

RIKEN Clone ID : RBb19C01

Lot# : 7676_A3Gb

Vector : pKA1U5

Gene	DCT		
RefSeq_mRNA	<i>NM_00192273</i>	2304 bp	1..2304
	<i>CDS</i>	1551 bp	428..1978
	<i>5'-terminal sequence</i>	1962 bp	343..2304

● **Plasmid DNA purification**

Date : 130711

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

Date : 130712

Purification : QIAGEN Miniprep kit -> dH₂O 100 ul

● **Digestion by restriction enzyme/Concentration calibration**

Date : 130712

DNA concentration (O.D.): 363 ng/ul

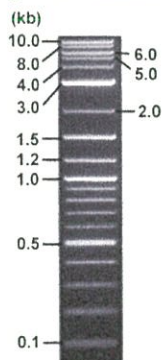
<Size of fragment expected from RBb19C01 >

DNA	0.2 ul
Enzyme (BamHI)	1 ul
Buffer H	1 ul
dH ₂ O	7.8 ul
Total	10 ul

BamHI 3183(vector), 2176 bp (unsure)

Electrophoresis : 1% agarose gel, 1x TAE Buffer

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



● **Confirmation of the insertion sequence**

Date : 130717

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

● **Adjustment of DNA concentration**

Shipping amount : 40 ul

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

Date : 130719

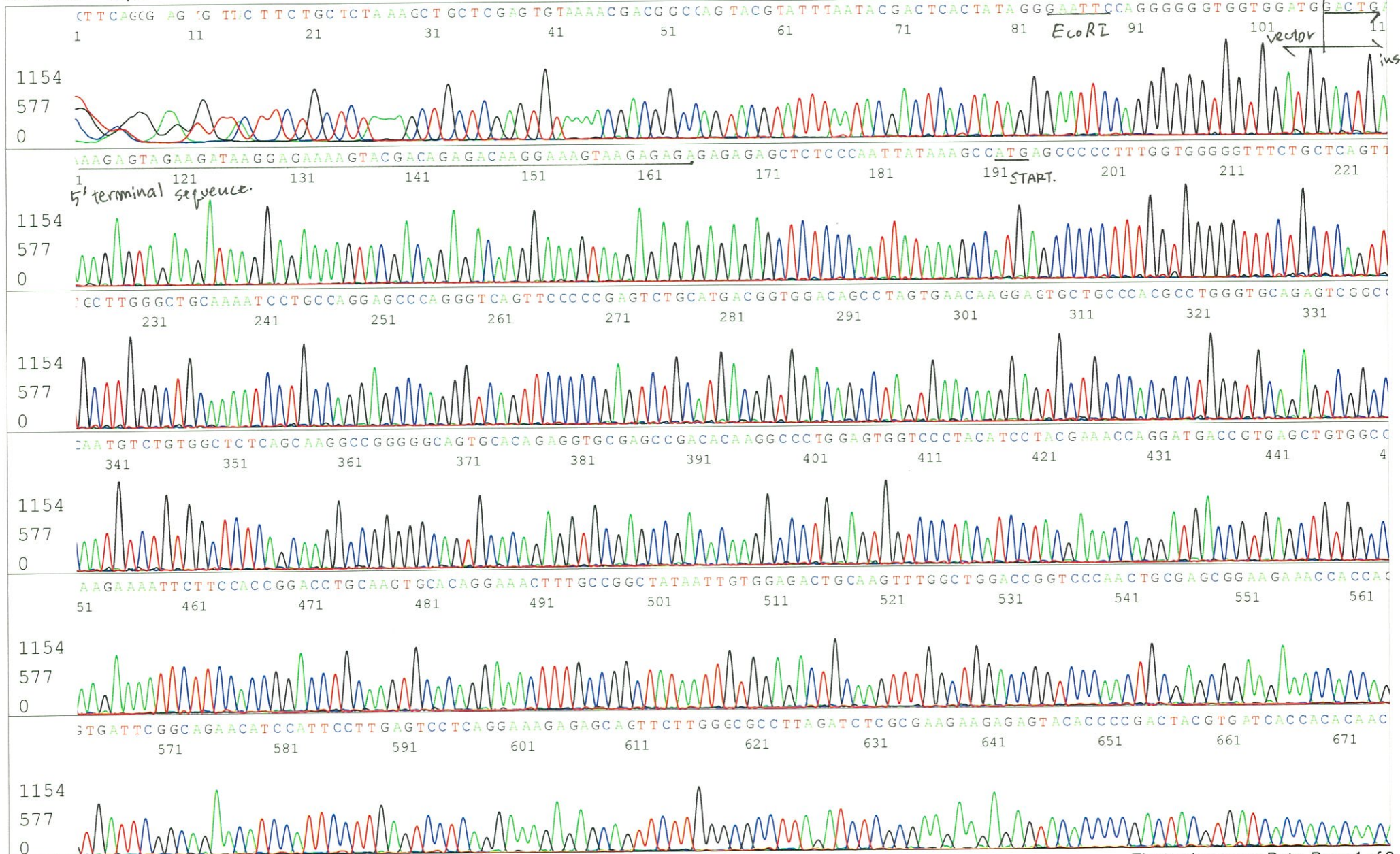
DNA (<u>363</u> ng/ul)	85.0 ul
10x TE	123.4 ul
dH ₂ O	1025.8 ul
Total	1234.2 ul



