



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ1903	Date of Submission:	7-13-04
Lexicon Contract Name:	DNA017	Mutation Type:	<input checked="" type="checkbox"/> Standard Knock out
LexVision Name:	SEC440N1		<input type="checkbox"/> Conditional
Reference accessions:	BC011475	Is this gene X-linked?	NO

Required Materials: X pKOS clone DNA(s) KOS 25
 X Target Vector DNA FTV25 NEO
 X Targeted ES Cell DNA 1H7
 X Genomic Map

Southern Blot Analysis:
External/Internal Probe Strategies

	<u>5' Internal</u>	<u>3' External</u>
Name of Probe:	15/16	13/14
Restriction Enzyme for Genomic Digest:	EcoRI	BamHI
Predicted Wild-type Band (kb):	7.4	10.6
Predicted Mutant Band (kb):	5.9	7.4
Probe Size:	443 BP	313 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:	DNA017-5	5' Primer Name:	NEO 3A
3' Primer Name:	DNA017-6	3' Primer Name:	DNA017-11
Predicted Wild-type Band (bp):	473 BP	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	507 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**

DNA017-15 5' – CCAGTAGATCACGGGTGGTA
DNA017-16 5' – GCCTGGGATTTATTCTCTGACGA
DNA017-13 5' – CTGGCCAGGAAGGGGATAGAATA
DNA017-14 5' – CTGTGTCTTGTAGGGGGTTC

PCR Genotyping

DNA017-5 5' – CTCCCTGAGTGTCCGATAAAGATG
DNA017-6 5' – CGCCATTTTTGTGACTCT
DNA017-11 5' –GGCCTTACCTGACGGACATCACC
Neo3a 5' - GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

AAGCTCCCGAACAAAGGGGATAACCTTCGGCCCTCTCGGCCCCACCCCCGCAGCGGGAGCCGCGCCCCCTTTGCCATA
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Genomic Locus: (the deleted sequence represents nt 8011-11199-in the sequence below)

CTTGTAAGCAACCAACCATGTGGATGCTGGGAAATGAACCTGAATAGTCTGCCAGAGCAGGAAGTGCTCTTAACCATT
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Selection Cassette:

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Targeted Locus:

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