

**RIKEN Clone ID : IRAK079K02**

Lot#: 7893\_A3I2

Vector : pCMV-SPORT6

|               |            |                   |
|---------------|------------|-------------------|
| Gene          | DEADC1     |                   |
| Accession No. | BC037955.1 | 2074 bp < 1..2074 |
|               | CDS        | 582 bp < 1..582   |

● Plasmid DNA purification

Date : 130902

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

Date : 130903

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

● Digestion by restriction enzyme/Concentration calibration

Date : 130903

DNA concentration (O.D.): 264 ng/ul

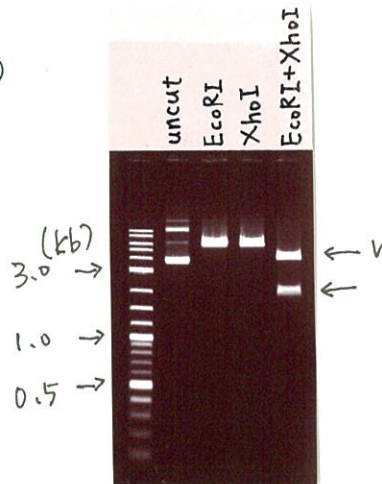
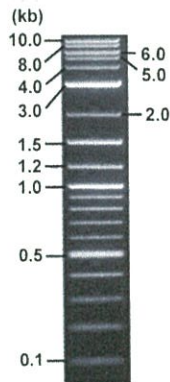
< Size of fragment expected from IRAK079K02 >

|                       |           |    |
|-----------------------|-----------|----|
| DNA                   | 0.2       | ul |
| Enzyme (EcoRI + XhoI) | 0.5 + 0.5 | ul |
| Buffer H              | 1         | ul |
| dH <sub>2</sub> O     | 7.8       | ul |
| Total                 | 10        | ul |

|              |                         |
|--------------|-------------------------|
| EcoRI        | 6456 bp                 |
| XhoI         | 6456 bp                 |
| EcoRI + XhoI | 4339bp (Vector) 2117 bp |

Electrophoresis : 1% agarose gel, 1x TAE Buffer

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



● Confirmation of the insertion sequence

Date : 130905

|                            |   |
|----------------------------|---|
| 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> |

● Adjustment of DNA concentration

Shipping amount : 40 ul

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

Date : 130905

|                   |       |    |
|-------------------|-------|----|
| DNA ( 264 ng/ul)  | 85.0  | ul |
| 10x TE            | 89.8  | ul |
| dH <sub>2</sub> O | 722.8 | ul |
| Total             | 897.6 | ul |



APPROVED BY :