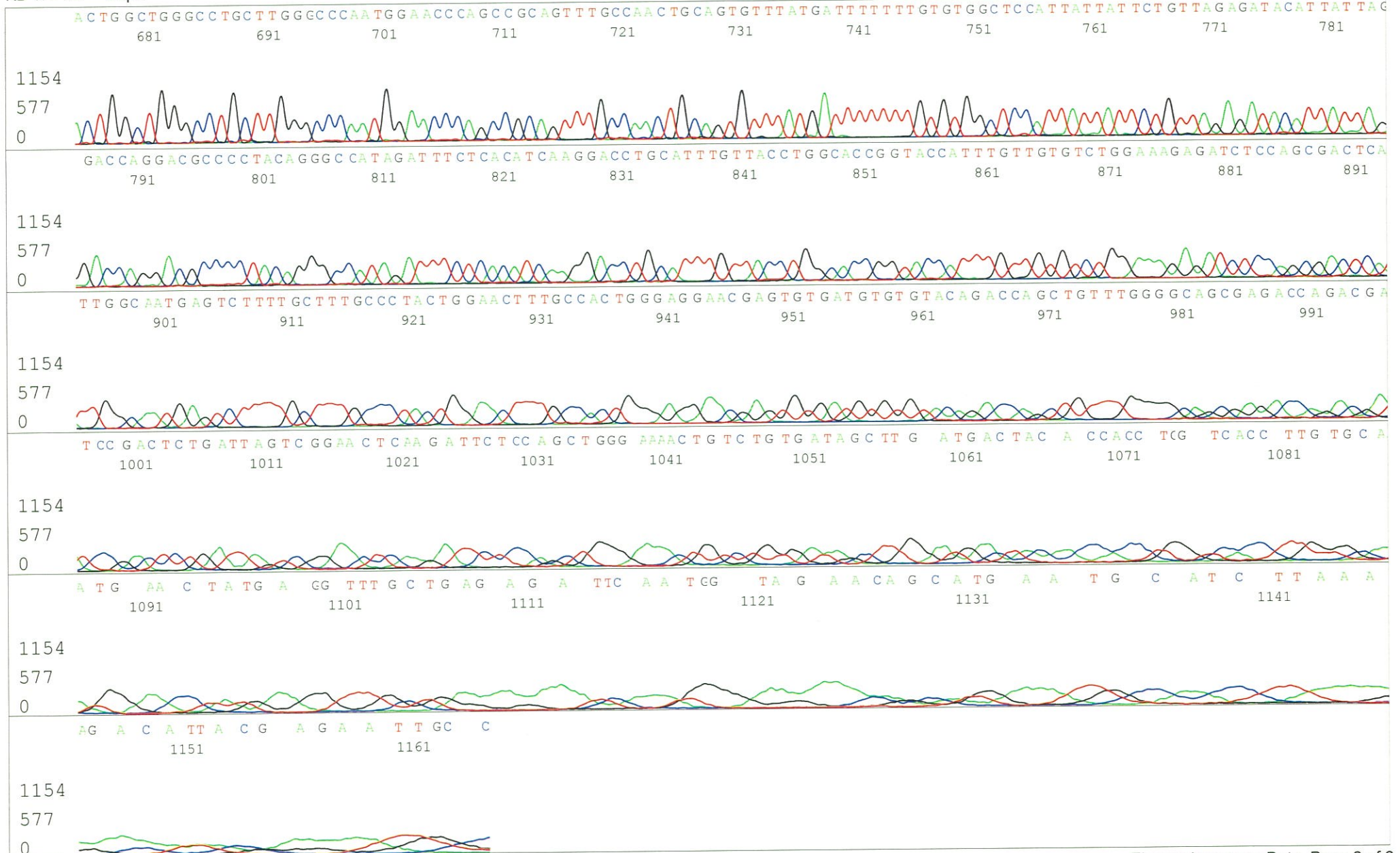
APPROVED BY :

