

***Rab3b* Cas9-CKO Strategy**

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Reviewer:

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Project Overview

Project Name

Rab3b

Project type

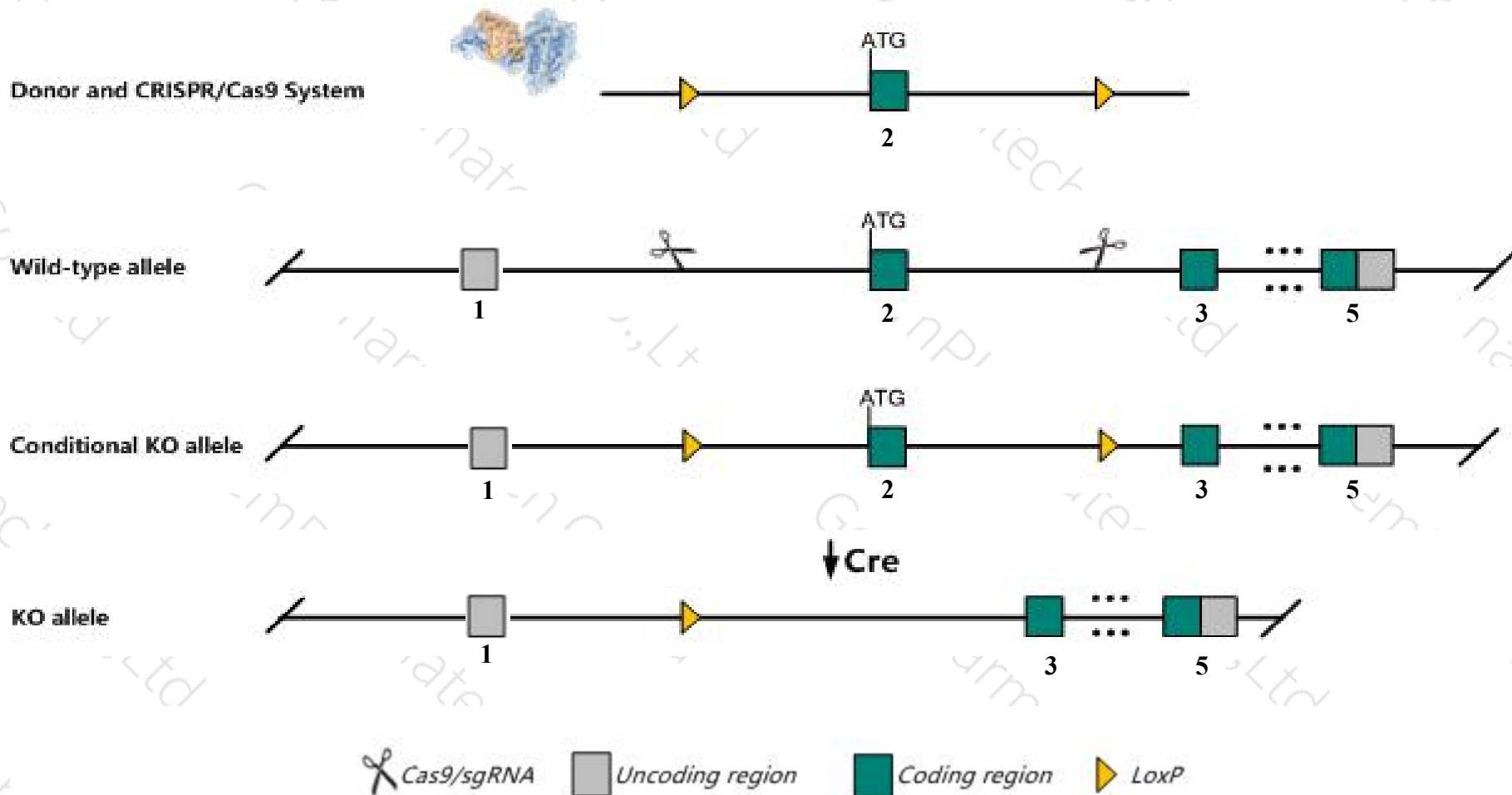
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rab3b* gene. The schematic diagram is as follows:



- The *Rab3b* gene has 4 transcripts. According to the structure of *Rab3b* gene, exon2 of *Rab3b-202* (ENSMUST00000106650.8) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rab3b* 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, Mice homozygous for a knock-out allele are viable with no apparent decrease in weight or fertility.
- The insertion of 5'Loxp may affect the 5-terminal regulation of *Rab3b-201* transcript.
- The *Rab3b* gene is located on the Chr4. 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)

