



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ675	Date of Submission:	11.19.04
Lexicon Contract Name:	DNA103	Mutation Type:	x Standard Knock out
LexVision Name:	SEC454N1		
Reference accessions:	NM_028880	Is this gene X-linked?	no

Required Materials:

- x pKOS clone DNA(s) pkos 3
- x Target Vector DNA TV 3 Bgeo/PURO
- x Targeted ES Cell DNA 1E8
- x Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' Internal</u>	<u>3' External</u>
Name of Probe:	28/29	30/31
Restriction Enzyme for Genomic Digest:	BamHI	SpeI
Predicted Wild-type Band (kb):	25	11.5
Predicted Mutant Band (kb):	5.3	7.2
Probe Size:	232 bp	348 bp

PCR Strategies:*For standard knockouts, give wildtype and mutant-specific strategies**For conditionals, give 5' loxP and cre-excision strategies*

Wild type-specific (absent in targeted allele)		Mutation-specific product (absent in wt)	
5' Primer Name:	SEC454N1-3	5' Primer Name:	SEC454N1-32
3' Primer Name:	SEC454N1-6	3' Primer Name:	IRES GT
Predicted Wild-type Band (bp):	409	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	589

Primer sequences:**Southern probes**

SEC454N1-28	5' – CCAGGCCTCTACTCTTAACCTC
SEC454N1-29	5' – TCCGGGGTAATGACAGATTTAG
SEC454N1-30	5' – ATGAATTCCGCTAGCATATAAC
SEC454N1-31	5' – GCTCTCCAGGCTGCAAAGCGTT

PCR Genotyping

SEC454N1-3	5' – AGCAGCTGAATGGAGGTTGTC
SEC454N1-6	5' – AGTGTAGACAGAGGCCGAGTAG
SEC454N1-32	5' – TGGAAAAGGGTGGGTAAGACACTTTT
IRES GT	5' – CCCTAGGAATGCTCGTCAAGA

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Selection Cassette: Bgeo/PGK PURO

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Targeted Locus:

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[illegible]

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CTTTCTGAGCTGGTGGCGAATTCTGGTTTTGTTTCTAGCTCTGATTTAGTAGCCAAGTGTAGAGTGTAACGCAGAAAG
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CTAAGATAATTTTTCTTCTACAGGTATACACATTAATATTAGCAACACAGACAATCTATATTATCAGAGGAAACACTAA
CCATTTGAGAATTGAGGACAGCTTTGATGTTCAATGTTTTGAGTCTTTGTTGACTTTTTGTTGCTGCTGCTGCTTATTTTC

[illegible]