

***Rnf6* Cas9-CKO Strategy**

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Project Overview

Project Name

Rnf6

Project type

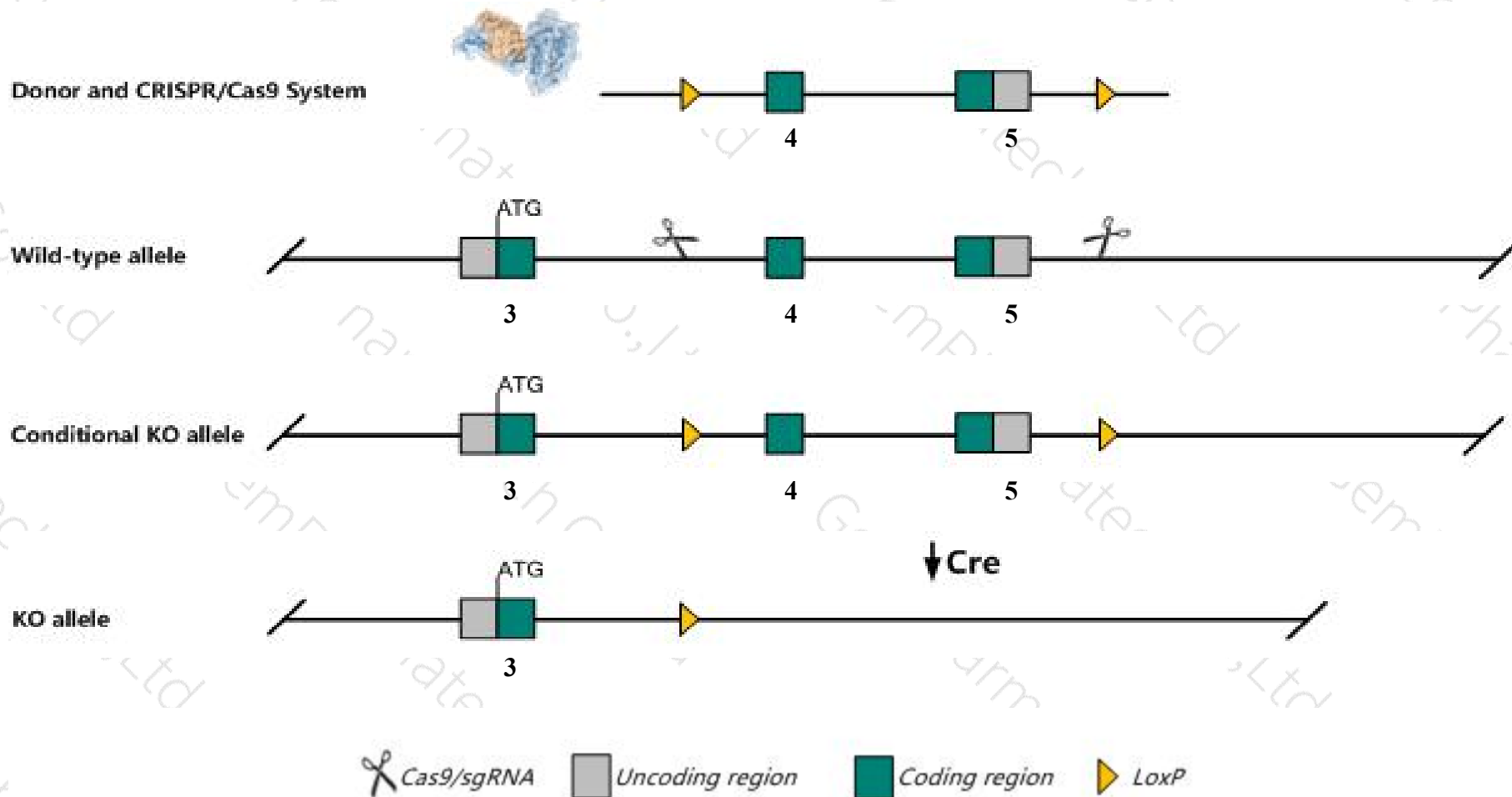
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rnf6* gene. The schematic diagram is as follows:



Technical routes

- The *Rnf6* gene has 8 transcripts. According to the structure of *Rnf6* gene, exon4-exon5 of *Rnf6*-206(ENSMUST00000161859.7) transcript is recommended as the knockout region. The region contains 1826bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rnf6* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was 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.

