



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ851	Date of Submission:	2/7/06
Lexicon Contract Name:	DNA433	Mutation Type:	X Standard Knock out
LexVision Name:	ENZ709N1		☐ Conditional
Reference accessions:	ENSMUST00000022821, NM_030686	Is this gene X-linked?	No

Required Materials:

- x pKOS clone DNA(s) __pKOS-80_____
- x Target Vector DNA __pKOS-80 TVneo_____
- x Targeted ES Cell DNA __1B12_____
- x Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	17/18	25/26
Restriction Enzyme for Genomic Digest:	Acc65I	Eco RI
Predicted Wild-type Band (kb):	Expected 19.2; Observed 15.9	18.5
Predicted Mutant Band (kb):	Expected 15.1, Observed 11.8	10.7
Probe Size:	307 bp	554 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:	DNA433-1	5' Primer Name:	Neo3a
3' Primer Name:	DNA433-29	3' Primer Name:	DNA433-7
Predicted Wild-type Band (bp):	515	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	250

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**

DNA433-17 5' – GCTTCATTCCCATGGTTAGG
DNA433-18 5' – GAGTGAGGCATTTAAGAGGC
DNA433-23 5' – GGAGACCAGGATATCAATCC
DNA433-24 5' – CGCTTAGATGCTCTACAAAG

PCR Genotyping

DNA433-1 5' – CCTGAACTATGCAGAAAGCG
DNA433-7 5' – GTATCTCACCAATGCAGTCC
DNA433-29 5' – GAAGTAAAGATCTGGGCACG
Neo3a 5' – GCAGCGCATCGCCTTCTATC

CAGTCCCCTCCCCTAGCAGAGTTCAACCCAACCCCTGGCCTGAACTATGCAGAAAGCGGGACGGCTGTTAGGTGGTTGGACTCAAGCGTGGATGTGCGGTGAGGATGGCCAGTCTCCGGGTGACTCTCGTGAAACCCCTCTCTCCAAATAAGGTGGCCCTGTGTACACAGCTCCACCGCAGGGTGAGTACCGAGCCGAGA GTCTTCTACCGAAGAACAGTACTCTTTAACTCTGGACTGTGTCTCTAGGCCTCAGATCGCGGCAAAAGGCCACCCCGAGGGAGAAAAAGGAAACCCA CATGGTGGACCTGAAATCGCTCCGAATCCCCGACCTGGCCCTGCCACAGGCTAGCCCTTGTATCACTGTTGCCTTCGGCTTTGTGAGAGCTCGGCGCTG CCAGAAGGACCCGAGGTCCCGGTGCTCTGGGCTGCCCAATCGACAGAGCTCCACCTAGAGTCTGGGAAGACCAAAACAACCGATCACGGCTCTCAAAGT CTGCTTGCTGTTCTTGCTCCCTAGGGACCCCTTACCTGGATCCCGTGCCAGATCTTTACTTCAGTCTGATATTTTGGGCTCCCGTCCCGAGCTGTCTCA CCCACCACCTCAACTATCCAGTGTCTTTTAAATCCCCACAGGACCTTACTAAATTAAAAAAATCTCTTGAAGATCTGTAATGCAGATGTCTATGAAAAGCTA ACATCTGGCTATTATAAAATATGTAATGATATTACATTTTGAGCCATGCTTTTTTGTGTGAAGGCCCCCCCAAAATATATCAAAGATAGAATTGTGA GGTGTAGTGTGGTAAGAGGATATTTGGGATATGGGGTCTCTTCCAGCAAGGCCCTTCAAGTCCATGGCAACTGAAACATCATCTATTTCTCTCCCCTTTT CCCAACTGGACATAGGTACAATAATCGGTATGAAGGCTGTGGCAGGTGATGTCAGTCCCGGGAATCTGGGGTAAAAAGACAAGATCATAATAAAATCA TCTGAGCTTATCCTTCTGTCCGCACCATTAAGAGCCTACTCTACCTAGTAGAAATGCGTGAAGCTCGAATACAGGGAGAGCGAGGGTCCAGTGTCTGAA AGCCTGAAAGCAGAGGCCACGCATGCTCTCGCCCTCTTCTCTCGGTAGAGGCGCTTGACAGATGTGTCACTTTTACCTCTCGTAGGATCGGCTTTGCCAT CGCCCGGCGCTGGCTGAAGATGGGGCCACGCTGGTCTGCAGAGCCGCAAAACAGCAGATGTGGACCTGTCAGTGGCCACACTACAGGGAGAGGGC CTGAGTGTGACTGGCATCGTGTGCCACGTGGGGAAGGCAGAGGATCGAGAAAAGCTGATAACCAACGGTGAGCAGGC

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