Genome Network Project_Full -Length cDNA clone Gene Engineering DivisionRIKEN BioResource Center

RIKEN_Clone_ID : IRAL026H10

Lot#: 7566_A3JI

Vector : pOTB7

Gene	SMO	
Accession No.	BC009989.2	3772 bp	1..3772
	CDS	2364 bp	281..2644

● Plasmid DNA purification

Date : 131021

Culture : LB (25 ug/ml Chloramphenicol) 5 ml -> at 37 deg C over night

Date : 131023

Purification : QIAGEN Miniprep kit -> dH_2O 100 ul

● Digestion by restriction enzyme/Concentrarion calibration

Date : 131023

DNA concentration (O.D.): 252 ng/ul

DNA	0.2 ul
Enzyme (BamHI + BglII)	0.5 + 0.5 ul
Buffer H	1 ul
dH_2O	7.8 ul
Total	10 ul

< Size of fragment expected from IRAL026H10 >

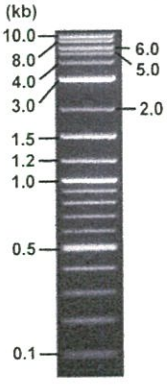
BamHI 2754, 2607, 212 bp

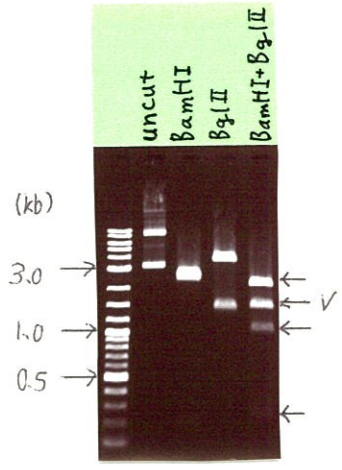
BglII 3966 , 1607 bp

BamHI + BglII 1615bp (Vector) 2351, 1139, 256, 212bp

Erectrophoresis : 1% agarose gel, 1x TAE Buffer

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





● Confirmation of the insertion sequence

Date : 131024

Primer A	pOTB7_F
Primer B	pOTB7_R
Primer C	poly(A) region primer #0
List of Sequencing Primers	http://dna.brc.riken.jp/en/GNPclone3en.html

● Adjustment of DNA concentration

Shipping amount : 40 ul

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

Date : 131024

DNA (252 ng/ul)	82.0 ul
10x TE	82.7 ul
dH_2O	661.9 ul
Total	826.6 ul

確認 13.10.2 笹沼

APPROVED BY :

