



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ698	Date of Submission:	10/13/2004
Lexicon Contract Name:	DNA299	Mutation Type:	X Standard Knock out
LexVision Name:	MAS252N1		☐ Conditional
Reference accessions:	NM_172205	Is this gene X-linked?	No

Required Materials: x pKOS clone DNA(s) __pKOS 17 _____
 x Target Vector DNA __DNA299 17TV _____
 x Targeted ES Cell DNA __2B9 _____
 X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	16/17	14/15
Restriction Enzyme for Genomic Digest:	HpaI	HindIII
Predicted Wild-type Band (kb):	15.0 kb	5.1 kb
Predicted Mutant Band (kb):	11.9 kb	8.7 kb
Probe Size:	360bp	370bp

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:	DNA299-25	5' Primer Name:	Neo3a
3' Primer Name:	DNA299-10	3' Primer Name:	DNA299-10
Predicted Wild-type Band (bp):	465 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	306 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**

DNA299-16 5' – GGTCAGGACAGACTTGTGTC
 DNA299-17 5' – GACCGTGGTGTCTCACTTC
 DNA299-14 5' – GTCAGTCAGTGGACGGAAAG
 DNA299-15 5' – TCGCTTCTGGTGAAAAGTGC

PCR Genotyping

DNA299-10 5' – CTGGGAAGTTGATAAATGGC
 DNA299-25 5' – GGTGAGGCGAGTTCATAAG
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

CTCCCTGCCTTCCCAACAACATGTATCTTGTACAGTTTGTCTAGCTCCTGCTGCCTCTTAGTGCTCCTGGGGACTCTGCCTGCCCGGGCAGCCCATGAGGA
TCCCGTTGAGAAGGTCATAGAAGGGTTAGCCGAGGGCTGAGCAATGCTGAGAGAGAGGTGGGCAAGGCCCTTGAAGGCATCAATAATGGAATCACT
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GGTCTCATCAAGGTCAAGCGGGTACGGAGGCCAGCACGGAGGGGCCCAACCACTACAGTAGTATCTGGGGTGAGTCT

Genomic Locus: 10kb flanking the deletion (the deleted sequence represents nt11494 – 14890 in the sequence below;

pKOS/17, used to generate the target vector, represents nt 3837 – 15624 in the sequence below)

TATGGAGTCTGTCATGCTTTTCAGTAAACATAGGATATTTTAAAGCCAGGTGTGATGGCTTGGTTCTCTAAGGCCAGACTCTCATGGGTGAGCTAGGC
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GACTGGAGTTTGCAGGCACACACAAACACTAATAAAGTTAAAAATTAAGAAAAAAGTATTCCATAAATAAATAATATCATAGAATATGTGGT
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GT
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GGCCTGAAGGGCCACCTGGCCTGTGCTGCTGAGGAGCAGCAGTAGAGAGCATGCTGGTGGGGGAGGGTCTCAGGACTCTAAGTACCCCTGGGGGA

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[illegible]

Selection Cassette:

CGCTCTAGAGGCCATAGCGGCCGATCTCTGAGGCGCGCCGGATCCCGGGCCGCTCTAGCTAGACTAGTCTAGCTAGAGAATTCCGCCCCCCCCCCCC
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Targeted Locus:

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