

Prss8 Cas9-CKO Strategy

Designer:

Ruirui Zhang

Reviewer

Huimin Su

Design Date:

2019-8-22

Project Overview

Project Name

Prss8

Project type

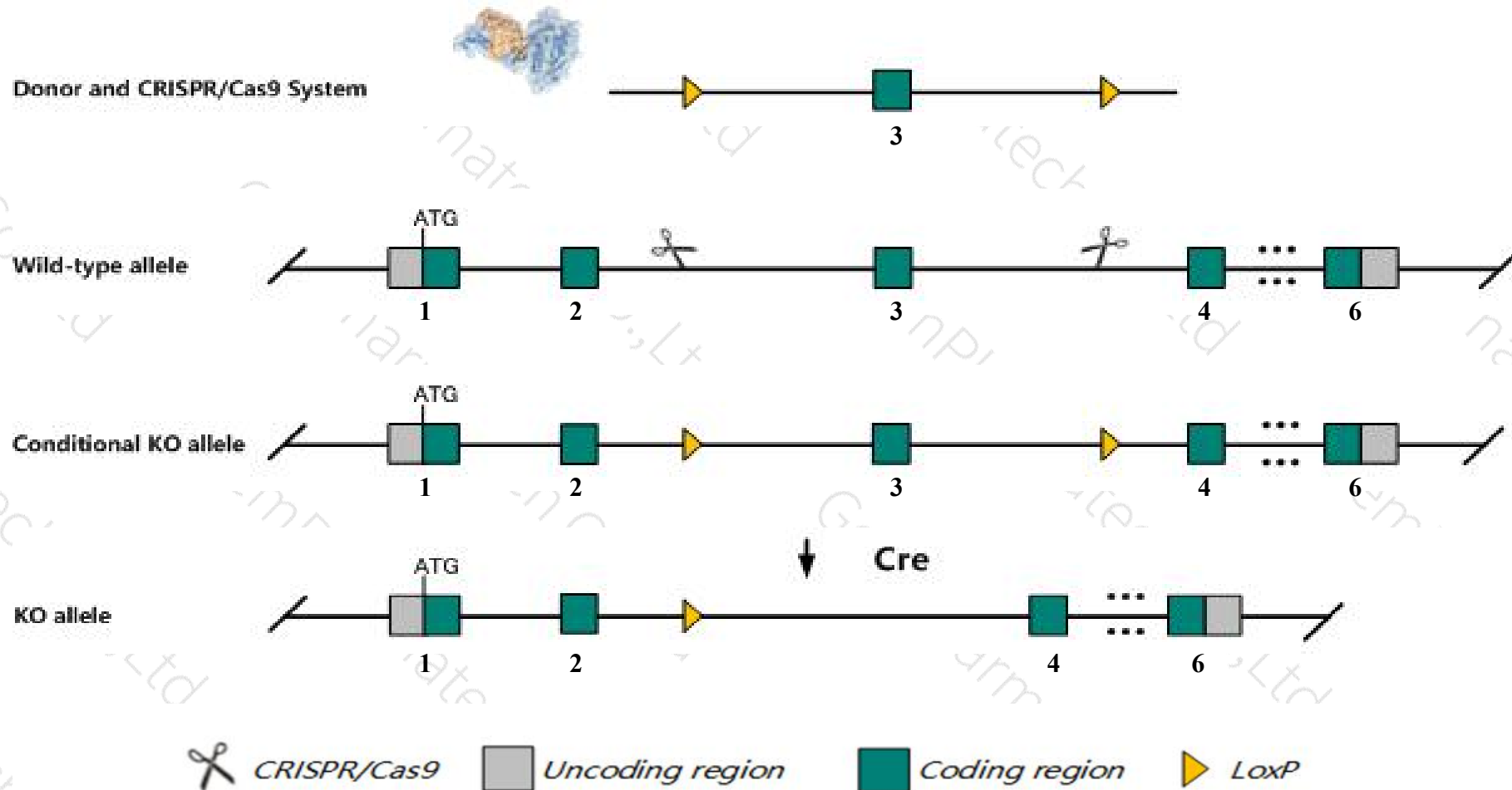
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Prss8* gene. The schematic diagram is as follows:



Technical routes

- The *Prss8* gene has 3 transcripts. According to the structure of *Prss8* gene, exon3 of *Prss8-201* (ENSMUST00000032988.9) transcript is recommended as the knockout region. The region contains 163bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Prss8* 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.

