

NIH-0976 Genotyping Strategies

Reaction Components	Vol (ul)
10X Sigma Buffer	5
25mM MgCl ₂	3.5
10mM dNTPs	2
Primer 20 uM	1.5
Primer 20 uM	1.5
5 U/ul Taq polymerase	0.5
Water	31
Total mix volume	45
Tail lysate (1:20 dilution)	5
Total reaction volume	50

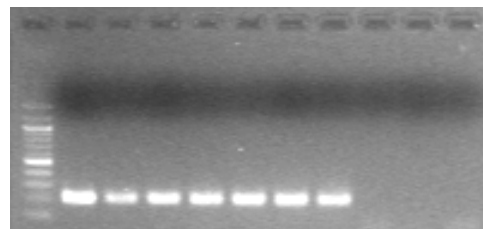
Step	Temp	Time	Note
1	94C	15"	
2	65C	30"	Decrease 1C/cycle
3	72C	40"	Go to 1, 10 cycles
4	94C	15"	
5	55C	30"	
6	72C	40"	Go to 4, 30 cycles

Primer Sequences (5' to 3'):	
Mutant PCR: Primer LTR-2 and Primer 0976-3', 204 bp	
Recommended Wt PCR: Primer 0976-5' and Primer 0976-3', 202 bp	
Primer LTR-2	AAATGGCGTTACTTAAGCTAGCTTGC
Primer 0976-3'	TGGCCTTAATCCATATGCCCTCTC
Primer 0976-5'	TCATGGGTAGCTAGTAAAAGGATGG

Well	Sample	Genotype
1	92	het
2	93	het
3	94	het
4	102	het
5	104	het
6	109	het
7	het	het
8	water	no amp
9	wt lysate	wt
10	water	no amp

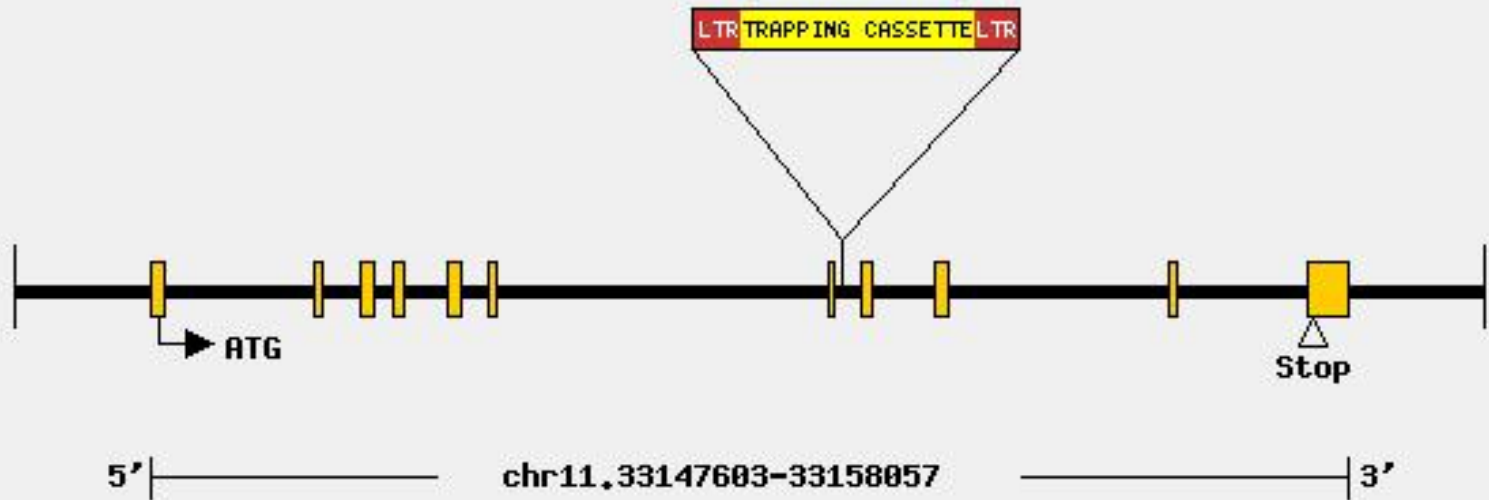
Mutant PCR

MK 1 2 3 4 5 6 7 8 9 10

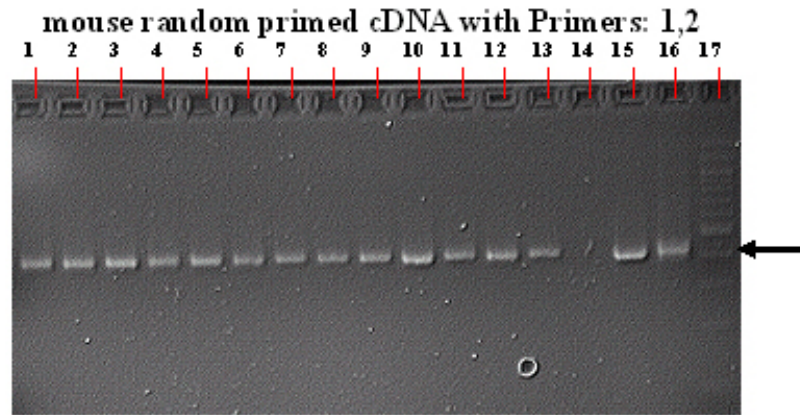


QC Image

Accession: NM_008722



RT-PCR WT Expression



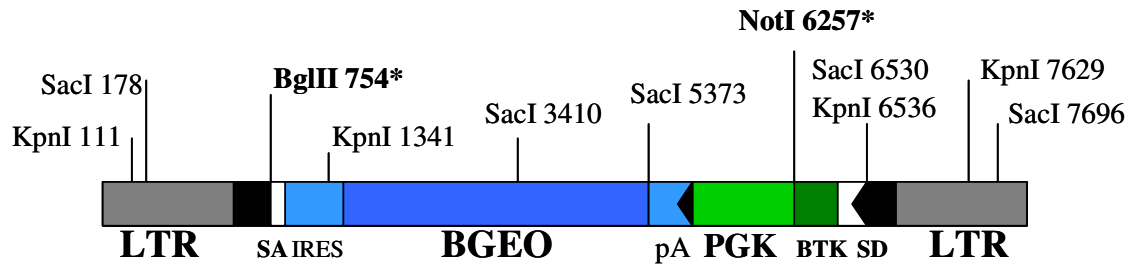
03/25/2002

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) Tail
- 14) (-) Control
- 15) (+) Control- ES cell cDNA
- 16) (+) Control- Genomic/Lex1 DNA
- 17) 100 bp ladder/marker

VICTR 37 Omnibank Vector



Total Size: 8108 nucleotides

Non-Cutters: SacII, DraI, XhoI

* Unique sites

Location of components in VICTR 37:

LTR (viral long terminal repeat): 1-590, 7519-8108

SA (splice acceptor): 759-888

IRES: 885-1469

Bgeo: 1471-5365

pA: 5370-5655

frt sites: 5670-5717, 6546-6593

PGK promoter: 5738-6254

BTK exon: 6289-6514

>VICTR 37

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