Rab3b RAB3B, member RAS oncogene family [*Mus musculus* (house mouse)]

Gene ID: 69908, updated on 12-Aug-2019

Summary

Official Symbol	Rab3b provided by MGI
Official Full Name	RAB3B, member RAS oncogene family provided by MGI
Primary source	MGI:MGI:1917158
See related	Ensembl:ENSMUSG00000003411
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2610528C18Rik
Expression	Biased expression in CNS E18 (RPKM 19.6), frontal lobe adult (RPKM 19.1) and 14 other tissues See more
Orthologs	human all

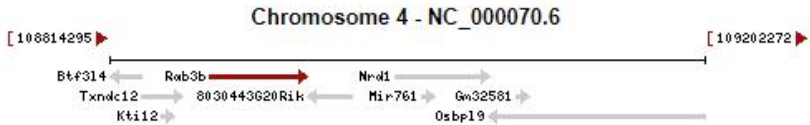
Genomic context

Location: 4; 4 C7

See Rab3b in [Genome Data Viewer](#)

Exon count: 5

Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	4	NC_000070.6 (108879070..108943324)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	4	NC_000070.5 (108551675..108615929)

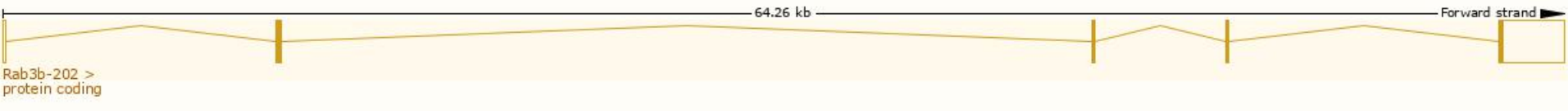


Transcript information (Ensembl)

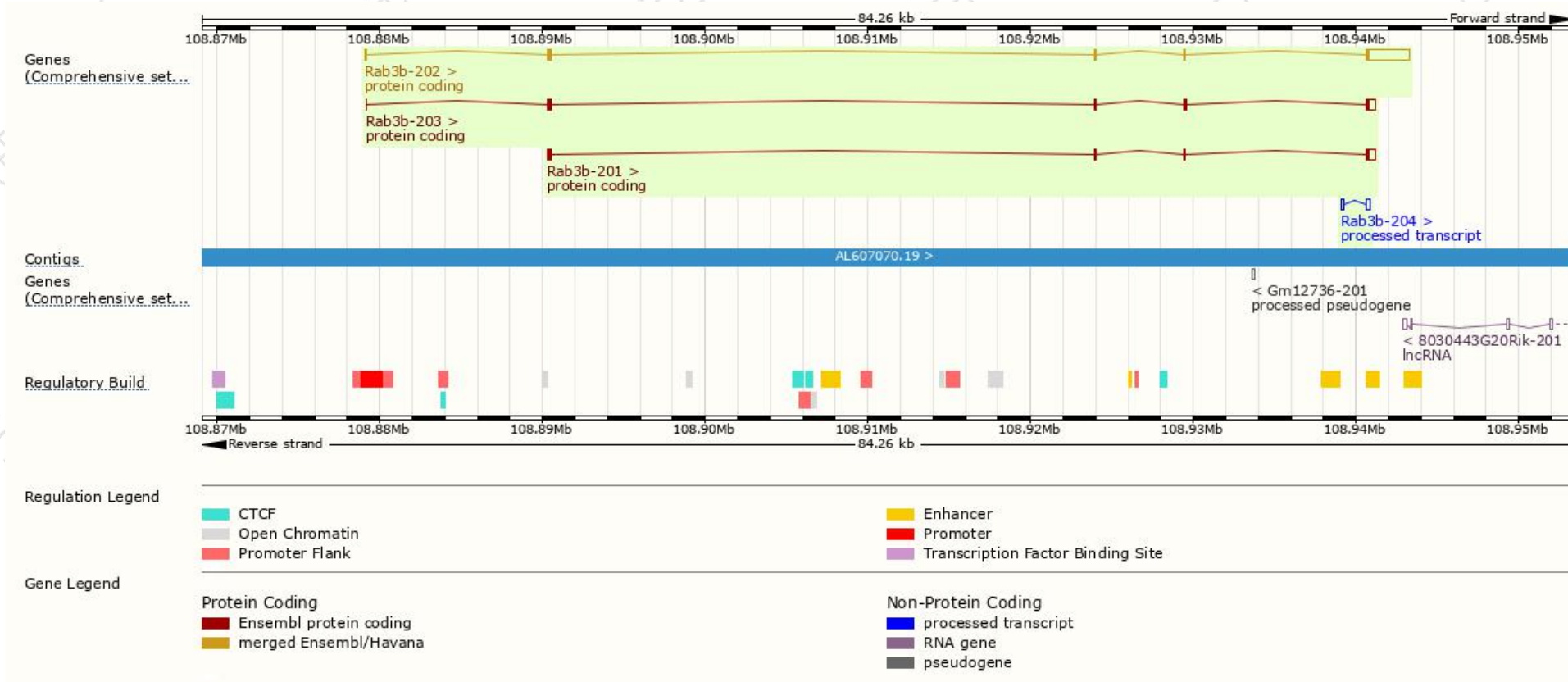
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rab3b-202	ENSMUST00000106650.8	3327	219aa	Protein coding	CCDS18459	Q0PD62 Q9CZT8	TSL:1 GENCODE basic APPRIS P1
Rab3b-201	ENSMUST000000003502.3	1053	219aa	Protein coding	CCDS18459	Q0PD62 Q9CZT8	TSL:1 GENCODE basic APPRIS P1
Rab3b-203	ENSMUST00000106651.8	1078	228aa	Protein coding	-	A2A7Z6	TSL:5 GENCODE basic
Rab3b-204	ENSMUST00000157062.1	432	No protein	Processed transcript	-	-	TSL:3