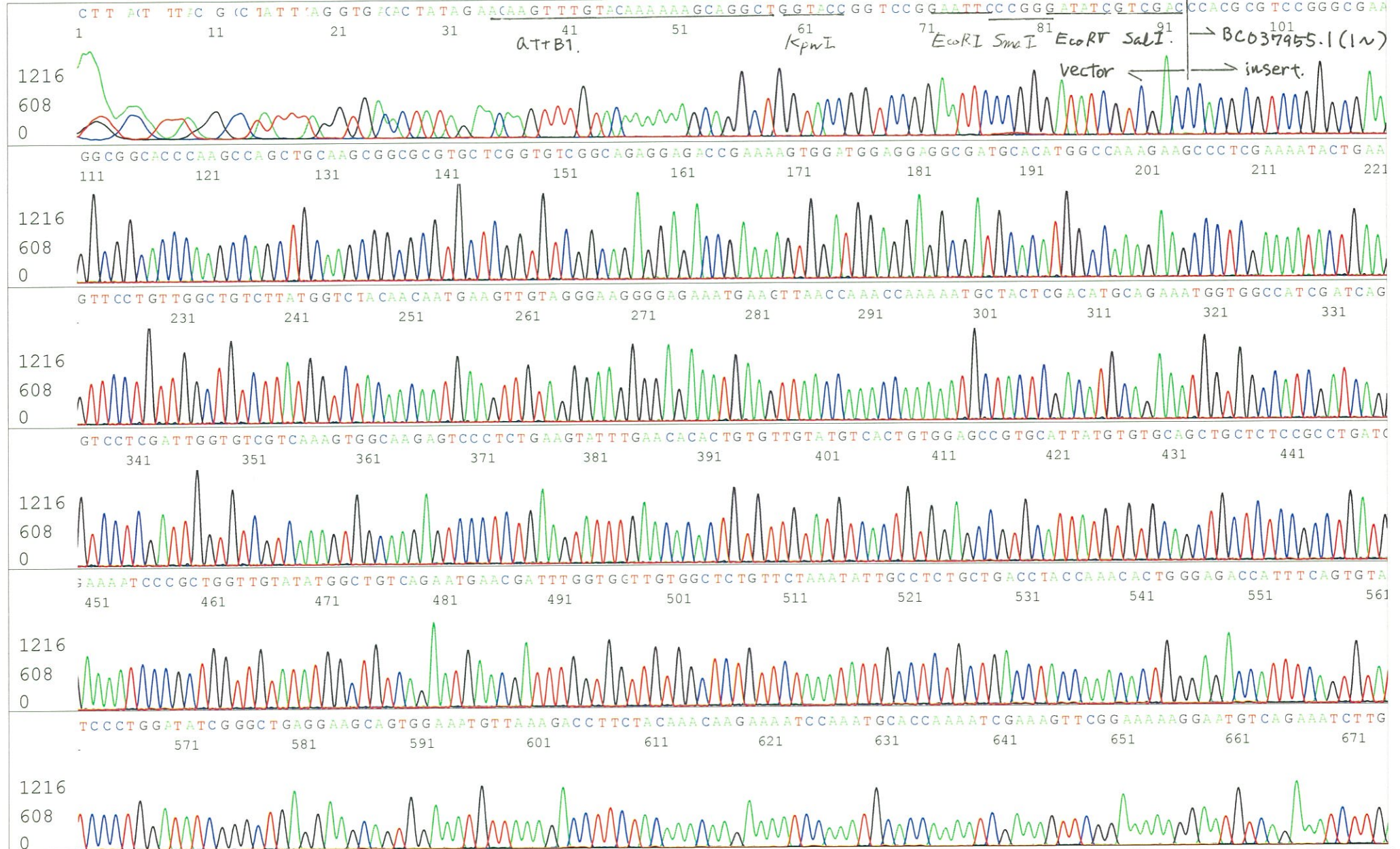
<http://dna.brc.riken.jp/index.html>

S/N G:188 A:125 T:101 C Primer A : Reverse2

KB.bcp

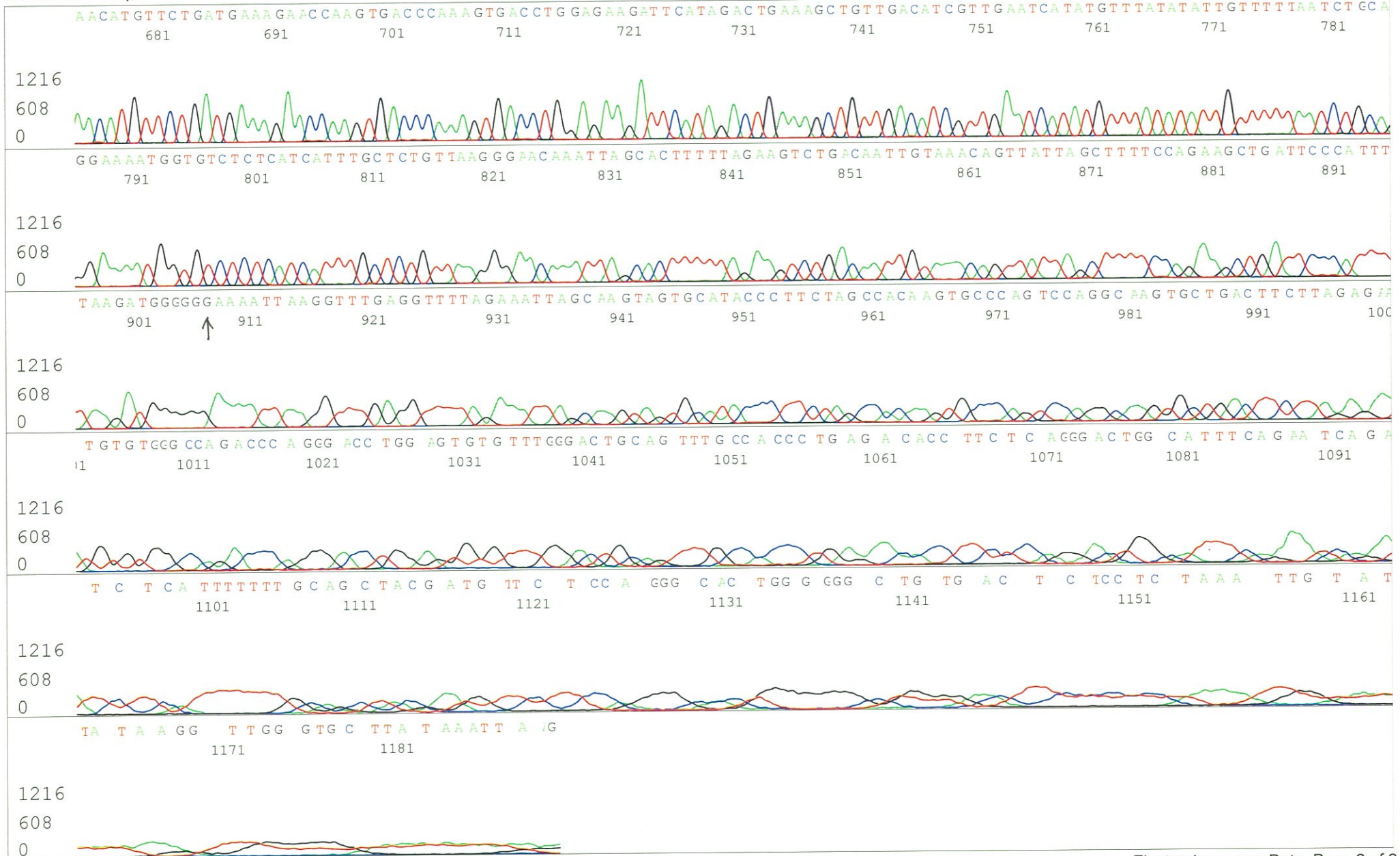
KB 1.4.1.8 Cap:1

5' GCGGATAACAATTTTCACACAGG 3'





