

**RIKEN clone ID: M01C003B02**

Vector : pDONR221

Gene	GJA1		
Accession No.	BC026329.1	3000 bp	1..3000
	<i>CDS</i>	1149 bp	89..1237
	<i>Insert Fragment</i>	1363 bp	89..1459

● Plasmid DNA purification

Date : 110516 Culture : LB (25 ug/ml Kanamycin) 3 ml → 37°C O/N

Date : 110517 Purification : QIAGEN Miniprep kit → dH<sub>2</sub>O 100 ul

● Digestion by restriction enzyme / Concentration calibration

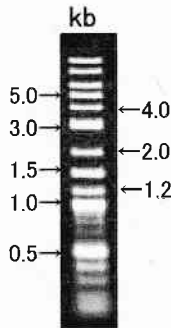
Date : 110517

DNA concentration (O.D.): 51.32 ng/ul

DNA	2	ul
Restriction enzyme (Hinc II + EcoRV)	0.5+0.5	ul
Buffer H	1	ul
dH <sub>2</sub> O	6	ul
<b>Total</b>	<b>10</b>	<b>ul</b>

Electrophoresis: 1% agarose gel, 1 × TAE Buffer

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



<Expected digestion pattern from DNA Sequence>

1436, 2263 bp 1658, 2263 bp

● Adjust plasmid DNA solution to 25 ng/ul ~preparation for shipping~

Date : 110519 Shipped : 25 ng/ul, 40 ul

Final concentration: 25 ng/ul

DNA ( <u>51.32</u> ng/ul)	82.0	ul
10 × TE	16.8	ul
dH <sub>2</sub> O	69.5	ul
<b>Total</b>	<b>168.3</b>	<b>ul</b>



