Mylk Cas9-CKO Strategy

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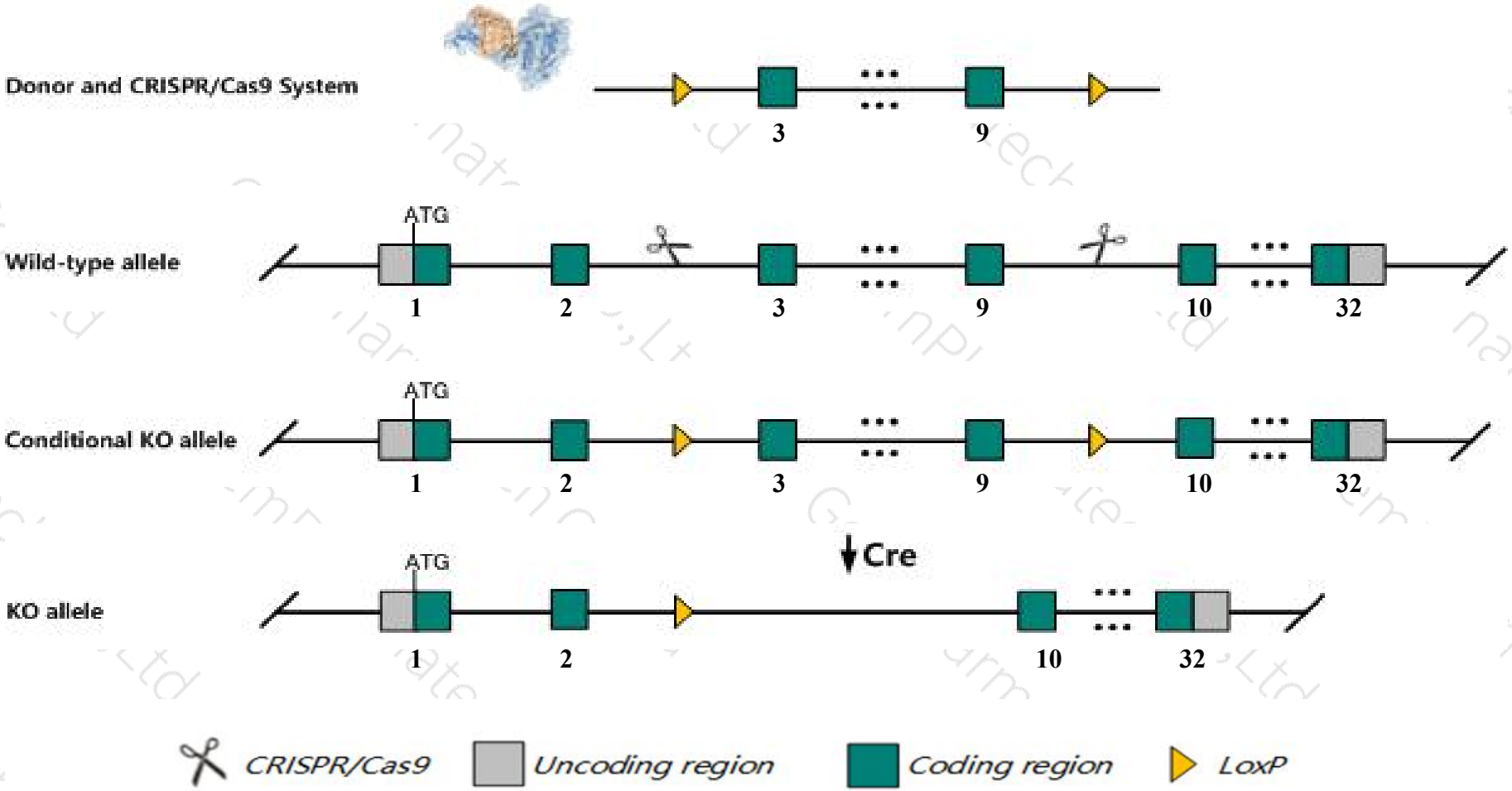
Design Date: 2019/10/15

Project Overview

Project Name	***Mylk***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Mylk* gene. The schematic diagram is as follows:



Technical routes

- The *Mylk* gene has 8 transcripts. According to the structure of *Mylk* gene, exon3-exon9 of *Mylk-201* (ENSMUST00000023538.8) transcript is recommended as the knockout region.The region contains 1315bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Mylk* gene. The brief process is as follows:CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

