Genome Network Project_Full -Length cDNA clone

Gene Engineering Division RIKEN BRC

RIKEN_Clone_ID : IRAK135G15

Lot#: 7893_A9Ee

Vector : pCMV-SPORT6

Gene	CD276	
Accession No.	BC062581.1	2550 bp
	CDS	1482 bp

● Plasmid DNA purification

Date : 190516

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

Date : 190517

Purification : QIAGEN Miniprep kit -> dH_2O 100 ul

● Digestion by restriction enzyme/Concentrarion calibration

Date : 190517

DNA concentration (O.D.): 88 ng/ul

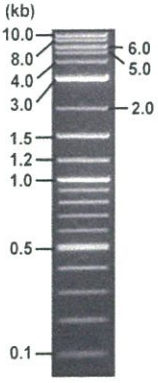
DNA	1.1 ul
Enzyme (EcoRI + XhoI)	0.5 + 0.5 ul
Buffer H	1 ul
dH_2O	6.9 ul
Total	10 ul

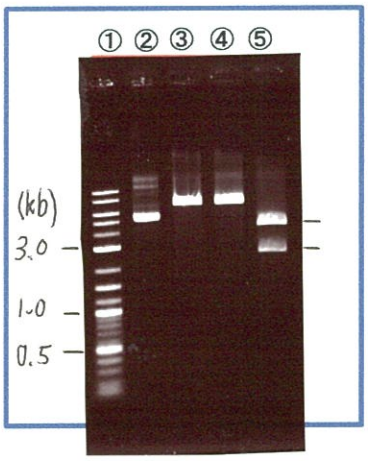
< Size of fragment expected from this clone >

EcoRI	6.9 kb	
XhoI	6.9 kb	
EcoRI + XhoI	4.3kb (Vector)	2.6 kb

Erectrophoresis : 1% agarose gel, 1x TAE Buffer

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





①:Marker
②:uncut
③:EcoRI
④:XhoI
⑤:EcoRI + XhoI

● Adjust plasmid DNA solution to 25 ng/ul

Date : 190529

DNA (88 ng/ul)	62.0	ul
10x TE	21.8	ul
dH_2O	134.4	ul
Total	218.2	ul

● Confirmation of the insertion sequence

Date : 190523

Primer A	Reverse2
Primer B	M13
Primer C	poly(A) region primer #0
List of Sequencing Primers	http://dna.brc.riken.jp/en/GNPclone3en.html

● Shipping

☑ Conc. : 25 ng/μl, Volume : 40 μl

☐ ~~Conc. : ng/μl, Volume : μl~~

確認 19.5.24 笹沼

APPROVED BY :