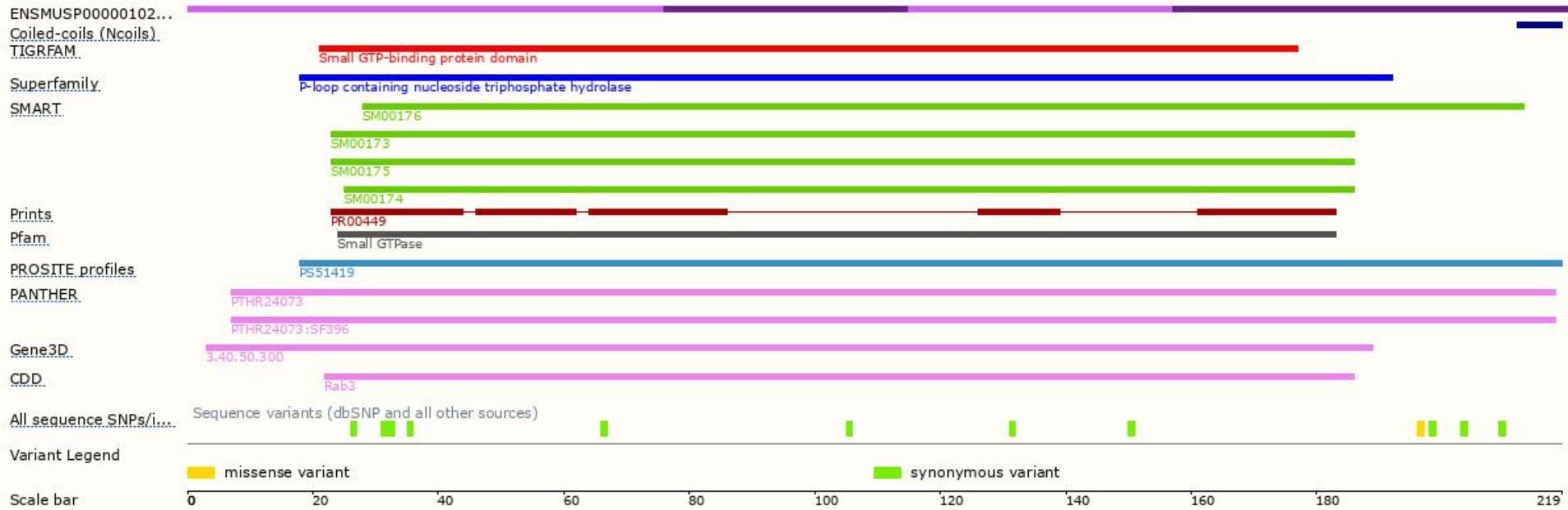
The strategy is based on the design of *Rab3b-202* transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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