Notice

- According to the existing MGI data,Mice that lack the isoform abundant in endothelial cells show a reduced susceptibility to acute lung injury. Mice lacking the smooth muscle isoform exhibit partial pre- or neonatal lethality, short small intestine and impaired smooth muscle contraction in the colon.
- *Mylk-203* transcript may be unaffected.
- The *Mylk* gene is located on the Chr16. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Mylk myosin, light polypeptide kinase [*Mus musculus* (house mouse)]

Gene ID: 107589, updated on 12-Aug-2019

Summary

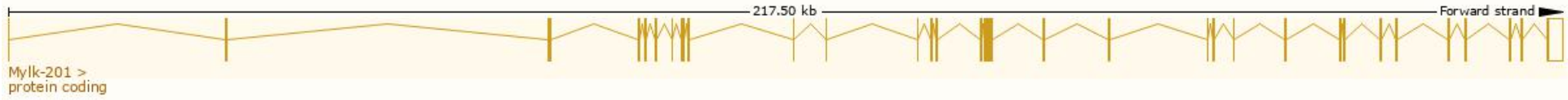
Official Symbol	Mylk provided by MGI
Official Full Name	myosin, light polypeptide kinase provided by MGI
Primary source	MGI:MGI:894806
See related	Ensembl:ENSMUSG00000022836
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	KRP; Mlck; nmMlck; smMLCK; MLCK108; MLCK210; AW489456; 9530072E15Rik; A930019C19Rik
Expression	Biased expression in bladder adult (RPKM 454.5), large intestine adult (RPKM 64.4) and 8 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

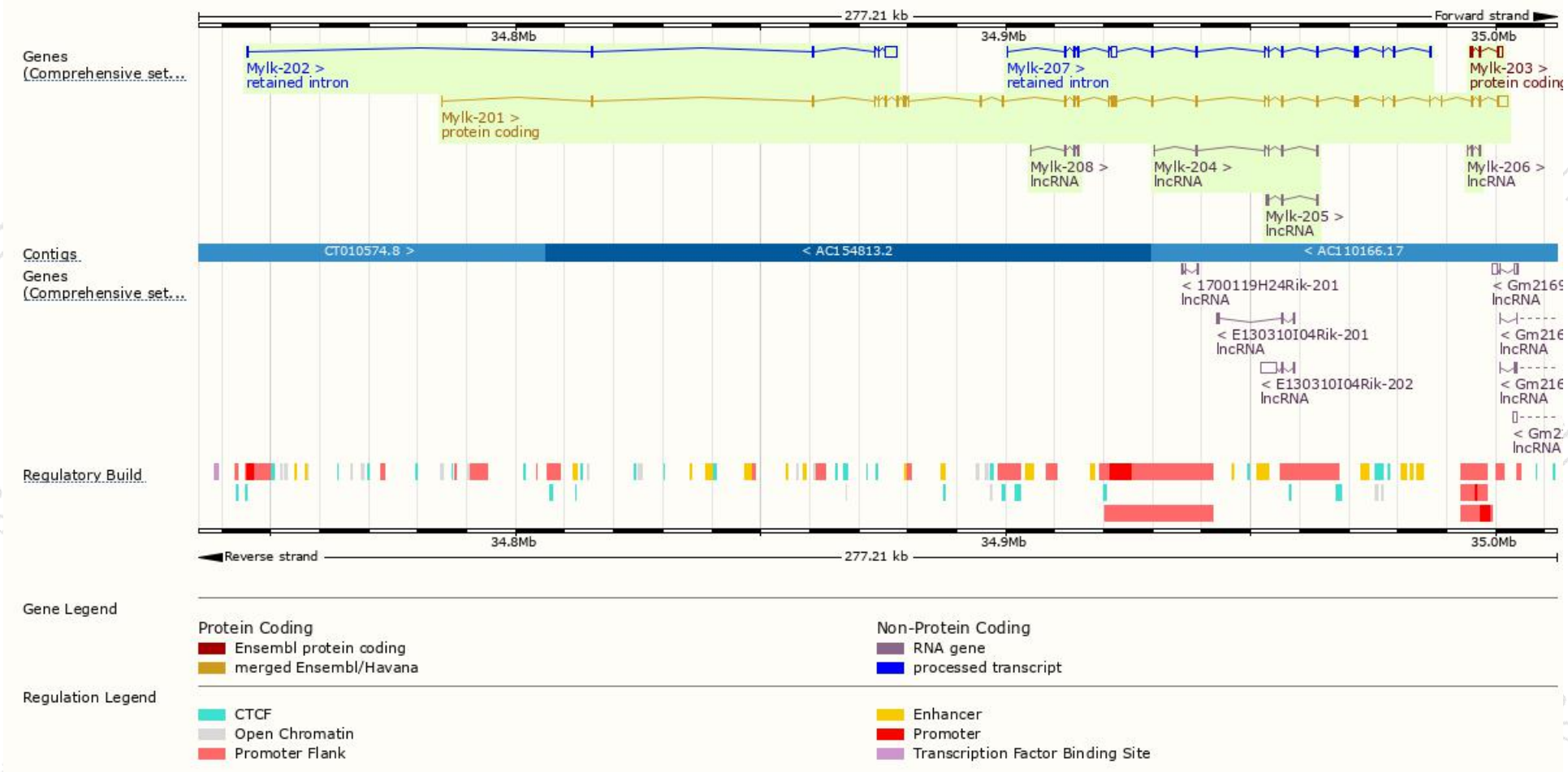
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mylk-201	ENSMUST00000023538.8	7824	1950aa	Protein coding	CCDS37320	B1B1A8	TSL:1 GENCODE basic APPRIS P1
Mylk-203	ENSMUST00000231589.1	1620	159aa	Protein coding	-	Q6PDN3	GENCODE basic
Mylk-207	ENSMUST00000232482.1	3730	No protein	Retained intron	-	-	-
Mylk-202	ENSMUST00000155268.1	3077	No protein	Retained intron	-	-	TSL:1
Mylk-204	ENSMUST00000231998.1	801	No protein	lncRNA	-	-	-
Mylk-205	ENSMUST00000232066.1	688	No protein	lncRNA	-	-	-
Mylk-208	ENSMUST00000232541.1	619	No protein	lncRNA	-	-	-
Mylk-206	ENSMUST00000232477.1	393	No protein	lncRNA	-	-	-

