

NIH-0953 Genotyping Strategies

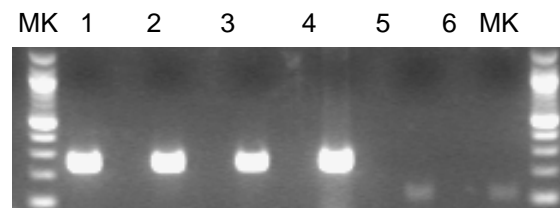
Reaction Components	Vol (ul)
5X GoTaq Buffer	10
25mM MgCl ₂	3.5
10mM dNTPs	1
Primer 20 uM	1
Primer 20 uM	1
5 U/ul Taq polymerase	0.5
Water	28
Total mix volume	45
Tail lysate (1:20 dilution)	5
Total reaction volume	50

Step	Temp	Time	Note
1	94C	15"	
2	65C	30"	Decrease 1C/cycle
3	72C	40"	Go to 1, 10 cycles
4	94C	15"	
5	55C	30"	
6	72C	40"	Go to 4, 30 cycles

Primer Sequences (5' to 3')	
Mutant PCR: Primer LTR-2 and Primer 0953-3', 242 bp	
Recommended Wt PCR: Primer 0953-wt and Primer 0953-3', 220 bp	
Primer LTR-2	AAATGGCGTTACTTAAGCTAGCTTGC
Primer 0953-wt	AGTTTGATTCCCAGAACTCACAGG
Primer 0953-3'	GAGGCTAACTAGACTGTGGGCTTCC

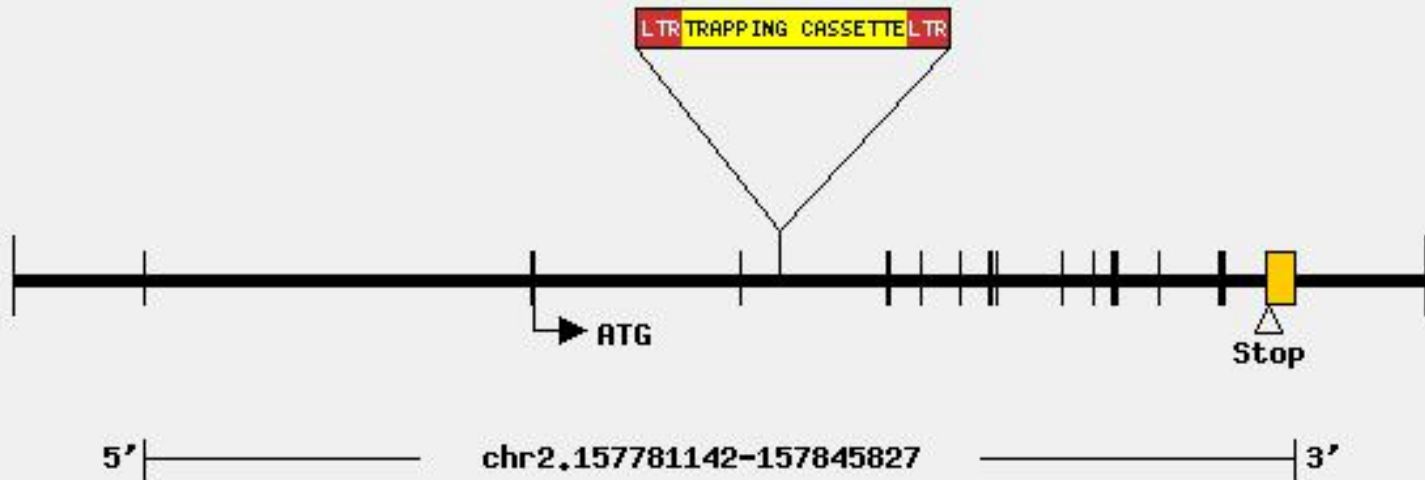
Well	Sample	Genotype
1	219	het
2	220	het
3	223	het
5	ES DNA	het
6	wt lysate	wt
7	water	no amp

Mutant PCR

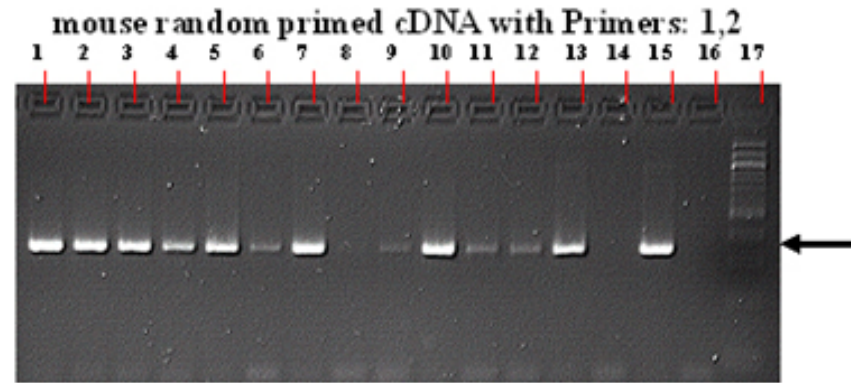


QC Image

Accession: NM_013865



RT-PCR WT Expression



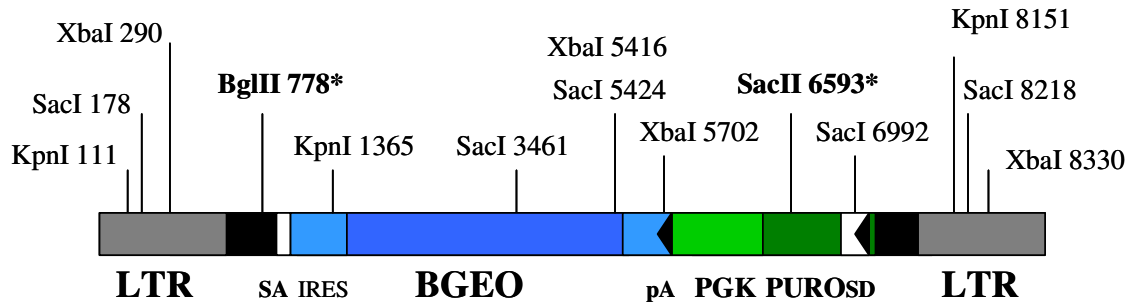
10/22/2002

Note: Expected band size denoted by arrow adjacent to 100bp ladder/marker.

Mouse cDNA Tissues

- 1) Brain
- 2) Thymus
- 3) Spleen
- 4) Lung
- 5) Kidney
- 6) Liver
- 7) Testis
- 8) Bone
- 9) Small Intestine & Colon
- 10) Skin Fibroblast
- 11) Heart
- 12) Adipose
- 13) Whole Blood
- 14) (-) Control
- 15) (+) Control- ES cell cDNA
- 16) (+) Control- Genomic/Lex1 DNA
- 17) 100 bp ladder/marker

VICTR 22 Omnibank Vector



Total Size: 8630 nucleotides

Non-Cutters:

* Unique sites

Location of components in VICTR 22:

LTR (viral long terminal repeat): 1-590, 8014-8630

SA (splice acceptor): 783-912

IRES: 909-1493

Bgeo: 1522-5416

pA: 5421-5706

lox p sites: 5726-5796, 7045-7121

PGK promoter: 5840-6356

PURO SD: 6379-7026

>VICTR 22

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