



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ8752	Date of Submission:	7.29.04.
Lexicon Contract Name:	DNA243	Mutation Type:	☒ Standard Knock out
LexVision Name:	ENZ759N1		☐ Conditional
Reference accessions:	NM_152922 ENSMUST00000015611	Is this gene X-linked?	

Required Materials: ☒ pKOS clone DNA(s) pKOS-17, -61
☒ Target Vector DNA DNA243-17-TV2
☒ Targeted ES Cell DNA 1A3
☒ Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	5' External	3' External
Name of Probe:	17+18	19+20
Restriction Enzyme for Genomic Digest:	ApaLI	AvrII
Predicted Wild-type Band (kb):	8.9 kb	6.8 kb
Predicted Mutant Band (kb):	5.6 kb	9.2 kb
Probe Size:	158 bp	152 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:	DNA243-23	5' Primer Name:	Neo3A
3' Primer Name:	DNA243-24	3' Primer Name:	DNA243-25
Predicted Wild-type Band (bp):	274 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	266 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**

DNA243-17 5' – CCTTACCTAGCTCACTTG
 DNA243-18 5' – CCTGTGTAAAAGTGCTCC
 DNA243-19 5' – CACTTCCAGTCCGTTCTC
 DNA243-20 5' – CTTCTACAACGGCTGGATC

PCR Genotyping

DNA243-23 5' – GTTCCTCTCCGTTACAACGAG
 DNA243-24 5' – CTTCTTCCAGCCCTGACA
 DNA243-25 5' – GCCCTGCTAGTTTAGTCAC
 Neo3A 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

TTTGGGAGTGTGCTCCAAGCAGACGCTGCTGGTTCCTCTCCGTTACAACGAGTCCATATAGTCAACCGGTGTACAAACCCCTACCTGACCTTGTGTGCGGG
GAGGCGCATATGTAGACCTACAGGTGAGAGGTGAAGCCCCTAAGGCTCCAGGTCTCAAAGGGACCCCAAGAGGCAAGTCCAGGATGGGTACCTAC
AGATATGAGCCTGCTTCTGTGTTCTAGGACACATACCGTGTGGCTGGCGGGAGGTAGGCGGGAGGTACCAACAGACACACGTTGGTGTCTGTCAGG
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GTCTTAACCTGTCTTCTCAAATGGACCCAGCCATCTGCTCCAAGCCTTGTCTTAATGGAGGTGTCTGCACTGGACCAGACCGGTGCGAGTGTGCCCCAG
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CACTCTCTTTTTCTTTGTCTAGTTCGGGAAGCAGACAGCGAAGAGGAGCGAGCCCTGAGGTGGGAGGTTGCAGAGCTTCGTGGGCGTCTGGAGAAG
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GCTGGGGCCTGGGTTGAGCAGTAGTGTCCCATGCCCCCTGAAGAGCTGCGGCTGAACAGGTGGCTGAGCTTTGGGGCGGGGTACAGAATTGAGTC
CCTCAGCGACCAGGTGCTGCTACTGGAGGAAAGACTGGGTGCCTGTGA

Genomic Locus: 10kb flanking the deletion (the deleted sequence represents nt 10026 – 11254 in the sequence below; pKOS-17, used to generate the target vector, represents nt 6278 – 13260 in the sequence below)

TTTTTGTTTGTTTGTGGTTTTTGTGGTTTTTAAGACAGGGTTTCTCAAGCCGTGCATGGTGGTGCACGCCCTTAATCCCAGCACTTGGGAGGCAGAGGC
NN
NNNNNNNNCTCTGAGTGTGGGATTAAGGTGTGCACCATGCTTCCCGCGCGCTTCTTTACCATATACGAAAGCCACCTCCGATGACACACATGC
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Selection Cassette:

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Targeted Locus:

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