Project : GNP

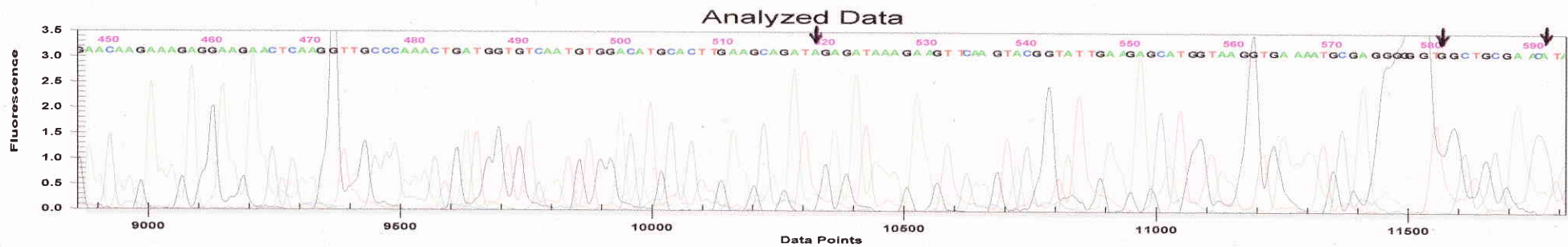
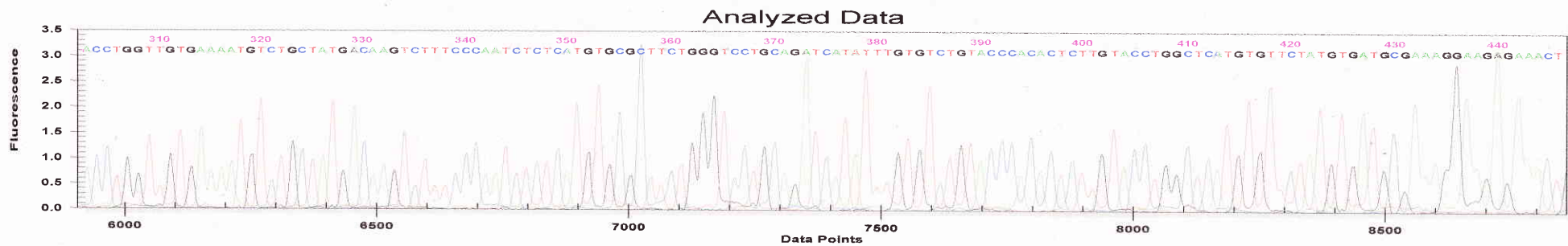
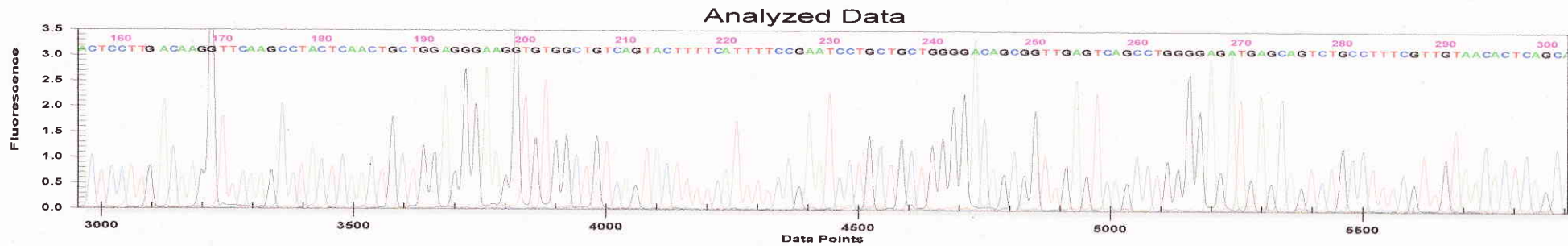
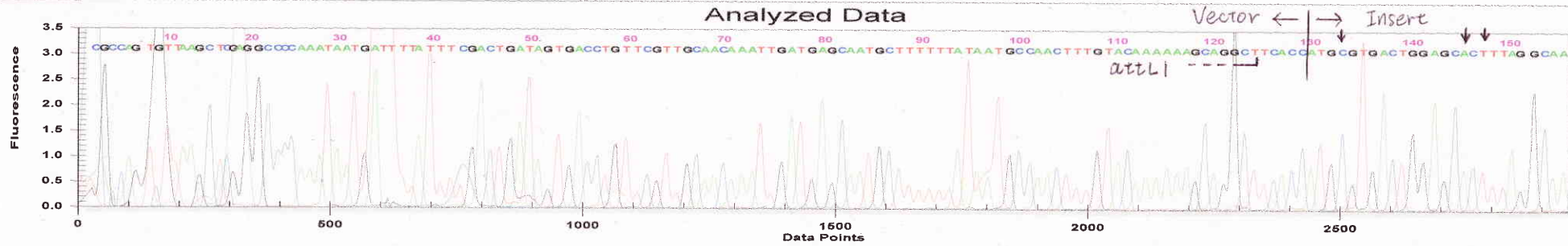
System : System 1

Operator : 1.110518.furu

Sample : M01C003B02\_M13.B04\_11051814GZ

Instrument : System 1 (Ver. 9.0.25)

