Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ731	**Date of Submission:**	10/19/05
Lexicon Contract Name:	DNA345	**Mutation Type:**	☒ Standard Knock out
LexVision Name:	MAS255N1		☐ Conditional
Reference accessions:	NM_146162	**Is this gene X-linked?**	No

Required Materials: ☒ pKOS clone DNA(s) __pKOS-9_____
☒Target Vector DNA __pKOS-9 TV_____
☒ Targeted ES Cell DNA __2B9_____
☒Genomic Map

Southern Blot Analysis:
External/Internal Probe Strategies

	5' External	**3' External**
Name of Probe:	**15/16**	**17/18**
Restriction Enzyme for Genomic Digest:	**BglI**	**BglII**
Predicted Wild-type Band (kb):	**~15.0 kb**	**~11.0**
Predicted Mutant Band (kb):	**~4.4 kb**	**~13.0**
Probe Size:	**597 bp**	**406**

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:	DNA345-9	5' Primer Name:	Neo3A
3' Primer Name:	DNA345-10	3' Primer Name:	DNA345-4
Predicted Wild-type Band (bp):	450	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	308

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
DNA345-15 5' – GTTCCCTCTGGTGCGAATCCT
DNA345-16 5' – TCAAAGAGCTGTCGGGACTG
DNA345-17 5' – AAGGCTGCCCCTCTTCTATGA
DNA345-18 5' – ACCCCAGGTCCCTCTACTCCC

PCR Genotyping
DNA345-9 5' – GAAGAGGTGCTCAGCGGTCA
DNA345-10 5' –CCAGGGTGGCAAGAGTCATTAAGG
DNA345-4 5' –CCTCCAGGACTCCTCAGGTACGTG
Neo3a 5' –GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

ATGGTCCCCTGGTTCCTCCTGTCTCTGCTGCTACTTGCGAGGCCTGTGCCTGGGGTGGCCTACTCTGTGTCACTCCCGGC
CTCCTTCCTGGAGGATGTAGCCGGCAGCGGGGAAGCTGAGGGTTCTTCAGCCTCTTCCCCGAGCCTGCCGCCGCCTGGG
ACTCCAGCCTTCAGTCCCACACCGGAGAGACCCCAGCCCACAGCTCTGGACGGCCCCGTGCCACCCACCAACCTCCTGG
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GTCTGCGCCGCCCTCATCACGCGCCAGAAGCACAAGGCCACAGCCTACTACCCATCCTCGTTCCCTGAAAAGAAGTATG
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GACAATGCTTAGGCTCTGTTCTGAGGGACTCCCCACACCTCCATTCAGGGCTCCCCGAGTCCTCAGCTCTTCGGATGTG
GATATATGACACACTAGCATAATAAATCTTGATCTGGCTTGAGTCTCTCTGG

Genomic Locus: (*The deleted sequence represents nt11144-13163-in the sequence below. KOS9 used to generate the TV represents nt9073-17768 in the sequence below.)*

GGAGGTGGGAAGGGGAGGGGAGGGAAGGATAGGGAAGGGGAGGGAAGGGAAGGAAAGAGGAGGGGAGGGAAGAGT
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Selection Cassette:

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Targeted Locus:

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