



Lexicon Genetics Incorporated – Genentech Project Materials

Genentech ID:	UNQ522	Date of Submission:	10.25.04
Lexicon Contract Name:	DNA419A	Mutation Type:	X Standard Knock out
LexVision Name:	PRT215N1		☐ Conditional
Reference accessions:	AK076281	Is this gene X-linked?	No

Required Materials: X pKOS clone DNA(s) __pKOS8__
X Target Vector DNA __DNA419-8-TV2.1 puro__
X Targeted ES Cell DNA __1E3__
X Genomic Map

Southern Blot Analysis: *External/Internal Probe Strategies*

	<u>5' External</u>	<u>3' External</u>
Name of Probe:	25/26	29/30
Restriction Enzyme for Genomic Digest:	Stu1	EcoR1
Predicted Wild-type Band (kb):	6.8 kb	9 kb
Predicted Mutant Band (kb):	10 kb	7 kb
Probe Size:	349 bp	241 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:	DNA419.18	5' Primer Name:	DNA419.34
3' Primer Name:	DNA419.33	3' Primer Name:	GT-IRES
Predicted Wild-type Band (bp):	355 bp	Predicted Wild-type Band (bp):	none
Predicted mutant band (bp)	none	Predicted mutant band (bp)	574 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**

DNA419.25 5' – CAGACATACTTGGTAAGGTGAC
 DNA419.26 5' – GTTCCTAAAGAGTTTATTTTGC
 DNA419.29 5' – CTGTCACAATTCAATATCTGG
 DNA419.30 5' – CAGTGCAAGAAACATTTTGAGG

PCR Genotyping

DNA419.18 5' – GCATCGAATGTCAGGAAGAG
 DNA419.33 5' – CTGCCTTTGACATAGTCCTTC
 DNA419.34 5' – TGGAGCAGTGAACACGCAGAG
 GT-IRES 5' – CCCTAGGAATGCTCGTCAAGA

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