



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ395	Date of Submission:	5/4/04
Lexicon Contract Name:	DNA074	Mutation Type:	X Standard Knock out
LexVision Name:	MEM655N1		☐ Conditional
Reference accessions:	NM_017378	Is this gene X-linked?	No

Required Materials: X pKOS clone DNA(s) __pKOS31_____
X Target Vector DNA __DNA074-FTV31_____
X Targeted ES Cell DNA __2C4_____
X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' Internal</u>	<u>3' External</u>
Name of Probe:	Ires 8/9	13/14
Restriction Enzyme for Genomic Digest:	EcoRV	EcoRI
Predicted Wild-type Band (kb):	N/A	13.1
Predicted Mutant Band (kb):	5.7	4.5
Probe Size:	291 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:	DNA074-1	5' Primer Name:	Neo3A
3' Primer Name:	DNA074-2	3' Primer Name:	DNA074-12
Predicted Wild-type Band (bp):	329	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	275

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**

IRES 8	5' –AGGAAGCAGTTCCTCTGGAA
IRES 9	5' –CACATGTAAAGCATGTGCACC
DNA074-13	5' – CCCTATCTGATCACAGAAGC
DNA074-14	5' – CAAGACTGTAGTGACTIONTAGGG

PCR Genotyping

DNA074-1	5' – AGGCTGACCAAGCCATTCAGGA
DNA074-2	5' – TCCTCAGAGTTCATCTGAACC
DNA074-12	5' – GCATCACCCAGTACTGAGTGAG
Neo3A	5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

CATGATGCTACTTCTGCCATTCTGCTAGGGCTCTTAGGGCCAGGAAGCTACTTGTTTCATTTCAGGGGATTGTCAGGAGGTGGCCACTGTTCATGGTGAA
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AGCCTGCTGGGACTCTCTCTGTCCAGGTACCTCTGG

Genomic Locus: (the deleted sequence represents nt 10426—13316 in the sequence below)

CACCTGAGAGGAAGGGAATGTTTCAAAGTAGAGTTAAGAATTTGATGGCAGGGCTGGTGAGATGGCTCAGTGGGTAAAGAGCACCTGACTGCTCTTCTGA
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Selection Cassette: LacZ/Neo

Targeted Locus:

[illegible]

[illegible]

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[illegible]

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