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

Applied Biosystems

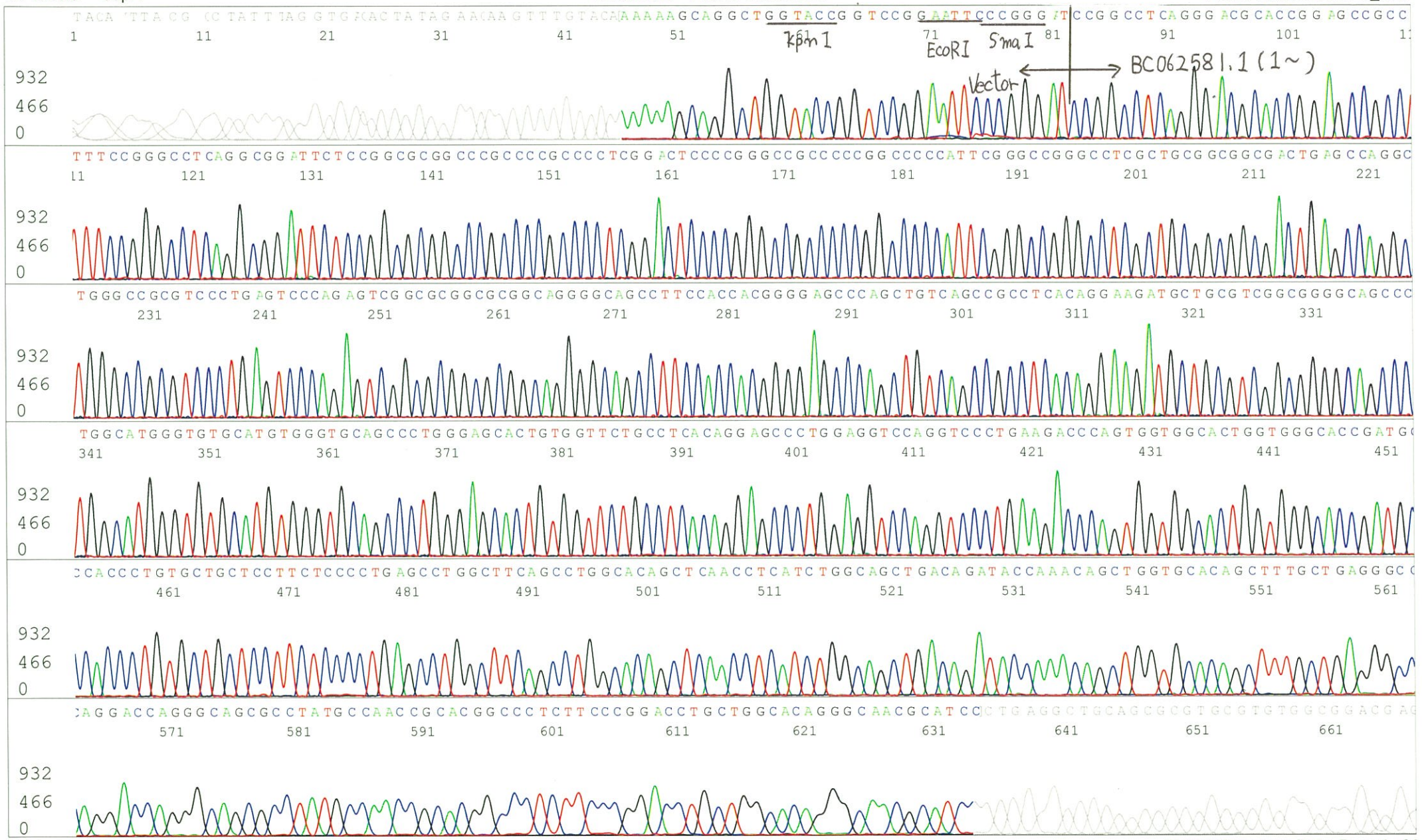
IRAK135G15_A9Ee_1_Reverse2_B01_04_ABI24
IRAK135G15_A9Ee_1_Reverse2
KB_3500_POP7_BDTv3.mob
Pts 1800 to 8706 Pk1 Loc:1768
Version 6.0 HiSQV Bases: 705

Inst Model/Name 3500/3500 Instrument
May 23,2019 09:34AM, JST
May 23,2019 10:01AM, JST
Spacing:10.46 Pts/Panel1350
Plate Name: 20190523_GNP

S/N G:204 A:175 T:112 C:31
KB.bcp
KB 1.4.1.8 Cap:4

Primer A : Reverse2
5' GCGGATAACAATTTCACACAGG 3'

Gray line indicates quality value (QV) < 40.



