



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ518	Date of Submission:	1-10-05
Lexicon Contract Name:	DNA204	Mutation Type:	X Standard Knock out
LexVision Name:	MEM720N1		☐ Conditional
Reference accessions:	NM_025453, ENSMUSESTT00000002362	Is this gene X-linked?	No

Required Materials: X pKOS clone DNA(s) __pKOS13_____
X Target Vector DNA __pKOS13 TV_____
X Targeted ES Cell DNA __1C12_____
X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' Internal</u>	<u>3' External</u>
Name of Probe:	1+2	16+18
Restriction Enzyme for Genomic Digest:	EcoRV	Bgl I
Predicted Wild-type Band (kb):	21.0 kb	10.7 kb
Predicted Mutant Band (kb):	11.0 kb	6.2 kb
Probe Size:	286 bp	516 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:	DNA204-4	5' Primer Name:	Neo3A
3' Primer Name:	DNA204-5	3' Primer Name:	DNA204-5
Predicted Wild-type Band (bp):	484 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	301 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**

DNA204-1 5' – GATATGCAGTGACAGGATGG
 DNA204-2 5' – GCTATTGTCTCACATGCAGC
 DNA204-16 5' – CATCCTTACCAGTTCTTGTCC
 DNA204-18 5' – ACAGGTGGTGTAACCTTGCC

PCR Genotyping

DNA204-4 5' – TGGACTCTGAACTGCTTTCC
 DNA204-5 5' – TAAGGTCCCTAAGATGGACC
 Neo3A 5' – GCAGCGCATCGCCTTCTATC

Genomic Sequence Deleted:

GGAAAAACCATGACGTGCTGTGAAGGGTGGACATCCTGCAATGGCTTCAGCCTGCTCATTCTGATCCTGCTAGGAGTGGT
TATCAATTGTATACCCCTGGGAATCAGCTTAGTGGAGGCAGACTCGACTTCTCAAACCCCATCTCCTGCTATGAGTGG
TGGTTTCCAGGAATTATAGGAGCAGGTCTGATGGTGAGTAATACCTACATGAACAGACTTGGAG

Genomic Locus: (The deleted sequence represents nt15393-15614 in the sequence below. KOS13 used to generate the TV represents nt-6733-17149 in the sequence below.)

[illegible]

AGTTACCCTCTGTGTTTTTCAGATAGTCTAATATTCTCAAAGGGTGTAAATAGCCAATCTTCAGTGCAAGGAAGTACAAAG
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Selection Cassette:

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[illegible]

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