



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ2491	Date of Submission:	10-11-05
Lexicon Contract Name:	DNA283	Mutation Type:	X Standard Knock out
LexVision Name:	MEM776N1		☐ Conditional
Reference accessions:	NM_019999	Is this gene X-linked?	NO

Required Materials: X pKOS clone DNA(s) __pKOS 88_____
X Target Vector DNA __pKOS-88TV Neo_____
X Targeted ES Cell DNA __2A6_____
X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' Internal</u>
Name of Probe:	21/22	25/26
Restriction Enzyme for Genomic Digest:	BstE II	BstE II
Predicted Wild-type Band (kb):	8.2	6.6
Predicted Mutant Band (kb):	18.4	18.4
Probe Size:	414 bps	519 bps

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:	DNA283-7	5' Primer Name:	NEO 3A
3' Primer Name:	DNA283-8	3' Primer Name:	DNA283-6
Predicted Wild-type Band (bp):	563	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	510

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**

21 5' – GTTTAATGCATGCGAGTTCC
 22 5' – CTATGTCCTTGGCCTAACAG
 25 5' – GAGTAGGAAAGCTGCCCTAT
 26 5' – CTCCGATAAGCAAATGCTAC

PCR Genotyping

6 5' – TCCTGTGGTCCAGACTTGGT
 7 5' – TCAGATTCGAGCCCTAGCCA
 8 5' – ACAGTGCCCCGAGTGCAG
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

TACACCGGAACCTGGCTTGGGTACCTCTTCTACCGACAGCAGCTTCGGAGGGCTCGGAATCGCTATCCCAAAGGCCACT
CCAAAACCTCAGCCCCGCCTCTTCAATGGTAATCTCTGTCTGCCCTGGCCAGCACCCACCTTGTTCCCTACGCTCACAC
ACACACACCCACACCCCGCGTGCCTACTTCCCTTCTCCCCCTGACCCCTTCAGTGGGCTCCCACCCCGCCCCAGCCTC
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Genomic Locus: (*The deleted sequence represents nt-18143-20673-in the sequence below. KOS 88 used to generate the TV represents nt—15779-25506 in the sequence below.*)

[illegible]

[illegible]

[illegible]

[illegible]

[illegible]

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GAAT

Selection Cassette: MC1 LACZ NEO

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