



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ8782	Date of Submission:	4-17-06
Lexicon Contract Name:	DNA452	Mutation Type:	x Standard Knock out
LexVision Name:	MAS264N1		☐ Conditional
Reference accessions:	ENSMUST00000048452, NM_032398	Is this gene X-linked?	no

Required Materials: x pKOS clone DNA(s) __pKOS64 (previously delivered)_____
 x Target Vector DNA __pKOS64-TVNeo (previously delivered)_____
 x Targeted ES Cell DNA __1H4 (in this shipment)_____
 x Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' Internal</u>	<u>3' External</u>
Name of Probe:	21 + 22	11 + 12
Restriction Enzyme for Genomic Digest:	EcoRI	Acc65I
Predicted Wild-type Band (kb):	11.5 kb	16.5 kb
Predicted Mutant Band (kb):	8.3 kb	7.8 kb
Probe Size (bp):	341 bp	331 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:	DNA452-1	5' Primer Name:	Neo3a
3' Primer Name:	DNA452-2	3' Primer Name:	DNA452-10
Predicted Wild-type Band (bp):	289 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	395 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**

DNA452-21 5' --CAAGAAACGGAGTAAGCAGG
 DNA452-22 5' – AATCCGAGCGGTATGCAGTA
 DNA452-11 5' – CAACTAGGTAGCTCAGGCTT
 DNA452-12 5' – GATCGACAATCTCACCTGGGT

PCR Genotyping

DNA452-1 5' – TCCTCTTCGTGTCGCTCATTCAG
 DNA452-2 5' – CTTACCAGGTCGCCTTGGCAC
 DNA452-10 5' – GTTGCATGTACTACACCAGG
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

ATGGGGCTCTCCATGGACCGCAGCCCATACGCCCGCACGGGGGACCAGCAGCGCGGCTGCTGGTACTACCTGCGCTATTTCTTCTCTTCGTGTCGCTC
ATTAGTTTCTCATCATCTCTGGGCTGGTCTCTTCATGATCTATGGCAATGTGCACGCCACCACTGAGTCCAGCCTGCGCGCCACGGAGATCCGCGCC
GACAGCCTGTACAGCCAGGTGGTTGGACTATCGGCCTCACAGGCTAACCTGAGCAAACAGCTGAACATCAGCTTGCTTGCAAGGAAACAGTCATGCA
GCAACTGTTGACTACGCGACGTGAGATGGAGCGCATCAACGCCAGCTTCCGCCAGTGCCAAGGCGACCTGGTAAGAGGCGGCATGTGTGCCCTG

Genomic Locus: (The deleted sequence represents nt 10000 – 10389 in the sequence below. KOS64 used to generate the TV represents nt 3495 – 13679 in the sequence below.)

[illegible]

[illegible]

Selection Cassette:

Targeted Locus:

[illegible]

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