- According to the existing MGI data, Nullizygous mutations result in impaired skin barrier function, dehydration, and postnatal lethality.
- *Kat8* gene is about 2.0kb away from *Prss8* exon3. This strategy may affect the 3-terminal regulation function of *Kat8* gene.
- The *Prss8* gene is located on the Chr7. 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)

Prss8 protease, serine 8 (prostasin) [*Mus musculus* (house mouse)]

Gene ID: 76560, updated on 12-Aug-2019

Summary



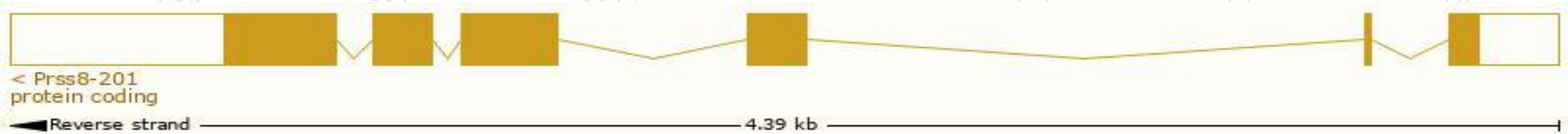
Official Symbol	Prss8 provided by MGI
Official Full Name	protease, serine 8 (prostasin) provided by MGI
Primary source	MGI:MGI:1923810
See related	Ensembl:ENSMUSG00000030800
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	fr; CAP1; mCAP1; C79772; AI313909; 2410039E18Rik
Expression	Biased expression in kidney adult (RPKM 285.2), placenta adult (RPKM 115.0) and 10 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

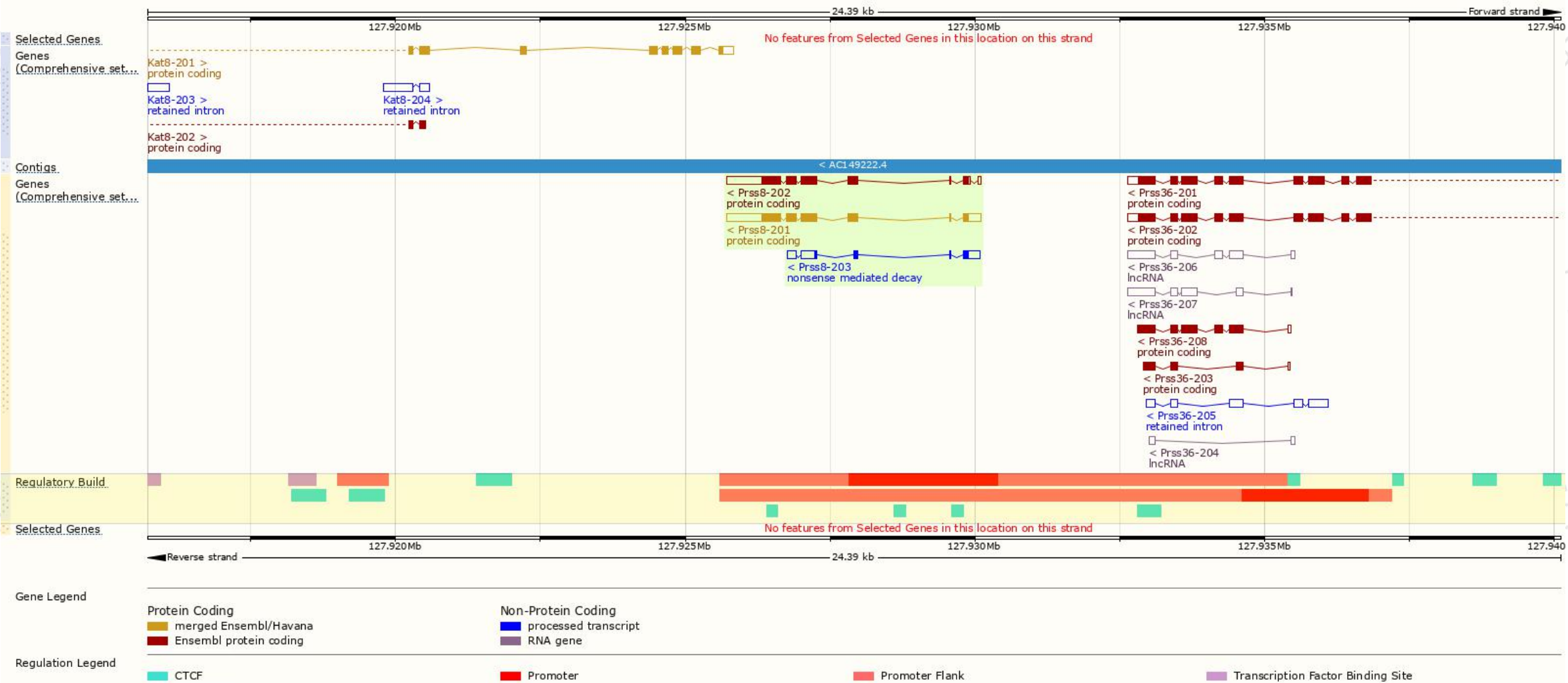
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Prss8-201	ENSMUST00000032988.9	1852	339aa	Protein coding	CCDS40147	Q99L44	TSL:1 GENCODE basic APPRIS P1
Prss8-202	ENSMUST00000206124.1	1709	339aa	Protein coding	CCDS40147	Q99L44	TSL:1 GENCODE basic APPRIS P1
Prss8-203	ENSMUST00000206568.1	795	62aa	Nonsense mediated decay	-	A0A0U1RQA6	TSL:3

