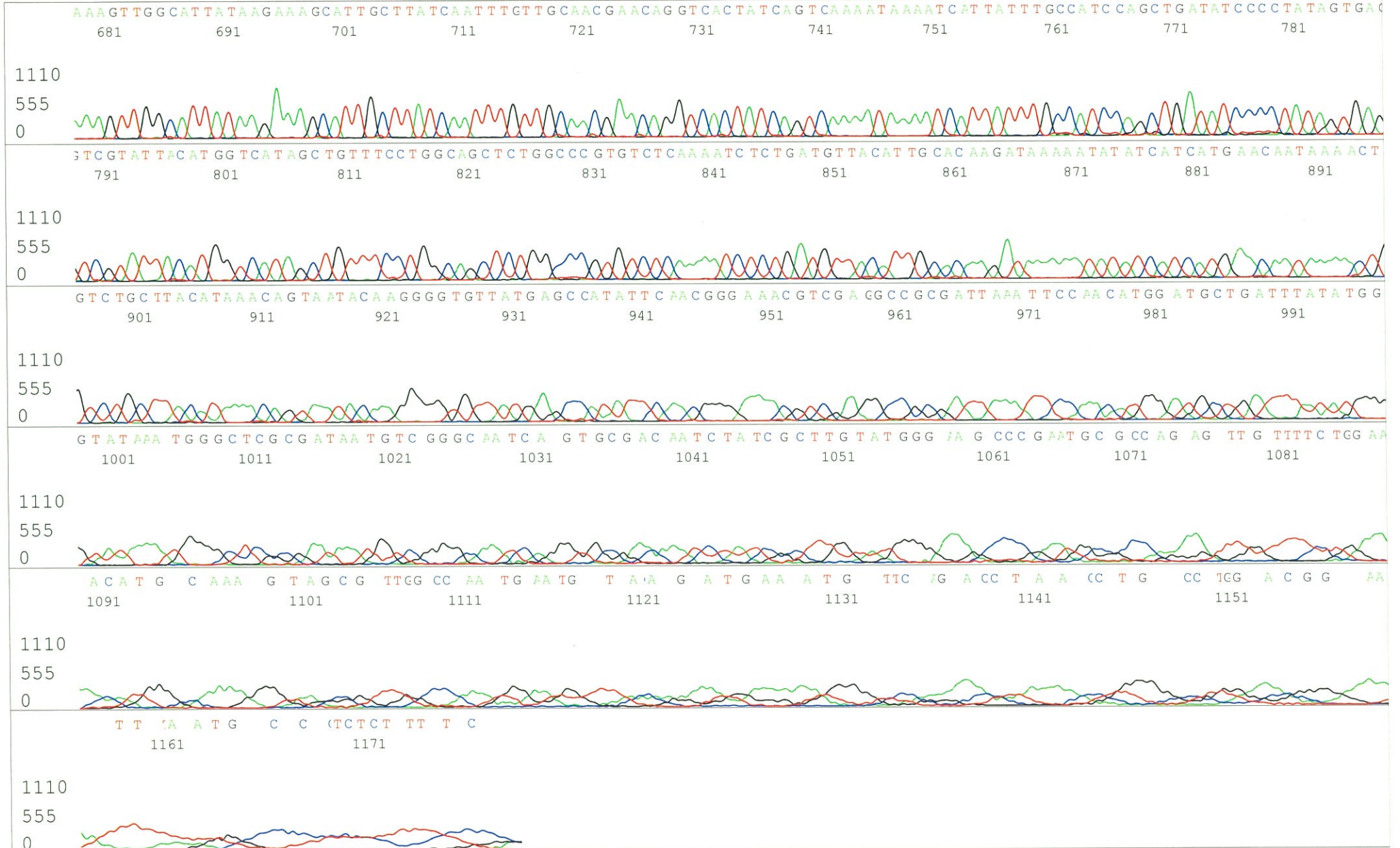


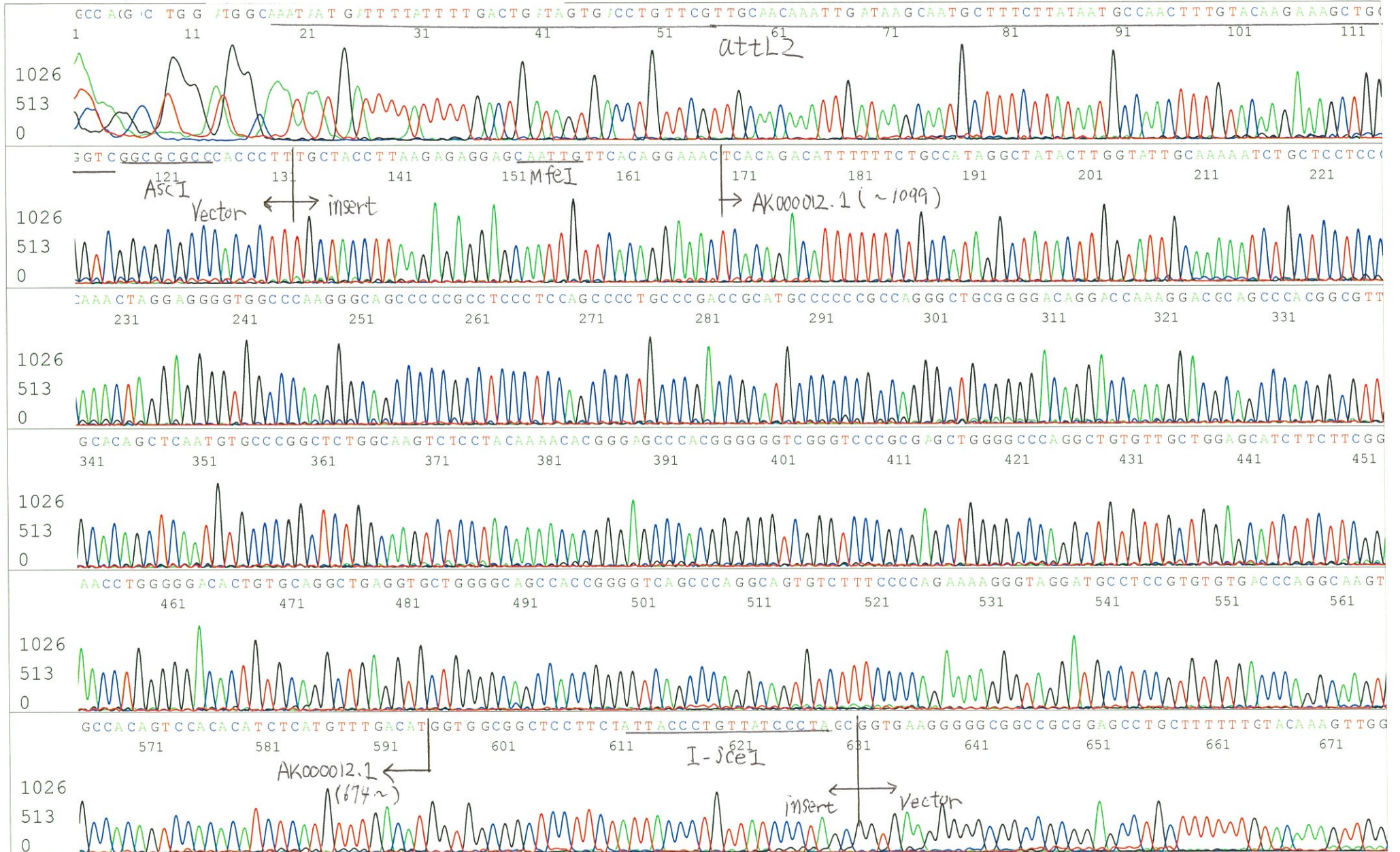
APPROVED BY :

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



S/N G:49 A:45 T:43 C:53  
KB.bcp  
KB 1.4.1.8 Cap:19





S/N G:58 A:63 T:61 C:74  
KB.bcp  
KB 1.4.1.8 Cap:22

