



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ9196	Date of Submission:	9-16- 03
Lexicon Contract Name:	DNA181	Mutation Type:	☒ Standard Knock out
LexVision Name:	GPR044N1		☐ Conditional
Reference accessions:	ENSMUST00000049962, NM_153835 (Hs)	Is this gene X-linked?	no

Required Materials: ☒ pKOS clone DNA(s) __pKOS-58, pKOS-4_____
☒ Target Vector DNA __pKOS-4TV_____
☒ Targeted ES Cell DNA __2A1_____
☒ Genomic Map

Southern Blot Analysis: External/Internal Probe Strategies

	<u>5' Internal</u>	<u>3' External</u>
Name of Probe:	66+67	57+58
Restriction Enzyme for Genomic Digest:	SpeI	EcoRI
Predicted Wild-type Band (kb):	~15	10
Predicted Mutant Band (kb):	~12.4	4.5
Probe Size (bp):	179	278

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:	71	5' Primer Name:	Neo3A
3' Primer Name:	70	3' Primer Name:	52
Predicted Wild-type Band (bp):	590	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	456

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**

66 5' – GAGCTACCATAGACCTGGCTG
67 5' – CATTCTGTCCGTGCACCACAG
57 5' – CAGCAGTTGGAGGCTAGCATC
58 5' – CGTGATAGAGACAGGAGCC

PCR Genotyping

71 5' – GCAGCAACTGAACCAGTACAGTG
70 5' – CAGATGCACATAGCTGGAGAC
neo3A 5' – GCAGCGCATCGCCTTCTATCGCC
52 5' – GTTCCTCACTACCTATCAG

TAGCTCTCTCCAGCTGCTCCTGGCCGTGACTCTTCCCCTGCTGGAGTTGGAGCCAACATTTGTCCCCACAGCCCAATCAGTAAGTACTGGGGTGAGTG
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[illegible]

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Targeted Locus:

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