The strategy is based on the design of *Prss8-201* transcript,The transcription is shown below



Genomic location distribution



Protein domain

ENSMUSP00000032...

Low complexity (Seq)

Conserved Domains

Cleavage site (Sign...

hmmpanther

PTHR24253:SF0

PTHR24253

Superfamily domains

Peptidase S1, PA clan

SMART domains

Serine proteases, trypsin domain

Prints domain

Peptidase S1A, chymotrypsin family

Pfam domain

Serine proteases, trypsin domain

PROSITE profiles

Serine proteases, trypsin domain

PROSITE patterns

Serine proteases, trypsin family, histidine active site

Serine proteases, trypsin family, serine act

Gene3D

2.40.10.10

All sequence SNPs/i....

Sequence variants (dbSNP and all other sources)

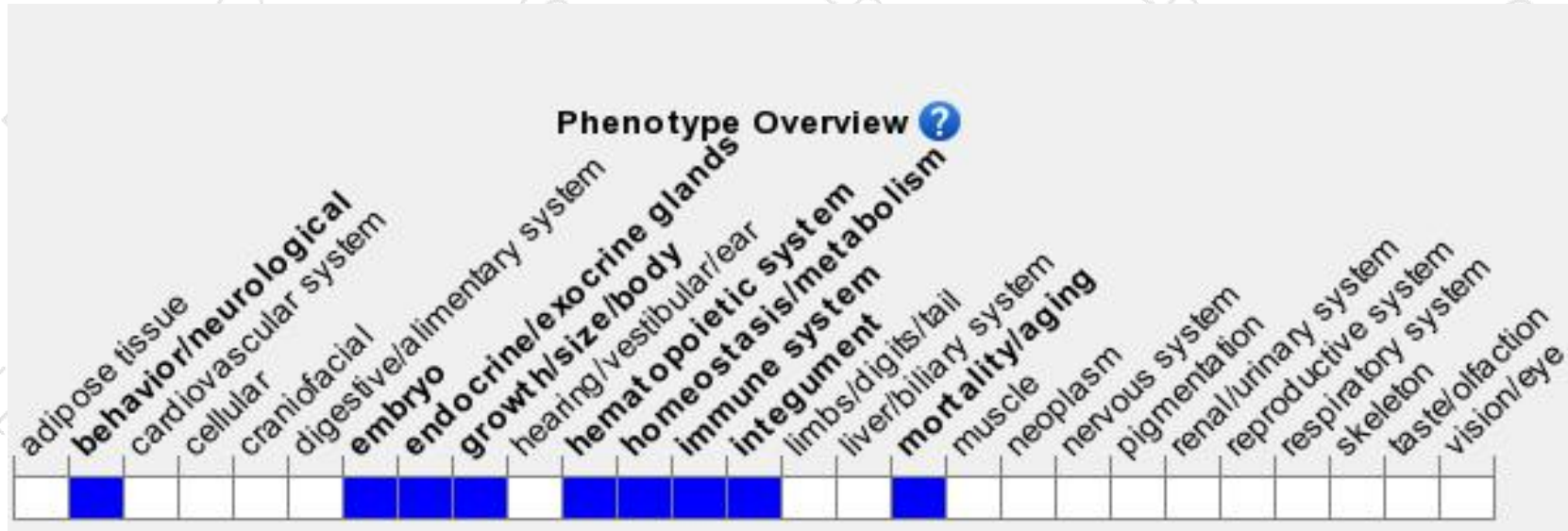
Variant Legend

missense variant
synonymous variant

Scale bar

0 40 80 120 160 200 240 280 339

Mouse phenotype description(MGI)



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

According to the existing MGI data, Nullizygous mutations result in impaired skin barrier function, dehydration, and postnatal lethality.

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

Tel: 400-9660890

