



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ9574	Date of Submission:	1-19-05
Lexicon Contract Name:	DNA494	Mutation Type:	X Standard Knock out
LexVision Name:	SEC264N1		☐ Conditional
Reference accessions:	AK003305	Is this gene X-linked?	No

Required Materials: ☐ pKOS clone DNA(s) __pKOS40_____
☐ Target Vector DNA __KOS40TV_____
☐ Targeted ES Cell DNA __1B6_____
☐ Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	20+21	24+25
Restriction Enzyme for Genomic Digest:	HpaI	EcoRI
Predicted Wild-type Band (kb):	17.1 kb	8.5 kb
Predicted Mutant Band (kb):	11.8 kb	7.1 kb
Probe Size:	608 bp	606 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:	DNA494-4	5' Primer Name:	Neo3A
3' Primer Name:	DNA494-5	3' Primer Name:	DNA494-19
Predicted Wild-type Band (bp):	519 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	655 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**

DNA494-21 5' – GGTCTGTAGCAAGCCTTCC
DNA494-20 5' – GTGTCCTTCCTGCCAAATCC
DNA494-24 5' – CATCTGTACAGCCCTTGTCC
DNA494-25 5' – CCAGAAGCACAAACAGCCAG

PCR Genotyping

DNA494-4 5' – GGTGATGCGGACCCGGAG
DNA494-5 5' – GTGTCTGATTGCAGCTCTCC
DNA494-19 5' – GGAGGCCTACACTAGAAGG
Neo3A 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

CAGTAGACAGAACAGCCTGAGCCAGGATGAAGGCTCTCAGGGCTGTCCTCCTGATCTTGCTACTAAGTGACAGCCAG
GGTGCGGGTCGCCTCAAGAAGGGATGGCCAGTCCTTACCCATGAGTACCTCCAGGCACCCCTTATCCTAGGGACAAG
GAATAGACCTGAGGAGTAAGATAGAAGGCCTGGGGCTCTTCTCTTCTGACGGCTTGGCTGCTTCGGCACTGCAGGGGTG
GGGGTGGGGGGAATTCTGTCTCCTTCCATAAAATGGAAAGGGTATGGGGAAGAGGGCATGTTAGGGTCAGGACTTAGC
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CAGGACGCCCTGGCCTCTCCTTCTGACAACCAGCTTCAGAGAATAAACTTGAATGTCGTTTGCCATCTAGCTCGTTGC
AGCTCC

Genomic Locus: *(The deleted sequence represents nt17415-18991-in the sequence below. KOS40 used to generate the TV represents nt 11892-21989 in the sequence below.)*

[illegible]

AAGGCTGGGTTTTCTGCAAGGGGTGGCAGATGCTATCAGAAAGGCTCTATACCCCCCTGGTGGCTTCTTGGGGTTTTTG
CTGGCACGCTGGGACAGGTGGTCAGCAGAGATTCTTTTCTGTGAGCAAGGCTCGCTGGCTTGCTGCCAGACTCTGCCC
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Selection Cassette:

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