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

Applied Biosystems

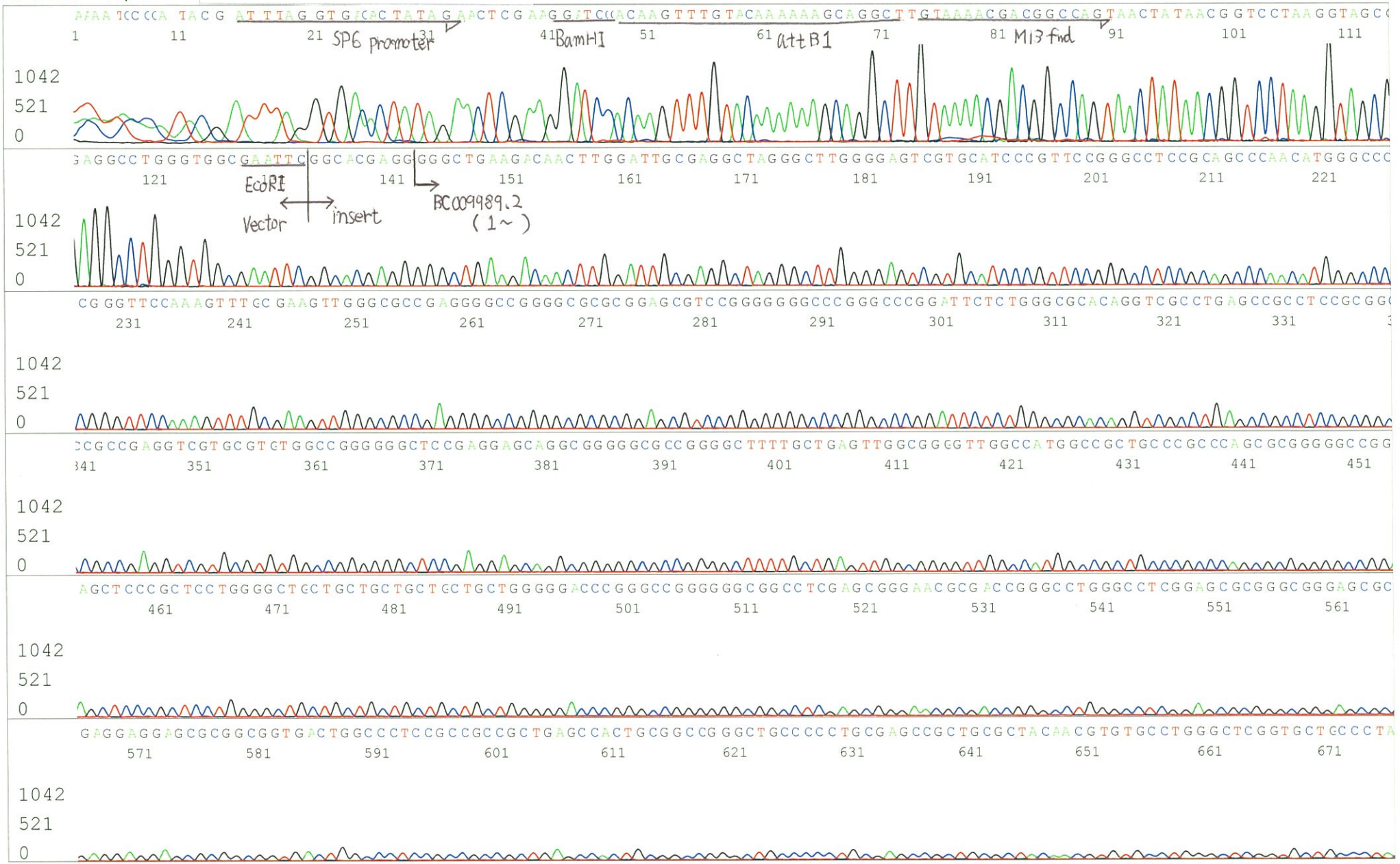
S/N G:117 A:60 T:51 C:50
KB.bcp
KB 1.4.1.8 Cap:12

