



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ6368	Date of Submission:	September 29, 2003
Lexicon Contract Name:	DNA099	Mutation Type:	X Standard Knock out
LexVision Name:	MEM481N1		☐ Conditional
Reference accessions:	NM_139299	Is this gene X-linked?	No

Required Materials: ☐ pKOS clone DNA(s) pKOS-66, & pKOS-74 (pKOS-53 N/A, but pKOS-74 very similar)
☐ Target Vector DNA pTV53
☐ Targeted ES Cell DNA 2G7
☐ Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	13+14	17+18
Restriction Enzyme for Genomic Digest:	EcoRV	SacI
Predicted Wild-type Band (kb):	24.8	11.8
Predicted Mutant Band (kb):	19.5	13.3
Probe Size:	330 bp	470 bp

PCR Strategies:

For standard knockouts, give wildtype and mutant-specific strategies

Wild type-specific (absent in targeted allele)		Mutation-specific product (absent in wt)	
5' Primer Name:	5	5' Primer Name:	Neo3a
3' Primer Name:	7	3' Primer Name:	7
Predicted Wild-type Band (bp):	274	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	348

Primer sequences:**Southern probes**

13 5' – GTGAAGGAGGTGCCTTCAGTTAG
14 5' – CCCAGCCTAAGTTTAAAGACAAG
17 5' – GATTCACCCCTGAGTCAAGTTAC
18 5' – CTTGGCTTATGGTTAGAACTG

PCR Genotyping

5 5' – GGGTGTGAACGCTGGAATAATG
7 5' – GATCATCTCTCCAAATTTACATG
Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

GTGAACGCTGGAATAATGTGGACCTTGGCACTGTGGGCATTCTCTTTCCTCTGCAAATTCAGCCTGGCAGGTAGGTTAGC

Genomic Locus: (the deleted sequence represents nt 8093-8172 in the sequence below)

[illegible]

[illegible]

ACGATCAGTCAGTTTATACTCTGAGGGCTAAGAAGGGTCACATGAGCAAAAAGTGTCCAATCACAAGGAGAAGCCATATGTGGCAAAAACATGCTTTTC
CGTAGAGGCACATAAGACTTCATCAATAACCAGTTAAAAATGAATAATCGGAAGTAAACTCAGCCCCATCCACTACACATTCCAAGAATGGTTTCATGC
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CCCAGGCAAGTGCCAAAGCCTTA

Selection Cassette:

ES cell selection cassette

The IRES-lacZ/MC1neo was inserted to replace nt 8093-8172 of the contig above.

GGCCGCTCTAGAGGCCATAGCGGCCATTTAAATGGCGCGCCGGATCCCGGGCCGCTCTAGCTAGACTAGTCTAGCTAGAGGAATTCGGCCCTCTCCCT
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Targeted Locus:

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[illegible]

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