- The *Rnfb6* gene is located on the Chr5. 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)

Rnf6 ring finger protein (C3H2C3 type) 6 [Mus musculus (house mouse)]

Gene ID: 74132, updated on 13-Mar-2020

Summary



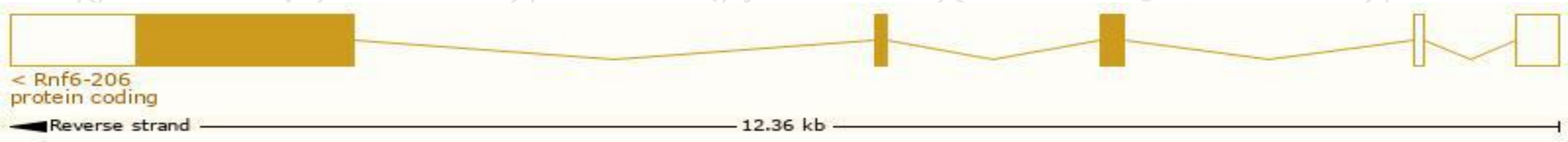
Official Symbol	Rnf6 provided by MGI
Official Full Name	ring finger protein (C3H2C3 type) 6 provided by MGI
Primary source	MGI:MGI:1921382
See related	Ensembl:ENSMUSG00000029634
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1200013I08Rik, 5730419H05Rik, AA537053
Expression	Ubiquitous expression in bladder adult (RPKM 14.8), cerebellum adult (RPKM 13.4) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

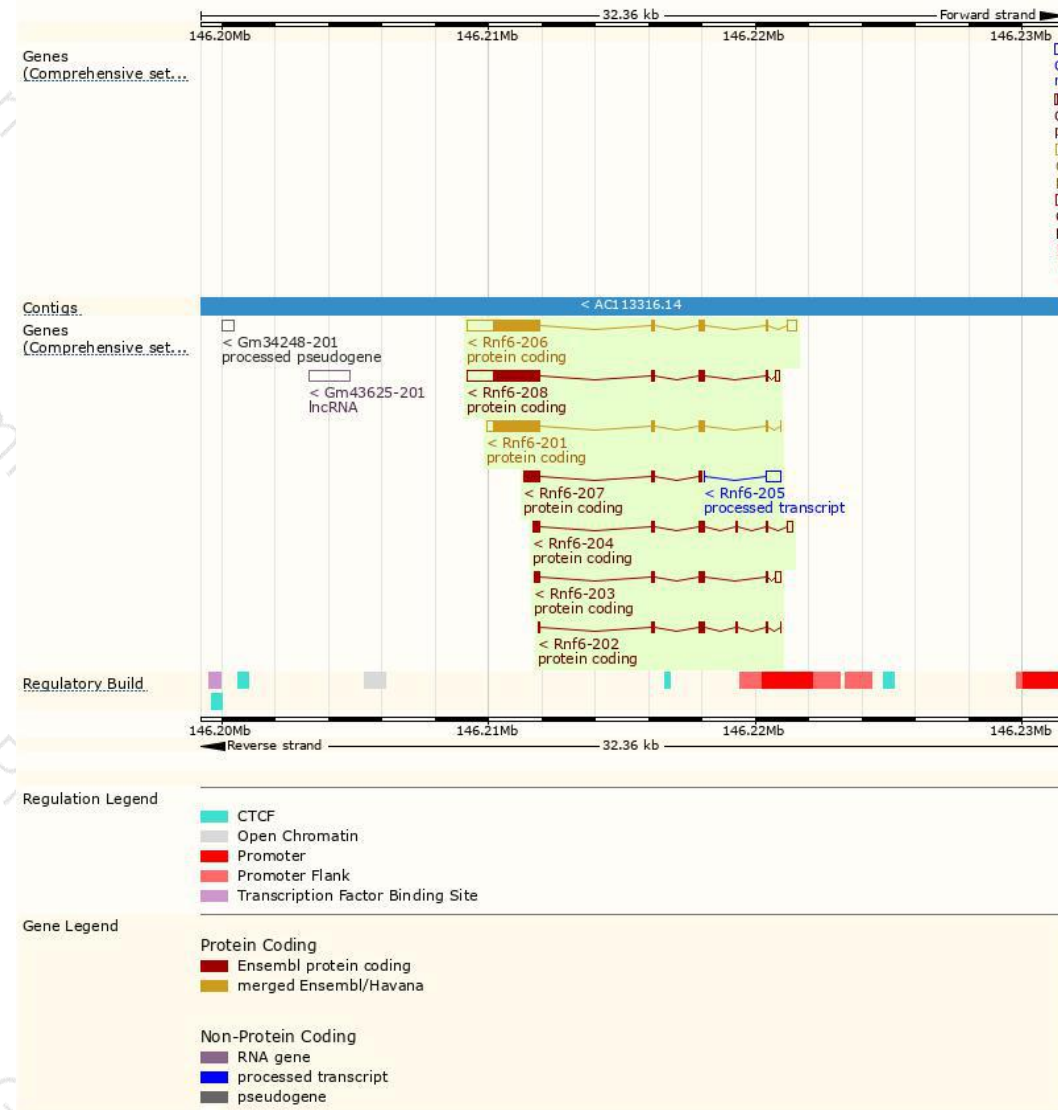
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rnf6-206	ENSMUST00000161859.7	3452	667aa	Protein coding	CCDS19868	Q9DBU5	TSL:1 GENCODE basic APPRIS P1
Rnf6-208	ENSMUST00000169407.8	3276	667aa	Protein coding	CCDS19868	Q9DBU5	TSL:2 GENCODE basic APPRIS P1
Rnf6-201	ENSMUST00000067837.9	2388	667aa	Protein coding	CCDS19868	Q9DBU5	TSL:1 GENCODE basic APPRIS P1
Rnf6-204	ENSMUST00000161574.7	909	188aa	Protein coding	-	E0CYZ9	CDS 3' incomplete TSL:5
Rnf6-207	ENSMUST00000162219.1	812	271aa	Protein coding	-	F6VG71	CDS 5' and 3' incomplete TSL:3
Rnf6-203	ENSMUST00000161331.7	802	167aa	Protein coding	-	E0CZ82	CDS 3' incomplete TSL:2
Rnf6-202	ENSMUST00000159074.2	542	129aa	Protein coding	-	E0CYS7	CDS 3' incomplete TSL:3
Rnf6-205	ENSMUST00000161725.1	563	No protein	Processed transcript	-	-	TSL:1