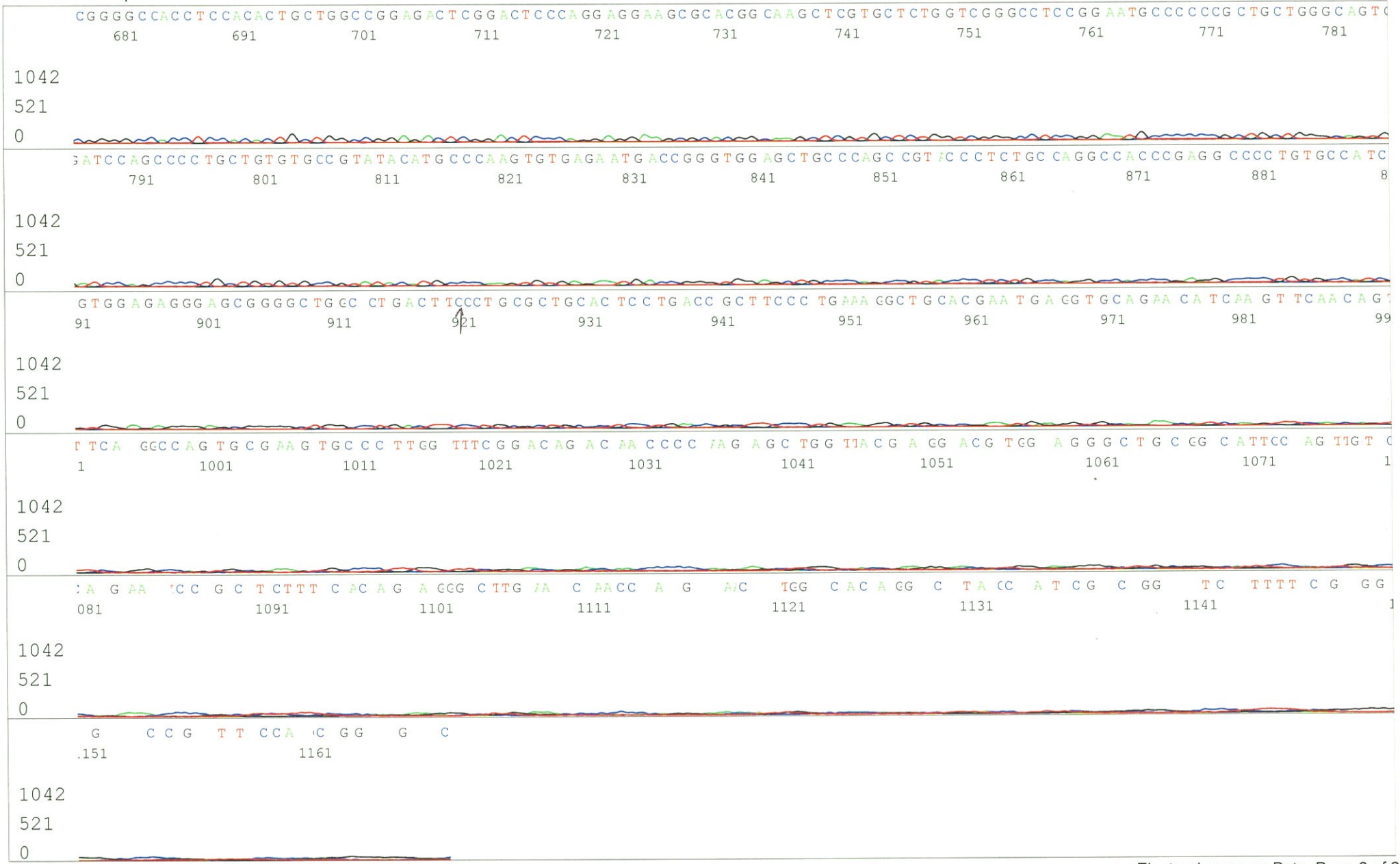
Primer A: pOTB7-F

5' AACGCGGCTACAATTAATACATAACC 3'

IRAL026H10_A3JI_pOTB7_F_D06_12
IRAL026H10_A3JI_pOTB7_F
KB_3500_POP7_BDTv3.mob
Pts 1328 to 13062 Pk1 Loc:1305
Version 5.4 HiSQV Bases: 1012

Inst Model/Name 3500/3500 Instrument
Oct 24,2013 11:10AM, JST
Oct 24,2013 11:36AM, JST
Spacing:11.65 Pts/Panel1350
Plate Name: 20131024_mix





Applied Biosystems

S/N G:107 A:78 T:72 C:51
KB.bcp
KB 1.4.1.8 Cap:15

Primer B: pOTB7-R

5' GTACTGCAGCCGATTCATTAATGC 3'

IRAL026H10_A3JI_pOTB7_R_E06_15
IRAL026H10_A3JI_pOTB7_R
KB_3500_POP7_BDTv3.mob
Pts 1328 to 13042 Pk1 Loc:1305
Version 5.4 HiSQV Bases: 889

