



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ598	Date of Submission:	6-20-05
Lexicon Contract Name:	DNA454	Mutation Type:	X Standard Knock out
LexVision Name:	MEM802N1		☐ Conditional
Reference accessions:	AK016865	Is this gene X-linked?	No

Required Materials: X pKOS clone DNA(s) __pKOS55, pKOS74_____
 X Target Vector DNA __pKOS55TV_____
 X Targeted ES Cell DNA __2H2_____
 X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	33+34	35+36
Restriction Enzyme for Genomic Digest:	EcoRI	NheI
Predicted Wild-type Band (kb):	6.6 kb	8.4 kb
Predicted Mutant Band (kb):	3.6 kb	12.4 kb
Probe Size:	200 bp	313 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:	DNA454-20	5' Primer Name:	Neo3a
3' Primer Name:	DNA454-21	3' Primer Name:	DNA454-3'
Predicted Wild-type Band (bp):	265 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	579 bp

5' loxP strategy		Distinguish Cre-excised and wt	
5' Primer Name:		5' Primer Name:	
3' Primer Name:		3' Primer Name:	
Predicted Wild-type Band (bp):		Predicted Wild-type Band (bp):	
Predicted mutant band (bp)		Predicted mutant band (bp)	

Primer sequences:**Southern probes**

DNA454-33 5' – GGGCATCTTGCTCCTTATCAG
 DNA454-34 5' – TTATCCAGGTGGGCGATGT
 DNA454-35 5' – TTCTGACAAAGGGCTAGATAA
 DNA454-36 5' – CCTCCTCGAACTAGTGACC

PCR Genotyping

DNA454-20 5' – GCTAAACTTTGGGACTGGTAG
 DNA454-21 5' – CCAAATGTTCAAGTGACGAA
 DNA454-3' 5' – GGCTCTATAGGGCTAACCAAGCAC
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

TGGGAACGCCCTACCACTGTTTAACATGTCAGCACTGAGCAAGAAGTGTCTGTTCAACCTGATGAAAGAGGTAAGGATG
CAGAGTAGACACTAGCCAAAGGCAGTAACGTTCCAACCTATAACTCCCTGAATGAGAGCCTCACGGGTGGGAACTCAG
CCCCTGGAGACCCCAGCTATCATGTAACCTGTTTGGTTTCTTAGAGCTAGCAAGATTTCTGTCTGATGAATCTCTCCAT
GGGGGACATACAATGAGAGATTCAAGACTGGGAAAGCGCCTGAGAAATAAGGGCACTGATGTGAAATCATGAACATG
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CACCTTTGGGATCTTCATCCCCTTTCTGCCATATAGCTGGAAATATGCTGCCAATATTGTATCCTTTGTAC

Genomic Locus: *(The deleted sequence represents nt11423-13146-in the sequence below. KOS55 used to generate the TV represents nt 9144-18308 in the sequence below.)*

[illegible]

CGAGGTGACCAATCATGCTGTTGAGTCTCAGGTTACAGGGAATATTGTAAGGCCTCAGTATGGGAACATGAGCAGAAG
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Selection Cassette:

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[illegible]

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GAGAGGATC