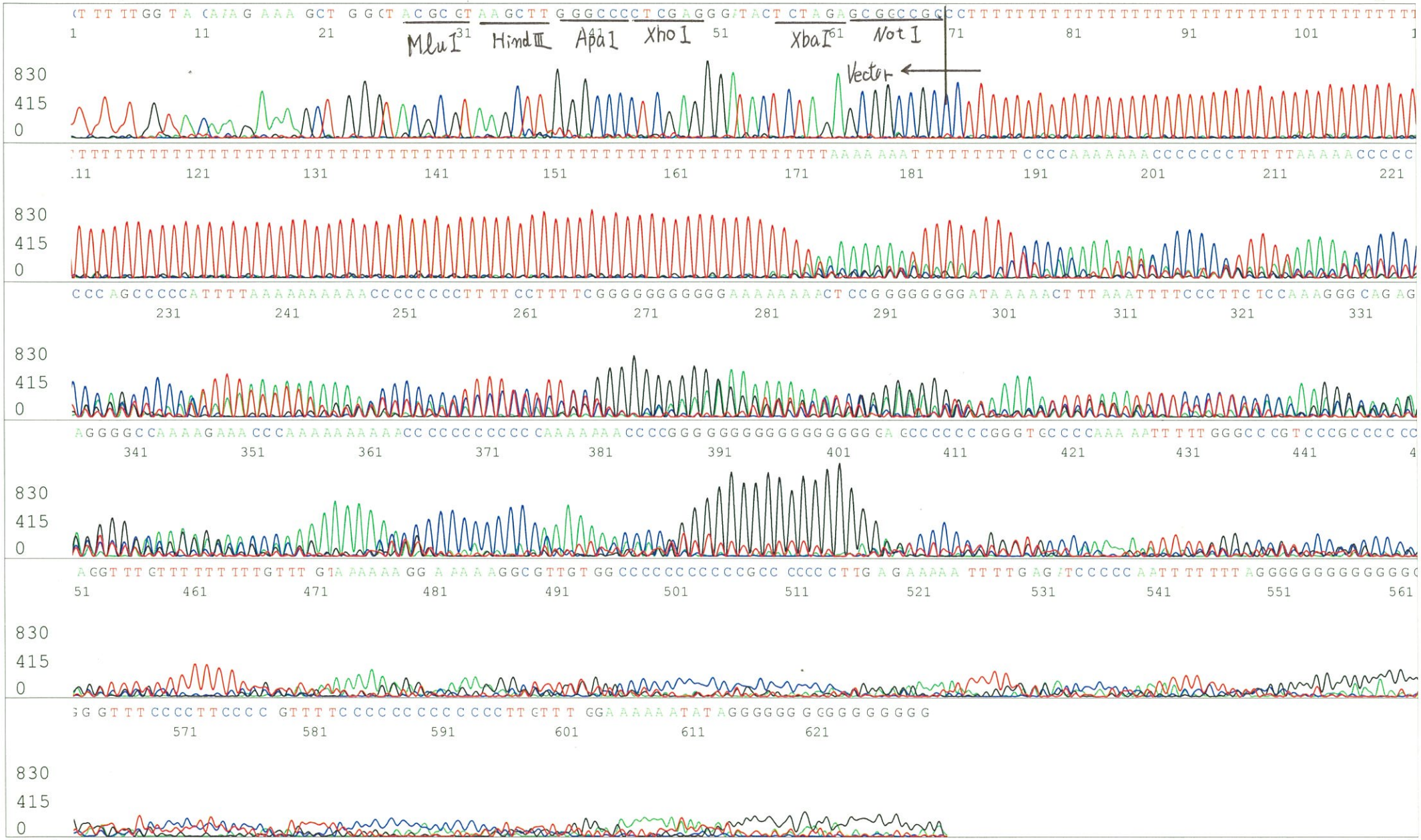
Applied Biosystems

S/N G:7 A:6 T:8 C:9
KB.bcp
KB 1.4.1.8 Cap:4

Primer B : M13
5' GTTTTCCCAGTCACGACGTTGTA 3'

IRAK135G15_A9Ee_1_M13(-40)_D01_04_ABI08
IRAK135G15_A9Ee_1_M13(-40)
KB_3500_POP7_BDTv3.mob
Pts 2075 to 8488 Pk1 Loc:2043
Version 6.0 HiSQV Bases: 250

Inst Model/Name 3500/3500 Instrument
May 22,2019 09:47AM, JST
May 22,2019 10:03AM, JST
Spacing:11.42 Pts/Panel1350
Plate Name: 20190522_GNP