Result : M01C003B02\_M13.B04\_11051814GZ

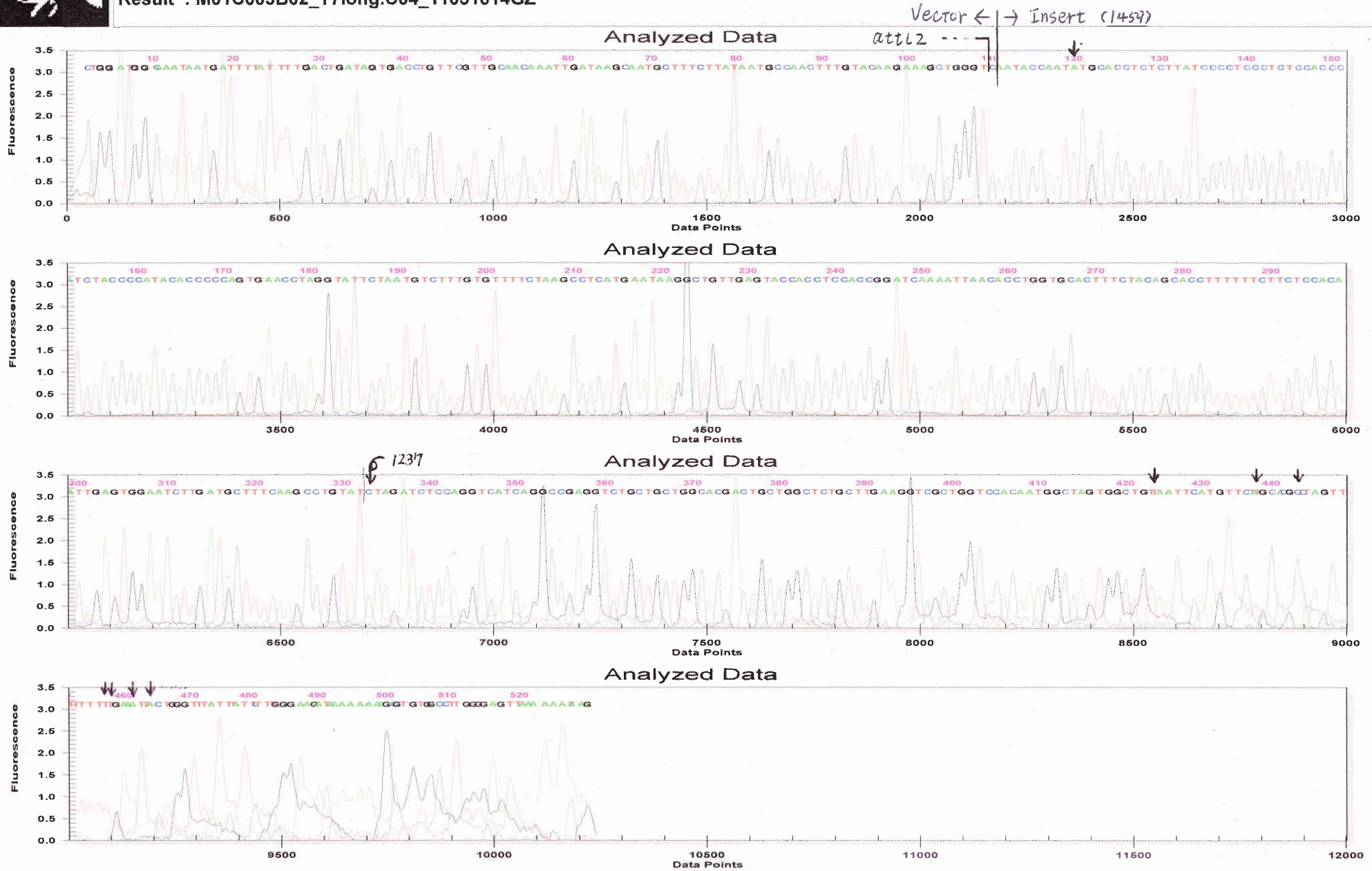




Project : GNP  
 Sample : M01C003B02\_T7long.C04\_11051814GZ  
 Result : M01C003B02\_T7long.C04\_11051814GZ

System : System 1

Operator : 1.110518.furu  
 Instrument : System 1 (Ver. 9.0.25)



[ GENETYX : Homology Data ]

Date : 2011.05.19

Query Sequence

File Name :
Sequence Name : BC026329
Sequence Size : 3000

Target Sequence

File Name : M01C003B02\_M13.B04\_11051814GZ
Sequence Name : M01C003B02\_M13.B04\_11051814GZ
Sequence Size : 711

Unit Size to Compare = 6
Pick up Location No. = 1

Query Range: 85 - 670
Sbjct Range: 125 - 710
587 bp, INT.Score: 2018, OPT.Score: 2229
Identity: 570 / 587 (97%)
Strand: Plus / Plus

Table with 4 columns: Query, Sbjct, Sequence alignment (with vertical bars and arrows), and Position. Includes handwritten annotations like 'insert' and '↑'.

