



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ2553	Date of Submission:	1.6.05
Lexicon Contract Name:	DNA342	Mutation Type:	x Standard Knock out
LexVision Name:	MEM734N1		☐ Conditional
Reference accessions:	ENSMUST00000001454, NM_020003	Is this gene X-linked?	No

Required Materials: x pKOS clone DNA(s) __KOS42, KOS21_____
x Target Vector DNA __TV42-2_____
x Targeted ES Cell DNA __1B3_____
x Genomic Map

Southern Blot Analysis: External/Internal Probe Strategies

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	13+14	19+20
Restriction Enzyme for Genomic Digest:	SpeI	EcoRI
Predicted Wild-type Band (kb):	9.9 kb	14.2 kb
Predicted Mutant Band (kb):	7.5 kb	7.1 kb
Probe Size:	249 bp	224 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:	-5	5' Primer Name:	Neo3a
3' Primer Name:	-21	3' Primer Name:	-9
Predicted Wild-type Band (bp):	268 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	376 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**

DNA342-13 5' – TAGCTGCATGTCTACACAGACGTG
 DNA342-14 5' – CACAAGAATGAGCTGCCATACC
 DNA342-19 5' – ACAGCCAGGTTAAAGAAGTAGAGG
 DNA342-20 5' – CCCGACCCAAAGAGCTATCT

PCR Genotyping

DNA342-5 5' – TGGCTCCTTTTAGTATGGGGTT
 DNA342-21 5' – TCAGCAGTACAGGCACTAAGTCAC
 DNA342-9 5' – ATGTAGGCCTTCAAGTTTCATC
 Neo3a 5' – GCAGCGCATCGCCTTCTATC

[illegible]

Selection Cassette: LacZ/Neo

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Targeted Locus:

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[illegible]

CAGGTGTGTGTGTGTGTGCTGTTCAAAGCATTGCCCCTTGTGCATACATAATAGCCCTGACACTCATCTGGAATCATGGACAAC TTCATGACTCCAGGT
GAGTGTCCCATGACTTCATGGGAGTCAGTCTTCTCCCTCACCATGTGGGTGGGGCTCCAGAGTCAGACTTGGGTTATCAGGCCTGACATCAAGTTTTGC
CAGCCCATGGTGCCTTATTTTGTTAAGATTTTTTTTAAAAAGTATATGCATATACATGAAAGGGTTGTGCATGTATGAGTCTCAGTTACTCACAGTCATG
TCCACTCTGTGTGTGCTTG