



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ756	Date of Submission:	10-10-04
Lexicon Contract Name:	DNA263	Mutation Type:	X Standard Knock out
LexVision Name:	ENZ757N1		☐ Conditional
Reference accessions:	ENSMUST00000036372	Is this gene X-linked?	No

Required Materials: X pKOS clone DNA(s) __KOS48, KOS84_____
 X Target Vector DNA __KSO48-FTV_____
 X Targeted ES Cell DNA __2C4_____
 X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	20+21	24+25
Restriction Enzyme for Genomic Digest:	SacI	BstX1
Predicted Wild-type Band (kb):	9.0 kb	10.2 kb
Predicted Mutant Band (kb):	8.0 kb	8.8 kb
Probe Size:	295 bps	397 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:	28	5' Primer Name:	Neo3A
3' Primer Name:	29	3' Primer Name:	30
Predicted Wild-type Band (bp):	261 bps	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	327 bps

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**

DNA263-20	5' – TGACGACGAAAACTTGGCC
DNA263-21	5' – AACGAGCTGTAGAGGAAACC
DNA263-24	5' – TCTAGTAAGAGGGCACTGTG
DNA263-25	5' – AAAACGTTTGAATGAGCCAGG

PCR Genotyping

DNA263-28	5' – CAGGAAC TTTCTCTTCGTGGG
DNA263-29	5' – ACACACTGAGGCCAGCGTAC
DNA263-30	5' – AGGCCAGCGTACTCAATCCC
Neo3A	5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

ATTTAAAAAACTAATATGCCGGGCTTGGTGCCGCTCGCCTTTAAGCCCAGCGCTCGAGAGGCAGAGACAGATCTCAGA
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AGGTGAACTGTGGCCGTGTGTGGCTCCAGCCCCGCCTGCAGCGTACAAAGCTGCAGTGTGGACCCCGCTGTCCTGGCCA
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CAGGGCTCGGCCGCGGAGGGCGTCCCGCGCGACAGGAACCTTCTCTTCGTGGGAGTCATGACAGCCCAGAAATACCTG
CAGACCCGCGCCGTGGCCGCTACAGGTGAGCCCCCT

Genomic Locus: (the deleted sequence represents nt 9,045-10,406 in the sequence below)

CTGTCCCTACATCCTGATAGGTGAAATAACTGGGCACAGGTGTTTGTGGACTTCAAGGCTTAAAAGCATGGTAGGATCC
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Selection Cassette:

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