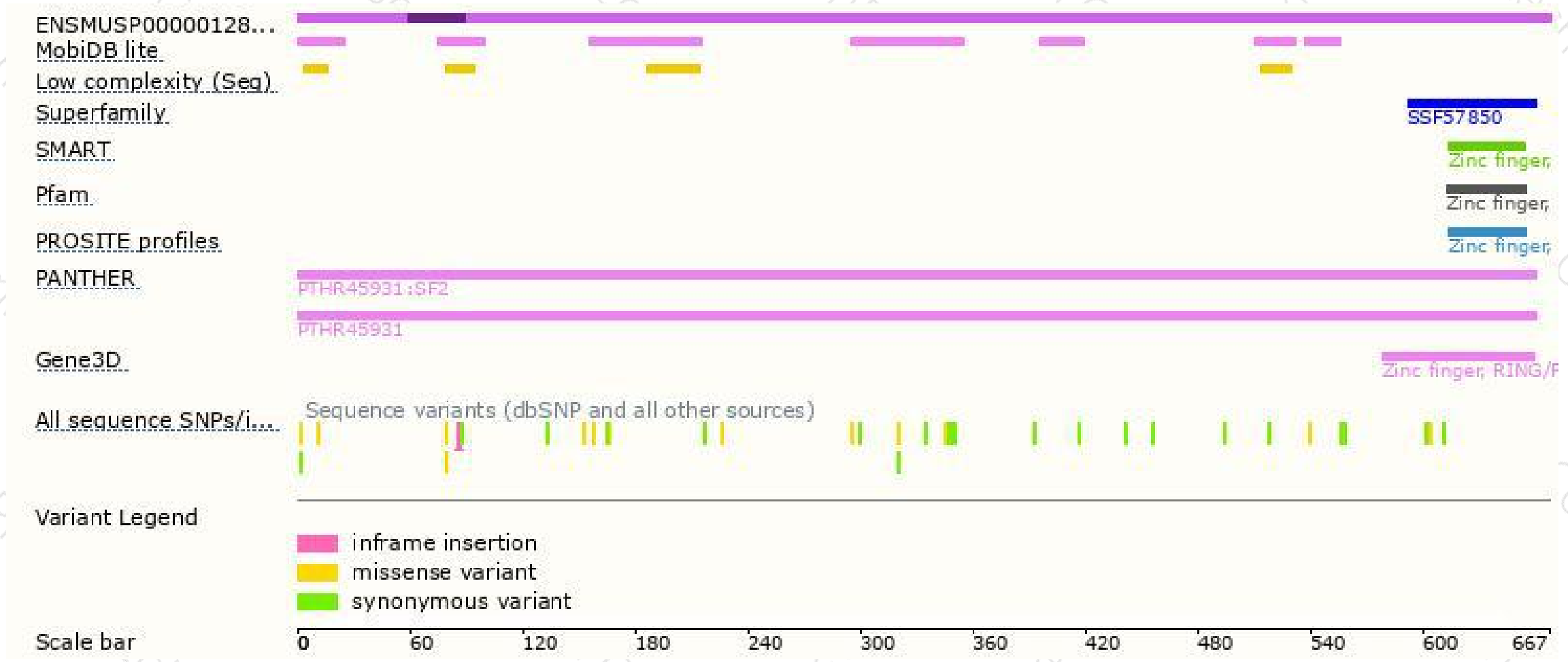
The strategy is based on the design of *Rnf6-206* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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