

NIH-0411 Genotyping Strategies

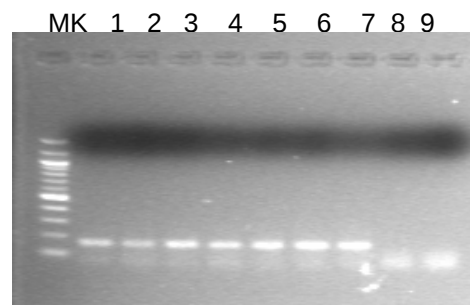
Reaction Components	Vol (ul)
10X Sigma Buffer	5
25mM MgCl ₂	3.5
10mM dNTPs	2
Primer 20 uM	1.5
Primer 20 uM	1.5
5 U/ul Taq polymerase	0.5
Water	31
Total mix volume	45
Tail lysate (1:20 dilution)	5
Total reaction volume	50

Step	Temp	Time	Note
1	96	17	
2	61	25	
3	72	25	Go to 1, 36 cycles

Primer Sequences (5' to 3'):	
Mutant PCR: Primer LTR-2 and Primer 0411-3', 165 bp	
Recommended Wt PCR: Primer 0411-5' and Primer 0411-3', 214 bp	
Primer LTR-2	AAATGGCGTTACTTAAGCTAGCTTGC
Primer 0411-3'	AATACTGGTCTTGGAGGCCTTGGA
Primer 0411-5'	AAGCACCCCTGAAAGCTGTACT

Well	Sample	Genotype
1	72	het
2	75	het
3	77	het
4	83	het
5	86	het
6	93	het
7	96	het
8	wt lysate	wt
9	water	no amp

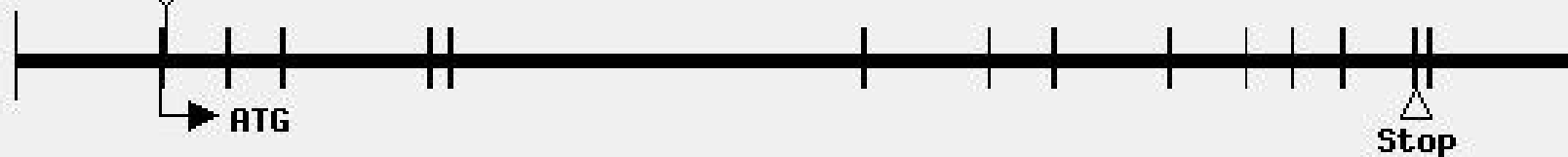
Mutant PCR



QC Image

Accession: AF071068

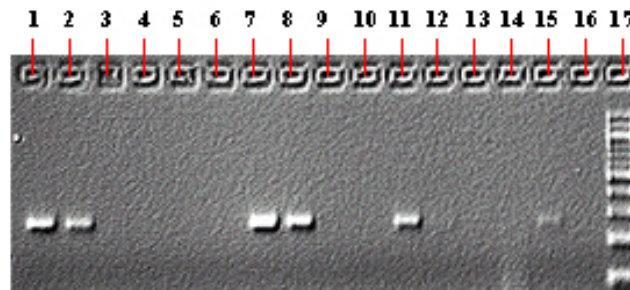
LTR TRAPPING CASSETTE LTR



5' |————— chr11.11735216-11807742 —————| 3'

RT-PCR WT Expression

mouse random primed cDNA with Primers: 1,2



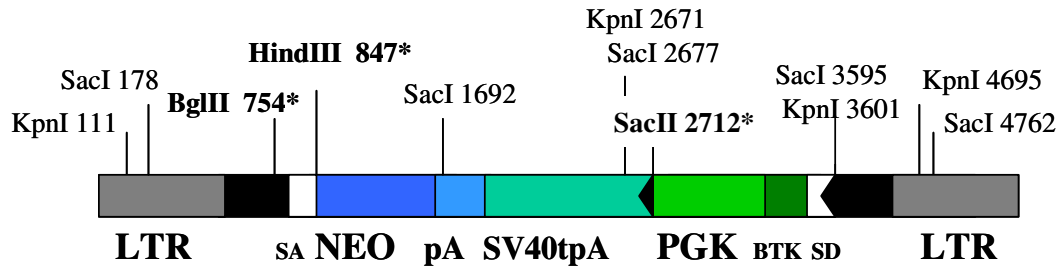
02/12/2004

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

Mouse cDNA Tissues

- 1) Brain
- 2) Spinal Cord
- 3) Eye
- 4) Thymus
- 5) Spleen
- 6) Lung
- 7) Kidney
- 8) Liver
- 9) Skeletal Muscle
- 10) Bone
- 11) Stomach, Small Intestine & Colon
- 12) Heart
- 13) Adipose
- 14) (-) Control
- 15) (+) Control- ES cell cDNA
- 16) (+) Control- Genomic/Lex1 DNA
- 17) 100 bp ladder/marker

VICTR 48 Omnibank Vector



Total Size: 5174 nucleotides

Non-Cutters: ApaI, XhoI, XmnI

* Unique sites

Location of components in VICTR 48:

LTR (viral long terminal repeat): 1-590, 4585-5174

SA (splice acceptor): 755-847

NEO: 867-1684

pA: 1688-1874

pA (SV40 poly adenylation sequence): 1875-2691

frt sites: 2733-2780, 3613-3661

PGK promoter: 2805-3321

BTK exon: 3356-3580

>VICTR 48

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TGAAAGACCCCGCTGACGGGTAGTCAATCACTCAGAGGAGACCCTCCCAAG
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