



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ785	Date of Submission:	1-20-05
Lexicon Contract Name:	DNA213	Mutation Type:	x Standard Knock out
LexVision Name:	MSC338N1		☐ Conditional
Reference accessions:	ENSMUST00000002911, NM_008233	Is this gene X-linked?	no

Required Materials:

- x pKOS clone DNA(s) __pKOS83____
- x Target Vector DNA __pKOS83-TVneo____
- x Targeted ES Cell DNA __2D5____
- x Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	11 + 12	9 + 10
Restriction Enzyme for Genomic Digest:	NheI	EcoRI
Predicted Wild-type Band (kb):	7.3 kb	10.2 kb
Predicted Mutant Band (kb):	14.9 kb	8.5 kb
Probe Size (bp):	318 bp	300 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:	DNA213-1	5' Primer Name:	Neo3a
3' Primer Name:	DNA213-2	3' Primer Name:	DNA213-5
Predicted Wild-type Band (bp):	337 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	300 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**

DNA213-11 5' – CATCATTGAAACTGCCCCAC
 DNA213-12 5' – ACAGGTGAAGATGGGATCTC
 DNA213-9 5' – CAGCCGCATTCATTTGCATG
 DNA213-10 5' – CAGGATCTCTGCTGAGTTAG

PCR Genotyping

DNA213-1 5' – GCTGGCTCAGGTGACTGCACT
 DNA213-2 5' – CGCATTTCTGACCTCTGGTAC
 DNA213-5 5' – GTAAAGCCCAGCAATACACC
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

CGCGGCCTGGGCCTCCGCCGACAGCATGCCGCACGCCTTCAAGCCCCGGGGACTTGGTGTTTGCCAAGATGAAGGGCTACCCGCACTGGCCGGCCCCGGG
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AACAACCCCATGCCAGTACAGTGCGCTCCG

Genomic Locus: (The deleted sequence represents nt 9999 – 12684 in the sequence below. KOS83 used to generate the TV represents nt 5801 – 14538 in the sequence below.)

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Selection Cassette:

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Targeted Locus:

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