



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ831	Date of Submission:	10-10-04
Lexicon Contract Name:	DNA267	Mutation Type:	x Standard Knock out
LexVision Name:	SEC501N1		☐ Conditional
Reference accessions:	NM_027829	Is this gene X-linked?	no

Required Materials:

- ☐ pKOS clone DNA(s) __kos18__
- ☐ Target Vector DNA __DNA267-FTV18__
- ☐ Targeted ES Cell DNA __1C8__
- ☐ Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	11 + 12	13 + 14
Restriction Enzyme for Genomic Digest:	BglII	ApaLI
Predicted Wild-type Band (kb):	5.7	12.2
Predicted Mutant Band (kb):	9.9	10.1
Probe Size:	256 bp	235 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:	DNA267-1	5' Primer Name:	Neo3a
3' Primer Name:	DNA267-2	3' Primer Name:	DNA267-8
Predicted Wild-type Band (bp):	276	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	276

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**

DNA267-11 5' – TGGA ACTCTCTAGCCTTGTC
 DNA267-12 5' – AGTTAGGTAGGACTCTCAGG
 DNA267-13 5' – CCTCACACTACATGGTGAG
 DNA267-14 5' – CACTGCTGAGTGTTCCTTCC

PCR Genotyping

DNA267-1 5' – CTATCGTCACACGTGAACCTC
 DNA267-2 5' – CTGGTACAGCTGGTCCATCCTC
 DNA267-8 5' – AGATGGTCTCGTACTGGTAG
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

CTGGGCCAGGCCATGGCCCTGCTGTTGTTTCTGGGCATGACAGCGGCGCTGGCCCCGGGGCTGCCTGCAGTGCAGACCCCAATTTTCGGGAGAAGTTCTCC
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TCCCCCTCCCCAGGCCGACTGTACTGTCAGCGTCACTGTGGTAAGTGAAC

Genomic Locus: (the deleted sequence represents nt10268 – 11300 in the sequence below)

TGACCTCTTGCTTCAACTGTGAGCTGAGACACCAAGCACTGCCGCTGCATAGTTAACCTTTTAACTCTATAAACTCCCATCAACCTCAGCACTT
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[illegible]

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Selection Cassette:

GCCGCTCTAGAGGCCATAGCGGCCATTTAAATGGCGCGCCGGATCCCGGGCCGCTCTAGCTAGACTAGTCTAGCTAGAGAATTCCGCCCTCTCCCTCC
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