Inst Model/Name 3500/3500 Instrument
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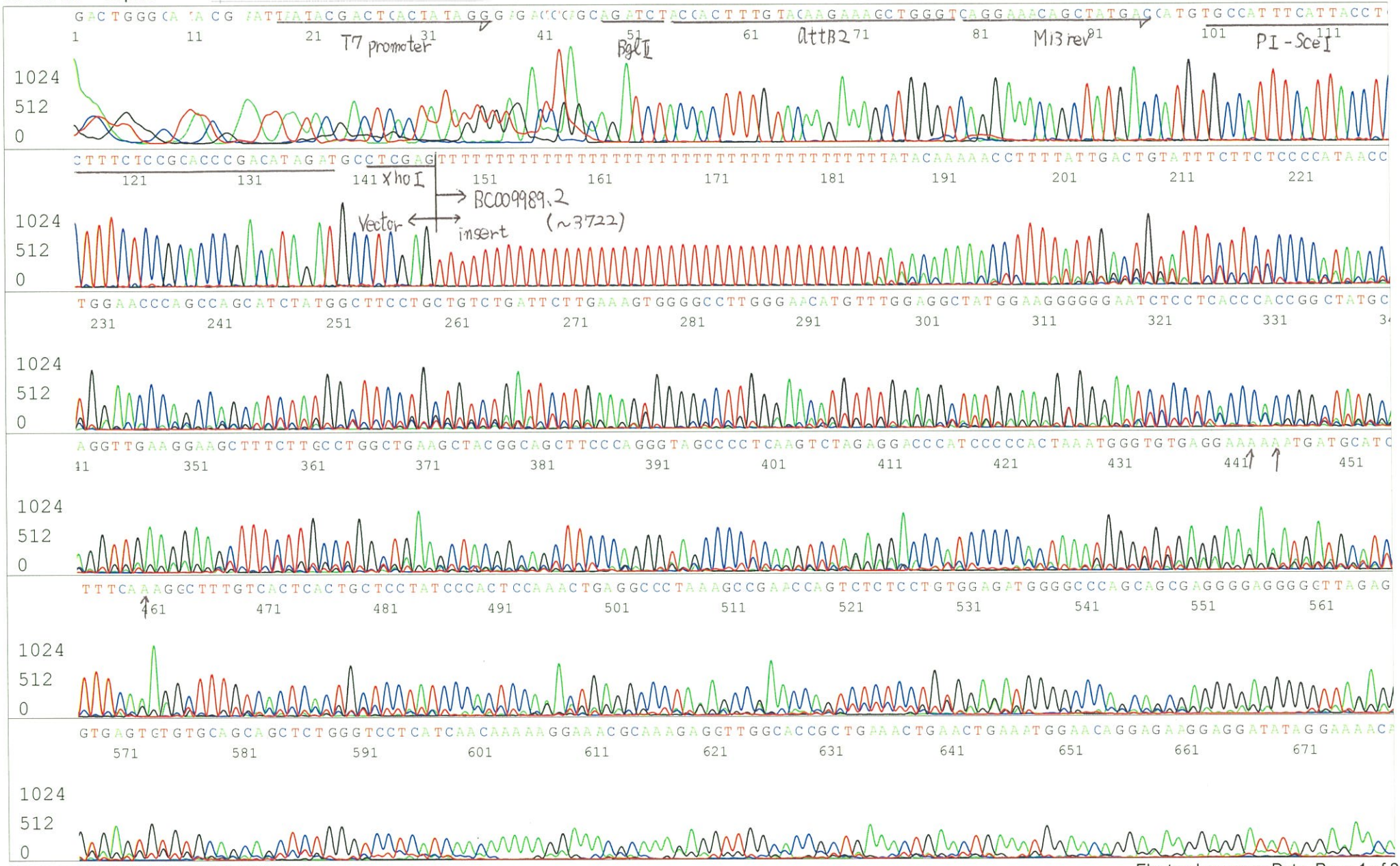
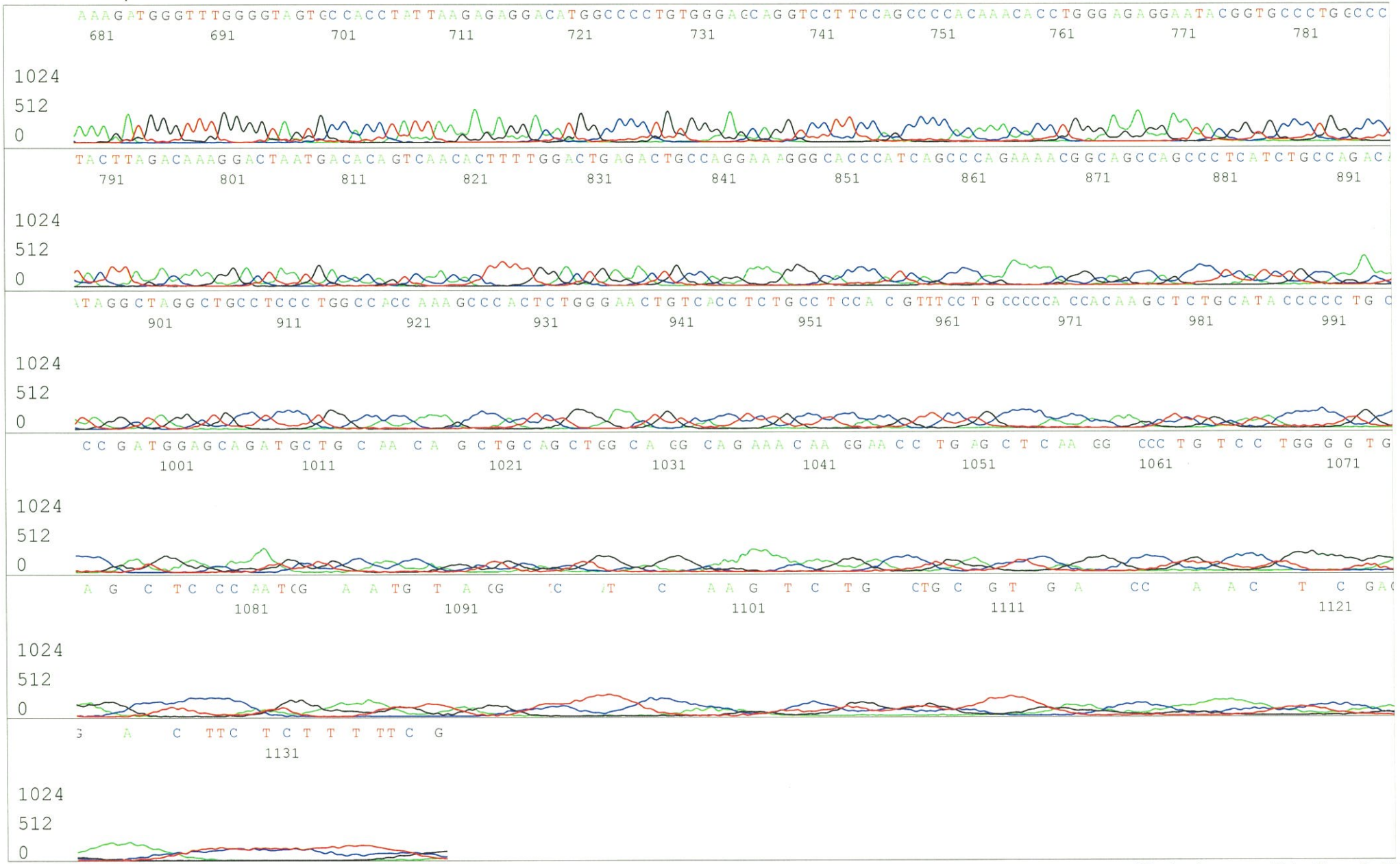


