Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ1821	**Date of Submission:**	11/05/03
Lexicon Contract Name:	DNA256	**Mutation Type:**	X Standard Knock out
LexVision Name:	MSC334N1		☐ Conditional
Reference accessions:	NM_026748	**Is this gene X-linked?**	No

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Required Materials:

X pKOS clone DNA(s) __KOS5, KOS46, KOS51, KOS54_________
X Target Vector DNA __KOS5-FTV____________
X Targeted ES Cell DNA __3F2____________
X Genomic Map

Southern Blot Analysis:

External/Internal Probe Strategies

	5' External	**3' External**
Name of Probe:	**17+18**	**21+22**
Restriction Enzyme for Genomic Digest:	**Bgl II**	**Bgl II**
Predicted Wild-type Band (kb):	**7.2 kb**	**4.8 kb**
Predicted Mutant Band (kb):	**14.9 kb**	**14.9 kb**
Probe Size:	**279 bp**	**414 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:	DNA256-24	5' Primer Name:	Neo3a
3' Primer Name:	DNA256-11	3' Primer Name:	DNA256-11
Predicted Wild-type Band (bp):	200	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	245

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

DNA256-17	5' – TCTGGCCTAGGTACGATGAAC
DNA256-18	5' – ACCACTGAGCACGCAAGCAC
DNA256-21	5' – CGCTGGTACAGTGAGGCTAGG
DNA256-22	5' – GCTCTGCCAATATGTAGTCCC

PCR Genotyping

DNA256-11	5' – ACTTGACTGGCATGTACGAG
DNA256-24	5' – CTGCTGCTAGGCCCGAAAG
Neo3a	5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

TACCCACCGTGCACGCTGACGGACGAGGAGACGAGGACGGAGATGATCAACCGGGAACTGCAGATCTCTCAGCGGGA
GAAGCAGGAGATCCTGGCCTTTGAGGGTCACCTGGCCGCCGCCTCCACCAAGCAGACCATCACCGAGAGCAGCAGCCT
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Genomic Locus: *(the deleted sequence represents nt 15,500-16,789 in the sequence below)*

GAACATATTATCATTTAAAAAATACTTTGAAACAAAAGTGAGCCCTGAGGCCACAACAAACACTTTCCTGACTGCGAC
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Selection Cassette:

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