



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ293	Date of Submission:	11.2.04
Lexicon Contract Name:	DNA334	Mutation Type:	x Standard Knock out
LexVision Name:	MSC336N1		☐ Conditional
Reference accessions:	ENSMUST00000044248, NM_172874	Is this gene X-linked?	No

Required Materials: x pKOS clone DNA(s) __KOS 68_____
x Target Vector DNA __TV68-2_____
x Targeted ES Cell DNA __1C1_____
x Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' Internal</u>	<u>3' External</u>
Name of Probe:	1+2	19+20
Restriction Enzyme for Genomic Digest:	BamHI	HindIII
Predicted Wild-type Band (kb):	5.1 kb	12.7 kb
Predicted Mutant Band (kb):	4.4 kb	10.7 kb
Probe Size:	190 bp	395 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:	DNA334-27	5' Primer Name:	Neo3a
3' Primer Name:	DNA334-28	3' Primer Name:	DNA334-10
Predicted Wild-type Band (bp):	494 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	342 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**

DNA334-1 5' – TCAGCTTGGCTCCTTAACAG
 DNA334-2 5' – CCTCACAAACACAAGGTACTCA
 DNA334-19 5' – CTCAGAGCTGGATGAATC
 DNA334-20 5' – GACAGGACGACATGATCTG

PCR Genotyping

DNA334-27 5' – CTGAACCTGCAGAACAAC
 DNA334-28 5' – ACTGAGTCATTTGTGGTAC
 DNA334-10 5' – GAAACTGAGATGCTAGGCAG
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

GGCTGGCAGCAGGGGCTGCCACTCCTACTGCTGGTGGCTTCAGTCTTCTGCTGGGCCCTGTGCTGCTGTGAGGGCACCTGTGTTTGGCCGAAGTGACAC
CCCCACCTTGAGCCCCGAGGAGAATGAATTTGTGGAGGAAGAGAATCAGCCAGTGTGTTTCTGAGCTCCGAGGAGCCAGCTGGCCCCAGCCACT
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AACGAGACCTTCTGGTGGGTCCCTGTGTCAGTGCATCCTCCCCATCACACCCGGCAGCGTGCAACACCGGCGGT

Genomic Locus: (The deleted sequence represents nt 7645-12931 in the sequence below. KOS68 used to generate the TV represents nt 4384-14783 in the sequence below.)

AGCTTTCTTACTTGAGAAGTAGTAGGCAAAACAGATCAAAAAGCTCAAGATATGGGCAACTTGATGAGGTCTGTCTCAAAATTTAAATAAATAACAA
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[illegible]

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Selection Cassette: LacZ/Neo

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[illegible]

[illegible]

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