Applied Biosystems

S/N G:107 A:78 T:72 C:51
KB.bcp
KB 1.4.1.8 Cap:15

IRAL026H10_A3JI_pOTB7_R_E06_15
IRAL026H10_A3JI_pOTB7_R
KB_3500_POP7_BDTv3.mob
Pts 1328 to 13042 Pk1 Loc:1305
Version 5.4 HiSQV Bases: 889

Inst Model/Name 3500/3500 Instrument
Oct 24,2013 11:10AM, JST
Oct 24,2013 11:36AM, JST
Spacing:11.74 Pts/Panel1350
Plate Name: 20131024_mix



Applied Biosystems

S/N G:91 A:56 T:50 C:36
KB.bcp
KB 1.4.1.8 Cap:18

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

IRAL026H10_A3JI_GNPmixture_F06_18
IRAL026H10_A3JI_GNPmixture
KB_3500_POP7_BDTv3.mob
Pts 1319 to 13096 Pk1 Loc:1296
Version 5.4 HiSQV Bases: 1000

Inst Model/Name 3500/3500 Instrument
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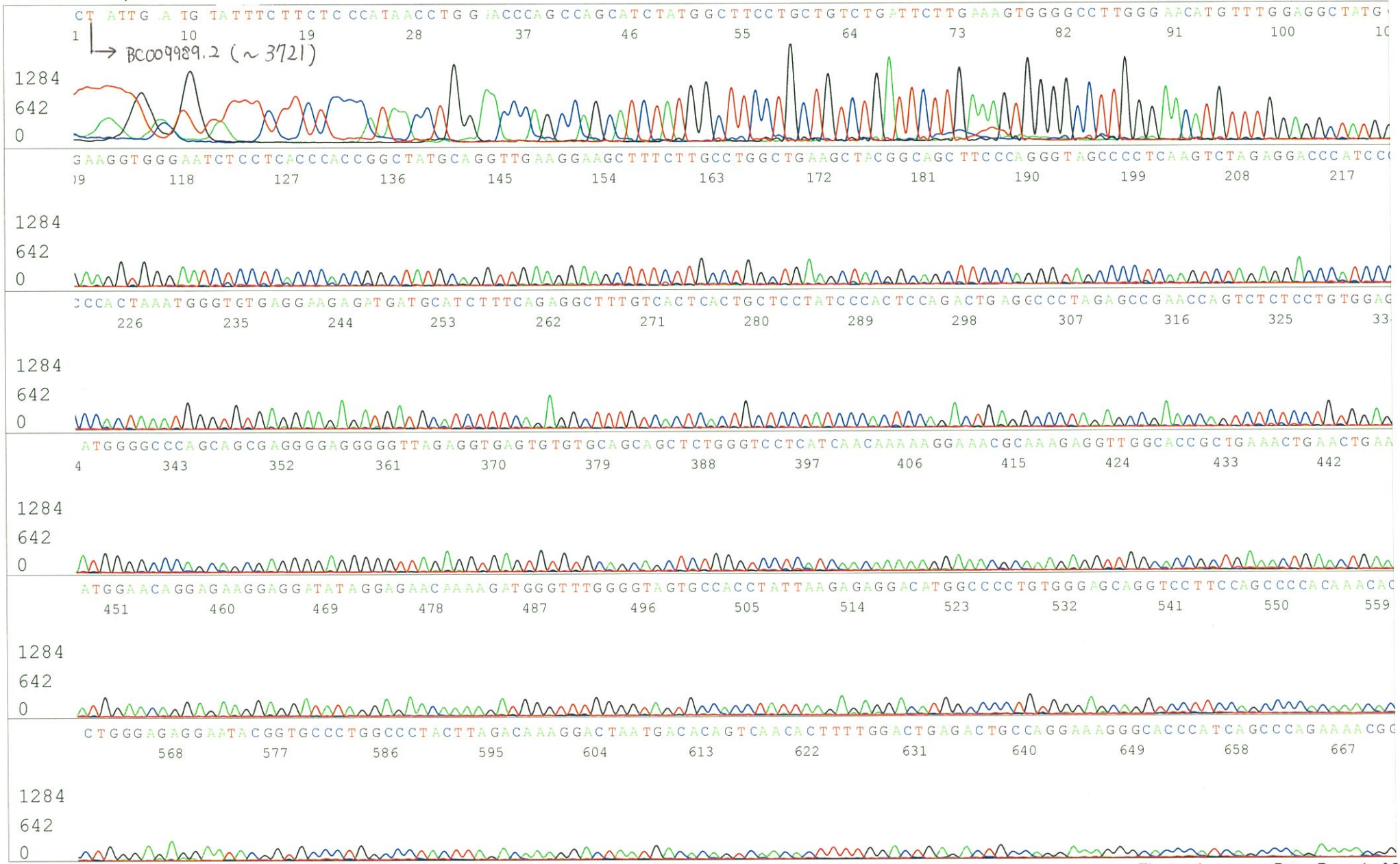