[ GENETYX : Homology Data ]

Date : 2011.05.19

## Query Sequence

File Name :  
 Sequence Name : BC026329  
 Sequence Size : 3000

## Target Sequence

File Name : M01C003B02\_T7long.C04\_11051814GZ  
 Sequence Name : M01C003B02\_T7long.C04\_11051814GZ (complement)  
 Sequence Size : 529

Unit Size to Compare = 6

Pick up Location No. = 1

Query Range: 1107 - 1552

Sbjct Range: 59 - 517

459 bp, INT.Score: 1246, OPT.Score: 1350

Identity: 393 / 459 (85%)

Strand: Plus / Minus

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Query 1107 ATAACCAG-- AATTC--AAA AAAACTA-GC -TGC--TGGA CATGAATT-A CAGCCACTAG 1157
          | | |||| | ||| ||| ||||| || | || | ||||| | |||||
Sbjct 59 AAACCAGTA ATTTCAAAA AAAACTAGGC GTGCATAGAA CATGAATTAA CAGCCACTAG 118

Query 1158 CCATTGTGGA CCAGCGACCT TCAAGCAGAG CCAGCAGTCG TGCCAGCAGC AGACCTCGGC 1217
          ||||| ||||| ||||| ||||| ||||| |||||
Sbjct 119 CCATTGTGGA CCAGCGACCT TCAAGCAGAG CCAGCAGTCG TGCCAGCAGC AGACCTCGGC 178

Query 1218 CTGATGACCT GGAGATCTAG; ATACAGGCTT GAAAGCATCA AGATTCCACT CAATTGTGGA 1277
          ||||| ||||| ||||| ||||| ||||| |||||
Sbjct 179 CTGATGACCT GGAGATCTAG; ATACAGGCTT GAAAGCATCA AGATTCCACT CAATTGTGGA 238
                          1237

Query 1278 GAAGAAAAAA GGTGCTGTAG AAAGTGCACC AGGTGTTAAT TTTGATCCGG TGGAGGTGGT 1337
          ||||| ||||| ||||| ||||| ||||| |||||
Sbjct 239 GAAGAAAAAA GGTGCTGTAG AAAGTGCACC AGGTGTTAAT TTTGATCCGG TGGAGGTGGT 298

Query 1338 ACTCAACAGC CTTATTCATG AGGCTTAGAA AACACAAAGA CATTAGAATA CCTAGGTTCA 1397
          ||||| ||||| ||||| ||||| ||||| |||||
Sbjct 299 ACTCAACAGC CTTATTCATG AGGCTTAGAA AACACAAAGA CATTAGAATA CCTAGGTTCA 358

Query 1398 CTGGGGGTGT ATGGGGTAGA TGGGTGGAGA GGGAGGGGAT AAGAGAGGTG CATGTTGGTA 1457
          ||||| ||||| ||||| ||||| ||||| |||||
Sbjct 359 CTGGGGGTGT ATGGGGTAGA TGGGTGGAGA GGGAGGGGAT AAGAGAGGTG CATATTGGTA 418

Query 1458 TTTTAAAGTAG TGGATTCAA GAACTTAG-A TTATAAATAA GAGTTCCAT- --TAGGTGAT 1513
          || | | | | | | | | | | | | | | | | | | | | | |
Sbjct 419 TTGACCCAGC TTTCTTGTA AAAGTTGGCA TTATAAGAAA GCATTGCTTA TCAATTTGTT 478
          Insert ← | → vector

Query 1514 ACATAGATAA GGGCTTTTTC TCCCCGAAA CACCCCTAA 1552
          || | | | | | | | | | | | | | | | |
Sbjct 479 GCAACGAACA GGTCACATC AGTCAAAATA AAATCATT 517
  
```