S/N G:402 A:342 T:196 C:258 Primer A : pGCAP-F
KB.bcp
KB 1.4.1.8 Cap:7

5' CTCAGTGGATGTTGCCTTTAC 3'



S/N G:402 A:342 T:196 C:258
KB.bcp
KB 1.4.1.8 Cap:7

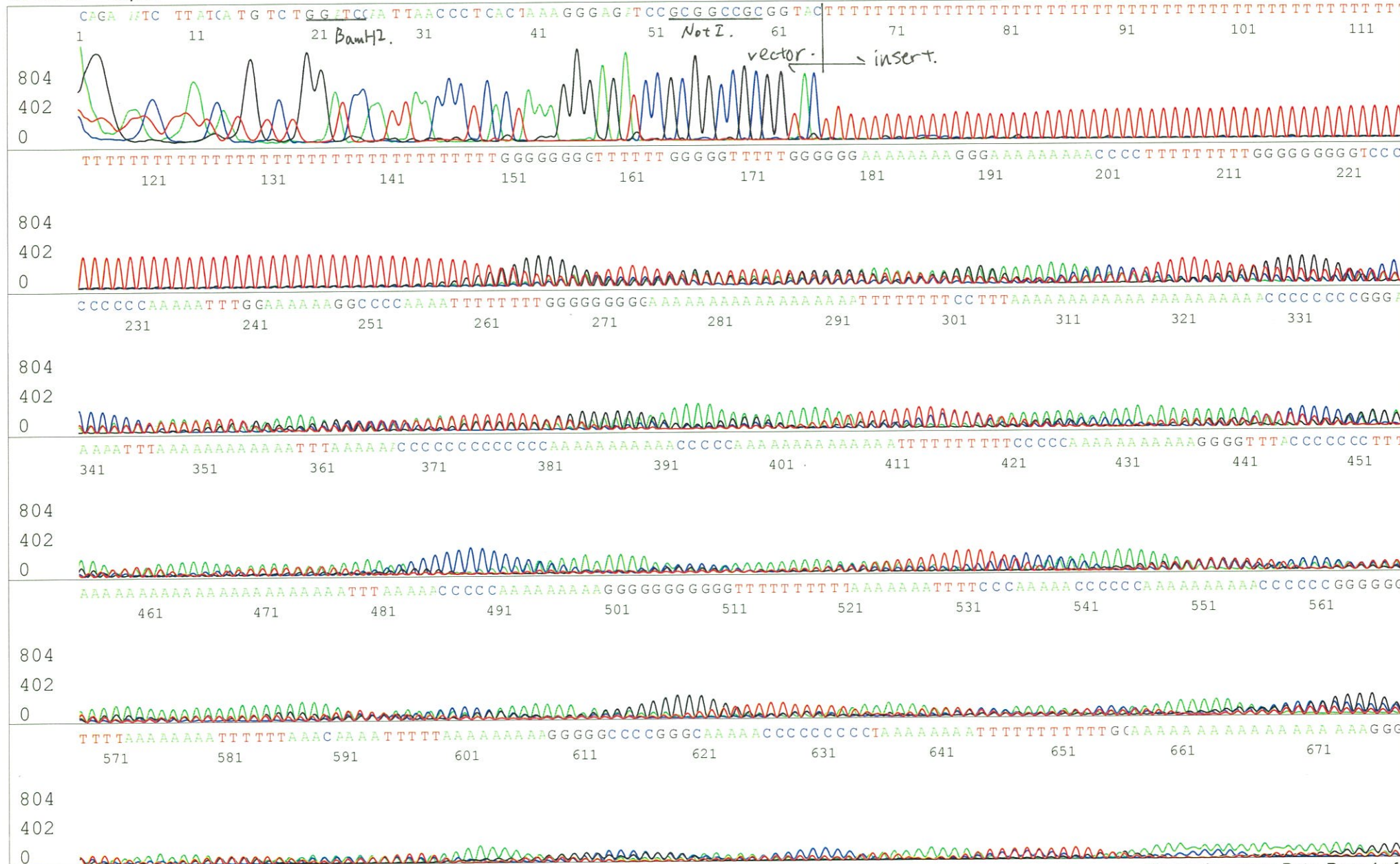


S/N G:46 A:53 T:62 C:32 Primer B : pGCAP-R

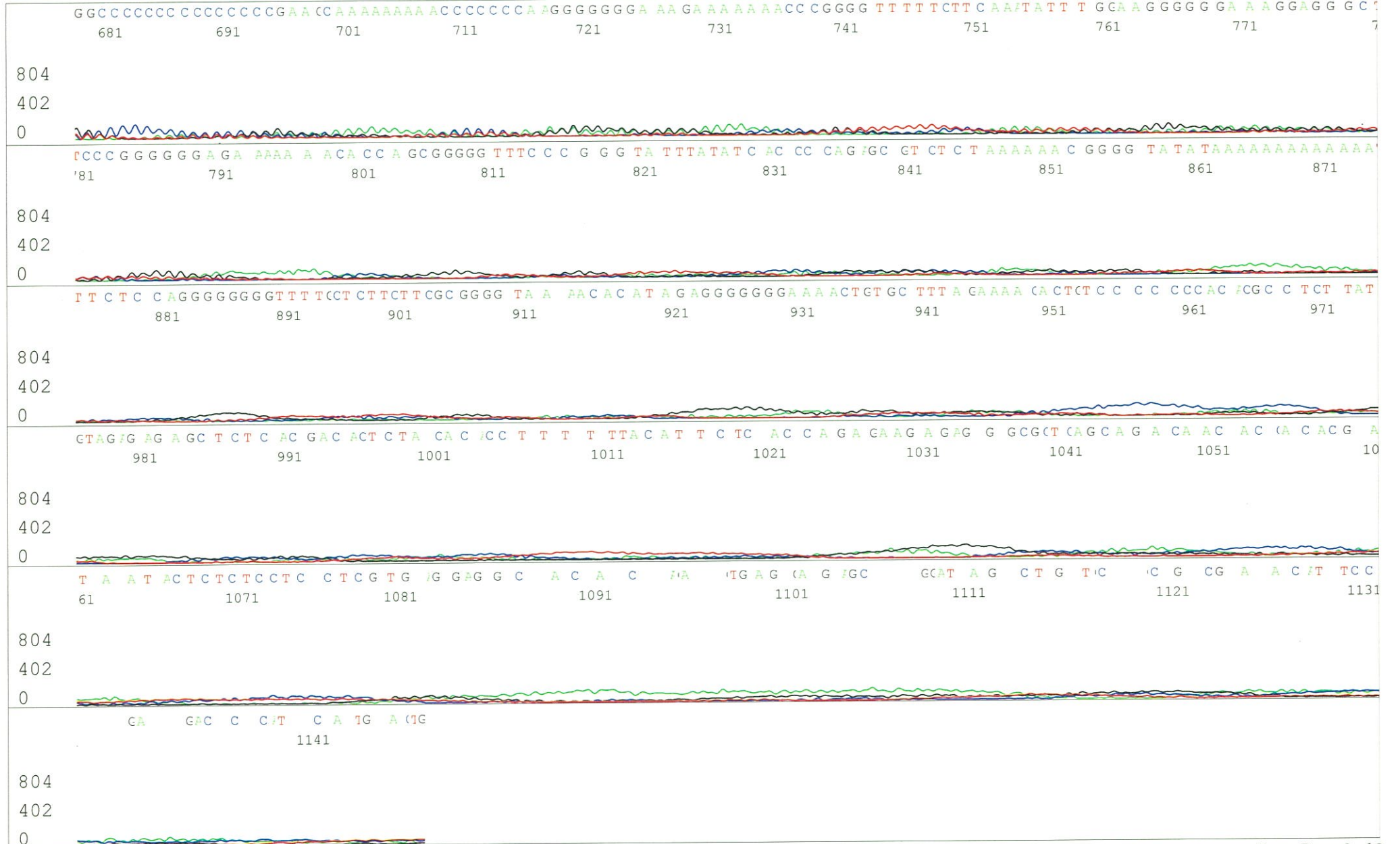
KB.bcp

5' GCATTCTAGTTGTGGTTTGTCC 3'