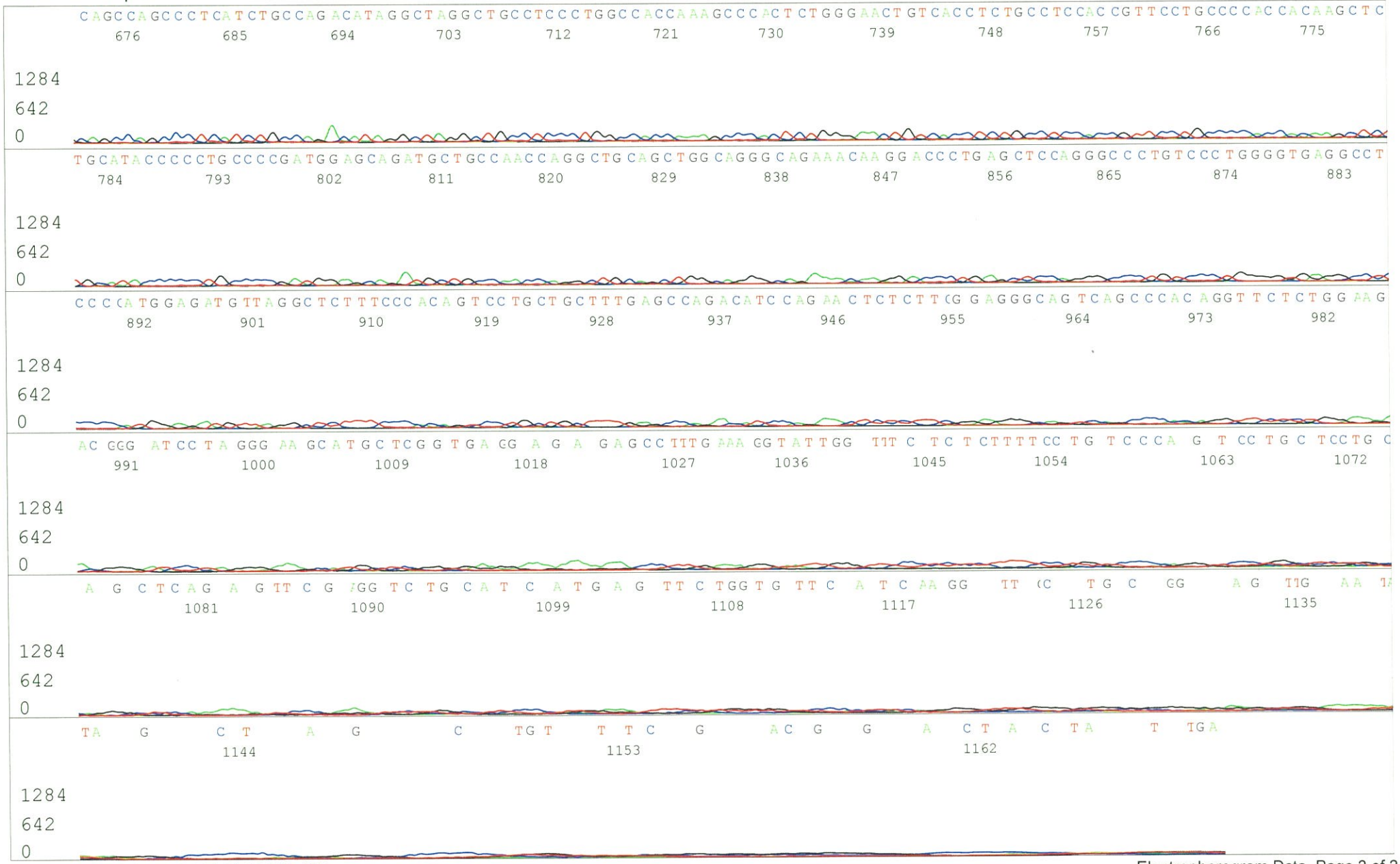
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784 793 802 811 820 829 838 847 856 865 874 883
892 901 910 919 928 937 946 955 964 973 982
991 1000 1009 1018 1027 1036 1045 1054 1063 1072
1081 1090 1099 1108 1117 1126 1135
1144 1153 1162
1284
642
0