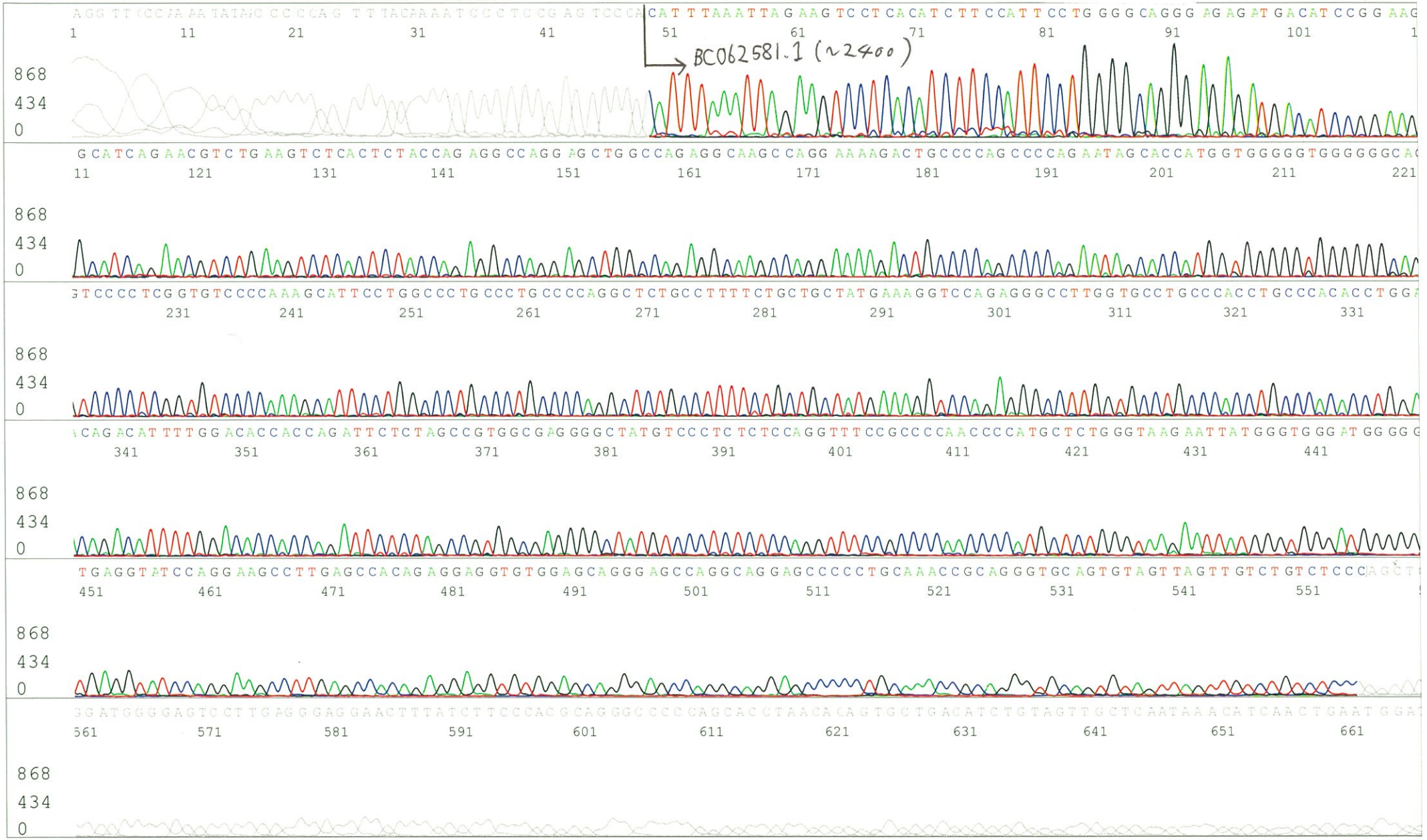


Applied Biosystems

IRAK135G15_A9Ee_1_GNP Mixture_C01_07_ABI24
IRAK135G15_A9Ee_1_GNP Mixture
KB_3500_POP7_BDTv3.mob
Pts 1810 to 8706 Pk1 Loc:1778
Version 6.0 HiSQV Bases: 690

Inst Model/Name 3500/3500 Instrument
May 23,2019 09:34AM, JST
May 23,2019 10:01AM, JST
Spacing:10.36 Pts/Panel1350
Plate Name: 20190523_GNP

S/N G:121 A:148 T:84 C:18
KB.bcp
KB 1.4.1.8 Cap:7

Primer C : Poly(A) region primer #0
5' TTTTTTTTTTTTTTTTTTTTNNN 3'

Gray line indicates quality value (QV) < 40.