



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ674	Date of Submission:	11.17.04
Lexicon Contract Name:	DNA490	Mutation Type:	X Standard Knock out
LexVision Name:	SEC383N1		☐ Conditional
Reference accessions:	NM_031380	Is this gene X-linked?	No

Required Materials: X pKOS clone DNA(s) pKOS 80
X Target Vector DNA DNA490.80 TVPuro
X Targeted ES Cell DNA 1A4
X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	19/20	30/33
Restriction Enzyme for Genomic Digest:	EcoRV	EcoRV
Predicted Wild-type Band (kb):	15.1 kb	15.1 kb
Predicted Mutant Band (kb):	10.1 kb	7.3 kb
Probe Size:	213 bp	177 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:	DNA490.38	5' Primer Name:	DNA490.38
3' Primer Name:	DNA490.36	3' Primer Name:	IRES up.LR
Predicted Wild-type Band (bp):	297 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	232 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**

DNA490.20	5' – CCAAGACTTTGAAGGTCCAAC
DNA490.19	5' – CTGCTACAATGAGGCGCAG
DNA490.30	5' – GCTAGGATGGCAAGAGTGGAG
DNA490.33	5' – CAGTCACCCTTGTAACCAAGAC

PCR Genotyping

DNA490.36	5' – GTCAGTCCTCTTTTGAGGCTG
DNA490.38	5' – GCTTTTATAGGCCGAGGCGTCT
IRES up.LR	5' – TGGAAAATAACATATAGACAAACGCACACC

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Selection Cassette: BetaGeo/ Puro

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