KB 1.4.1.8 Cap:10



S/N G:46 A:53 T:62 C:32
KB.bcp
KB 1.4.1.8 Cap:10



S/N G:56 A:80 T:61 C:35

Primer C : Poly(A) region primer #0

7676_RBb19C01_A3Gb_GNPmixture

Jul 17,2013 03:29PM, JST

KB.bcp

KB_3500_POP7_BDTv3.mob

Jul 17,2013 03:55PM, JST

KB 1.4.1.8 Cap:13

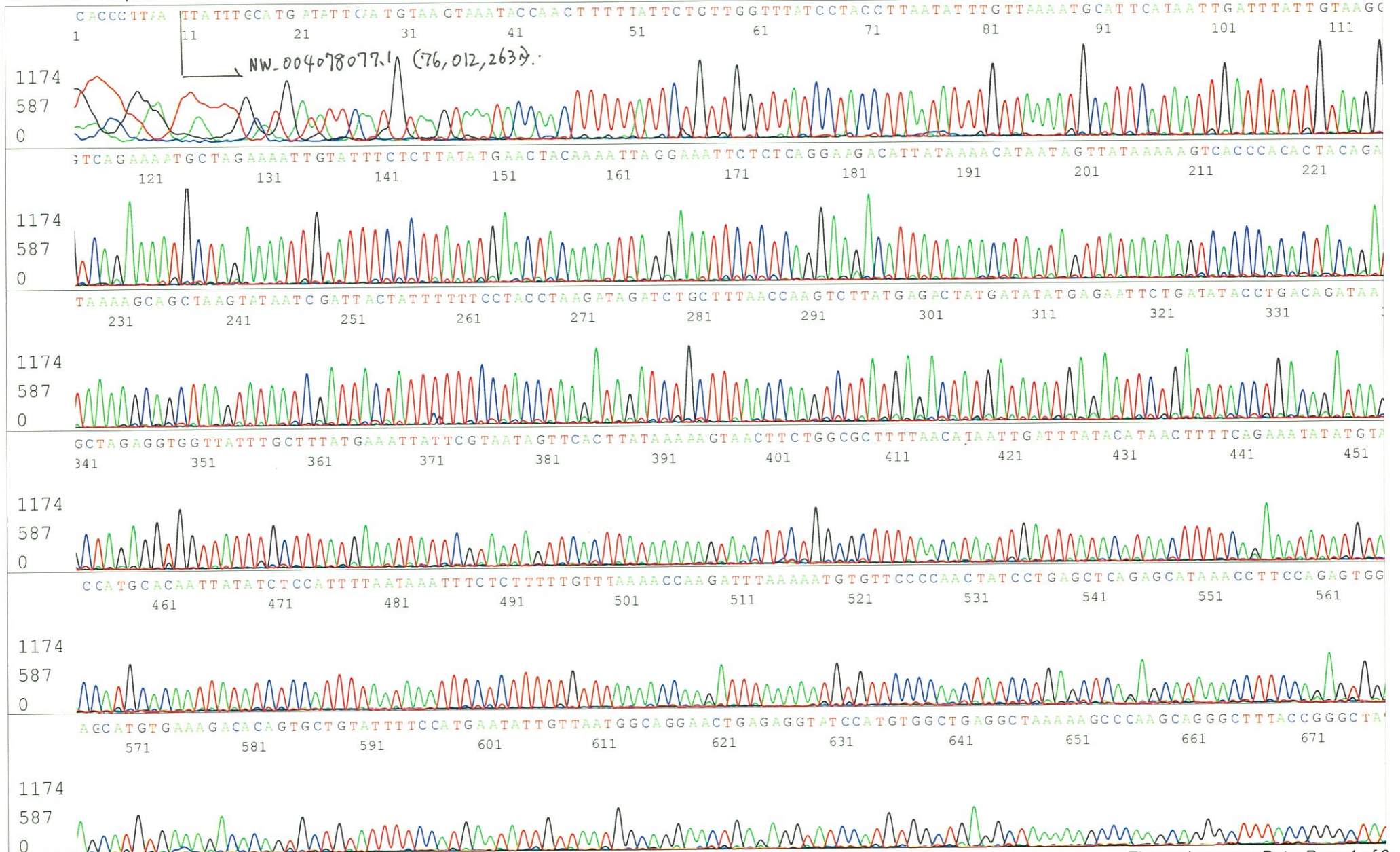
5' TTTTTTTTTTTTTTTTTTTTTNNN 3'

Pts 1299 to 13070 Pk1 Loc:1276

Spacing:11.6 Pts/Panel1350

Version 5.4 HiSQV Bases: 1006

Plate Name: 20130717_mix



S/N G:56 A:80 T:61 C:35
 KB.bcp
 KB 1.4.1.8 Cap:13

