



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ200	Date of Submission:	4-6-05
Lexicon Contract Name:	DNA470	Mutation Type:	X Standard Knock out
LexVision Name:	SEC407N1		☐ Conditional
Reference accessions:	NM_021474	Is this gene X-linked?	No

Required Materials: ☐ pKOS clone DNA(s) __ pKOS20, pKOS63, pKOS65, _____
☐ Target Vector DNA __pKOS20.TV_____
☐ Targeted ES Cell DNA __2F1_____
☐ Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	19+20	21+22
Restriction Enzyme for Genomic Digest:	EcoRV	EcoRI
Predicted Wild-type Band (kb):	20.1 kb	9.1 kb
Predicted Mutant Band (kb):	17.2 kb	5.1 kb
Probe Size:	336 bp	259 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:	DNA470-10	5' Primer Name:	Neo3A
3' Primer Name:	DNA470-11	3' Primer Name:	DNA470-23
Predicted Wild-type Band (bp):	581 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	317 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**

DNA470-19 5' – AGAGACGTCCACAAACTGC
DNA470-20 5' – GGAGTTTGGCCAGTTATACG
DNA470-21 5' – CGTCTCACTGTGTTACCTG
DNA470-22 5' – AACCCATTTTAGAAAGGGTCC

PCR Genotyping

DNA470-10 5' – CTGCGAAAGGATTCTGTCC
DNA470-11 5' – TCCTGAAGATGGCTAGTACC
DNA470-23 5' – ACACGCCTATAATCCCAGC
Neo3A 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

CCACCTCCCAGCCTCAGGATGCTCCCTTTTGCCTCCTGCCTCCCCGGGTCTTTGCTGCTCTGGGCGTTTCTGCTGTTGCTC
TTGGGAGCAGCGTCCCCACAGGATCCCCGAGGAGCCGGACAGCTACACGGTGGGGACCGTGGGGTCAACCAGCTTGGGAT
GGGATGGGAAGGACAGTGAGATCAGGATCCTGGGGAGGCTGCTTCTGGGCAAGATTCTCCCCGGCTTGGTTGGTGGAA
AGCTGGAGGCCATTGTGTGGAGCTGACCCGAGCATGACTCTGAACTCTGGCCCTGGTCTTGGCCTGGCTAGTTGGTGGAA
GACAGCCAGGGTGGTCACTGGCCACATCGGGTTCCCTTCCAGGAATGCACAGATGGCTATGAGTGGGATGCAGACAGC
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GAGTGCCTGACCATCCCGAGGCTTGCAAGGGTGAGATGAAATGCATCAACCACTACGGGGGTTATTTGTGTCTGCCTC
GCTCTGCTGCCGTCATCAGTGATCTCCATGGTGAAGGACCTCCACCGCCAGCGGCCCATGCTCAACAACCAACCCCTTG
CCCGCAGGGCTACGAGCCTGATGAACAGGAGAGCTGTGTGGGTAAGCAAGGGC

Genomic Locus: (The deleted sequence represents nt 15511-16745 in the sequence below. KOS20 used to generate the TV represents nt8339-18526 in the sequence below.)

TTTTCCCCCATTTTTTCGAGACAGGATTTCTCTGTGTAGCCCTGGCTGTCTGAACTCACTCTGTAGACCAGGCTAACCT
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[illegible]

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Selection Cassette:

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Targeted Locus:

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[illegible]

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