S/N G:188 A:125 T:101 C:83  
KB.bcp  
KB 1.4.1.8 Cap:1



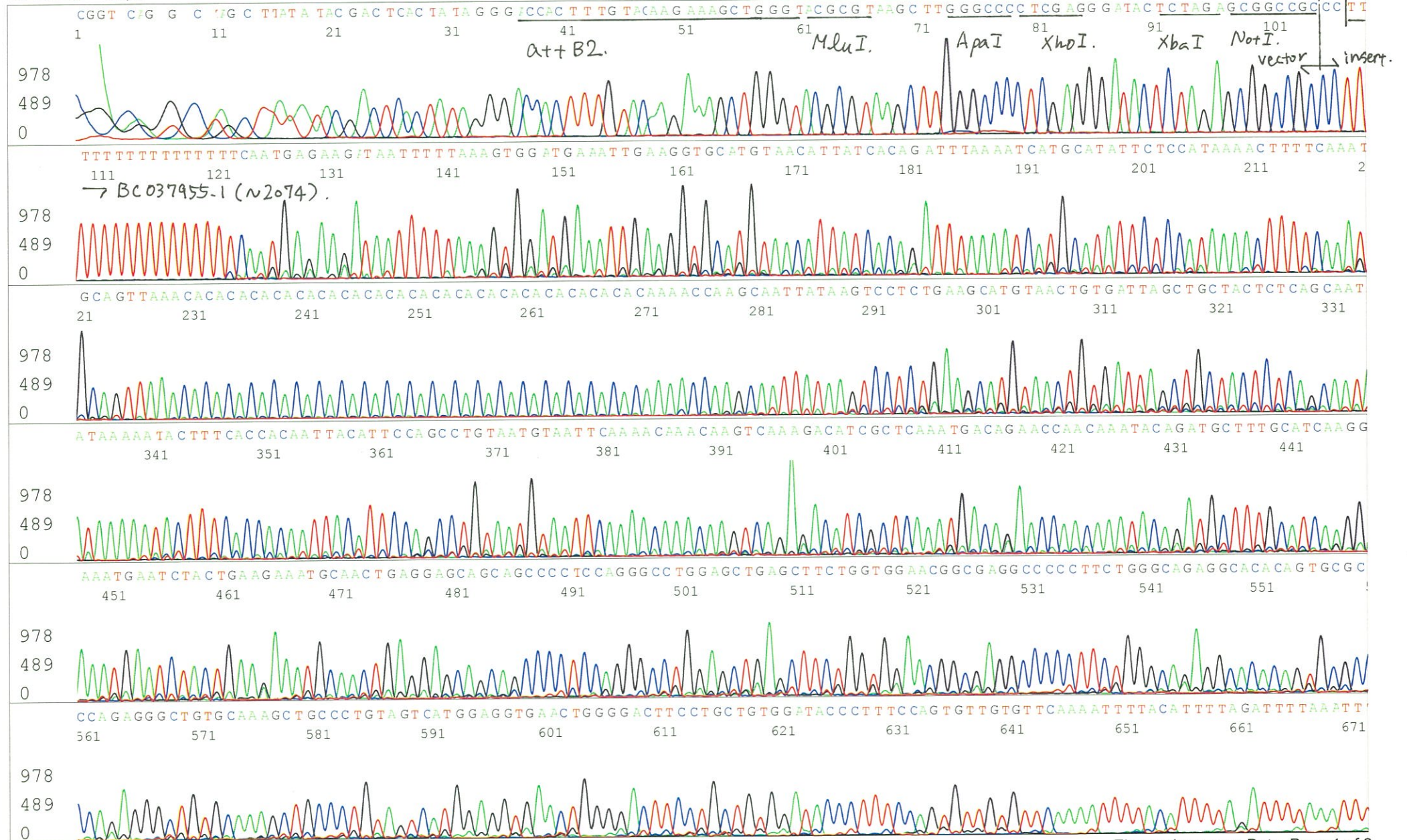


S/N G:395 A:417 T:369 (Primer B : M13

KB.bcp

KB 1.4.1.8 Cap:4

5' GTTTTCCCAGTCACGACGTTGTA 3'





S/N G:395 A:417 T:369 C:270  
KB.bcp  
KB 1.4.1.8 Cap:4