The strategy is based on the design of *Mylk-201* transcript,The transcription is shown below

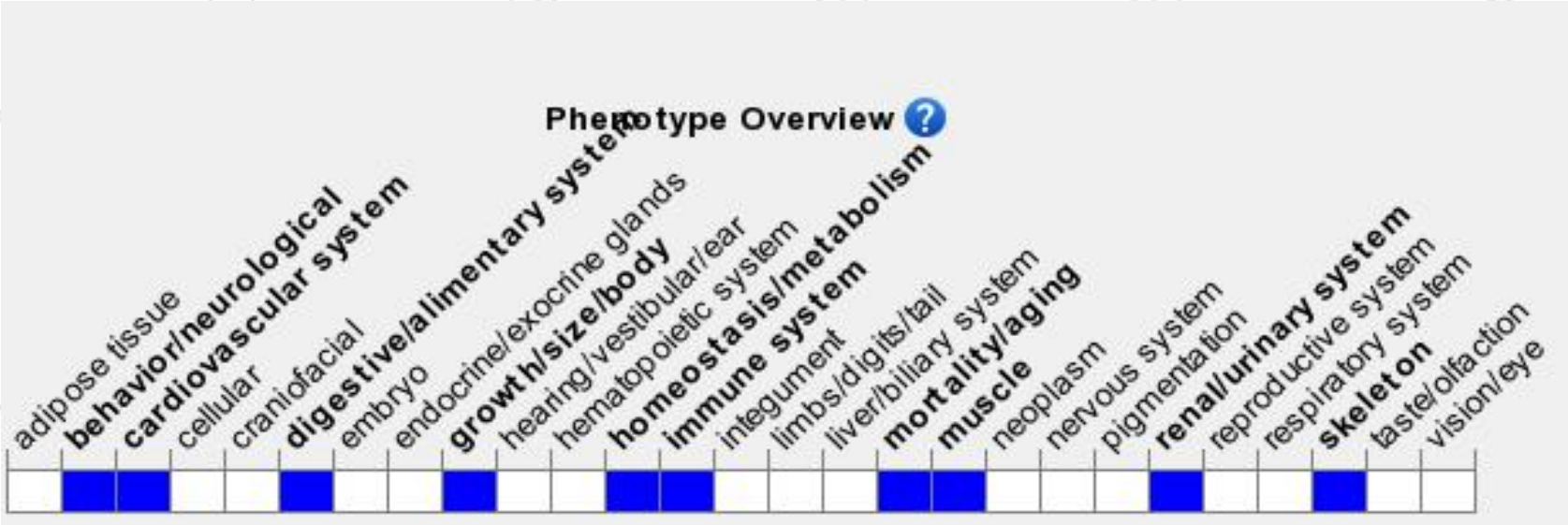


Genomic location distribution



Protein domain

Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

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If you have any questions, you are welcome to inquire.

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