



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ728	Date of Submission:	1.6.04
Lexicon Contract Name:	DNA226	Mutation Type:	x Standard Knock out
LexVision Name:	SEC507N1		☐ Conditional
Reference accessions:	AK087615.1	Is this gene X-linked?	No

Required Materials:

- x pKOS clone DNA(s) __KOS42, KOS66__
- x Target Vector DNA __TV66-1__
- x Targeted ES Cell DNA __3B3__
- x Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' Internal</u>
Name of Probe:	1+2	7+8
Restriction Enzyme for Genomic Digest:	HindIII	SacI
Predicted Wild-type Band (kb):	6.1kb	4.9kb
Predicted Mutant Band (kb):	5.5kb	7.1kb
Probe Size:	400bp	180bp

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:	DNA226-12	5' Primer Name:	Neo3a
3' Primer Name:	DNA226-13	3' Primer Name:	DNA226-6
Predicted Wild-type Band (bp):	497	Predicted Wild-type Band (bp):	None
Predicted mutant band (bp)	none	Predicted mutant band (bp)	311

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**

DNA226-1 5' – TCCTTGGCCTTTATTAGAACCT
DNA226-2 5' – GCAATCATACCCTAGCCCTAAC
DNA226-7 5' – GTGCAGGTTCCAGTACACTT
DNA226-8 5' – AACTGTGAAGCCAACATACTCA

PCR Genotyping

DNA226-12 5' – ATGCCGCTAAGCCTGTTACCC
DNA226-13 5' – TTGTAAACCACCATGCCAGGCC
DNA226-6 5' – TCATACCTTTAATCTTGCC
Neo3a 5' – GCAGCGCATCGCCTTCTATC

Contact Information:

Laura L. Kirkpatrick, Ph.D.
Senior Scientific Group Leader, Molecular Genetics
Lexicon Genetics, Inc.
8800 Technology Forest Place
The Woodlands, TX 77381-1160
Phone: 281-863-3016
Email: laurak@lexgen.com

Additional Contact:

Kathleen Holt, Ph.D.
Senior Director Molecular Genetics & Alliance Management
Lexicon Genetics, Inc.
8800 Technology Forest Place
The Woodlands, TX 77381-1160
Phone: 281-863-3013
Email: kholt@lexgen.com

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