

Rph3a Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Rph3a

Project type

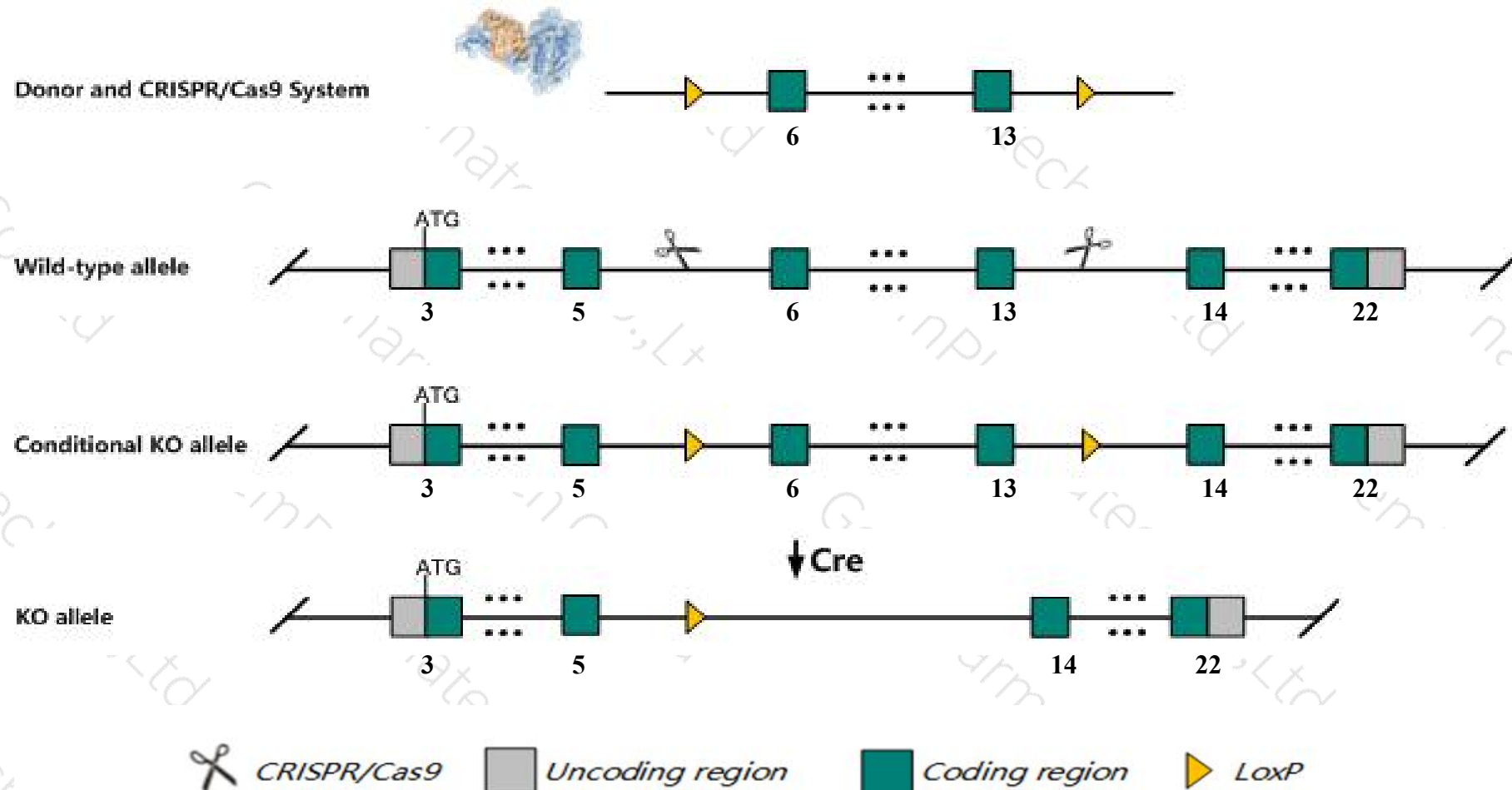
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rph3a* gene. The schematic diagram is as follows:



- The *Rph3a* gene has 4 transcripts. According to the structure of *Rph3a* gene, exon6-exon13 of *Rph3a-201* (ENSMUST00000079204.8) transcript is recommended as the knockout region. The region contains 914bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rph3a* 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, Homozygous mutants are viable and fertile and do not exhibit any obvious abnormal phenotypes.
- Some amino acids will remain at the N-terminus and some functions may be retained.
- The *Rph3a* 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)

Rph3a rabphilin 3A [Mus musculus (house mouse)]

Gene ID: 19894, updated on 19-Mar-2019

Summary



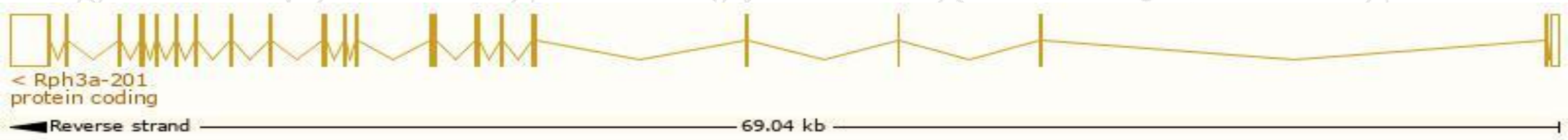
Official Symbol	Rph3a provided by MGI
Official Full Name	rabphilin 3A provided by MGI
Primary source	MGI:MGI:102788
See related	Ensembl:ENSMUSG00000029608
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2900002P20Rik, AU022689, AW108370
Expression	Biased expression in cerebellum adult (RPKM 64.9), cortex adult (RPKM 57.6) and 4 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

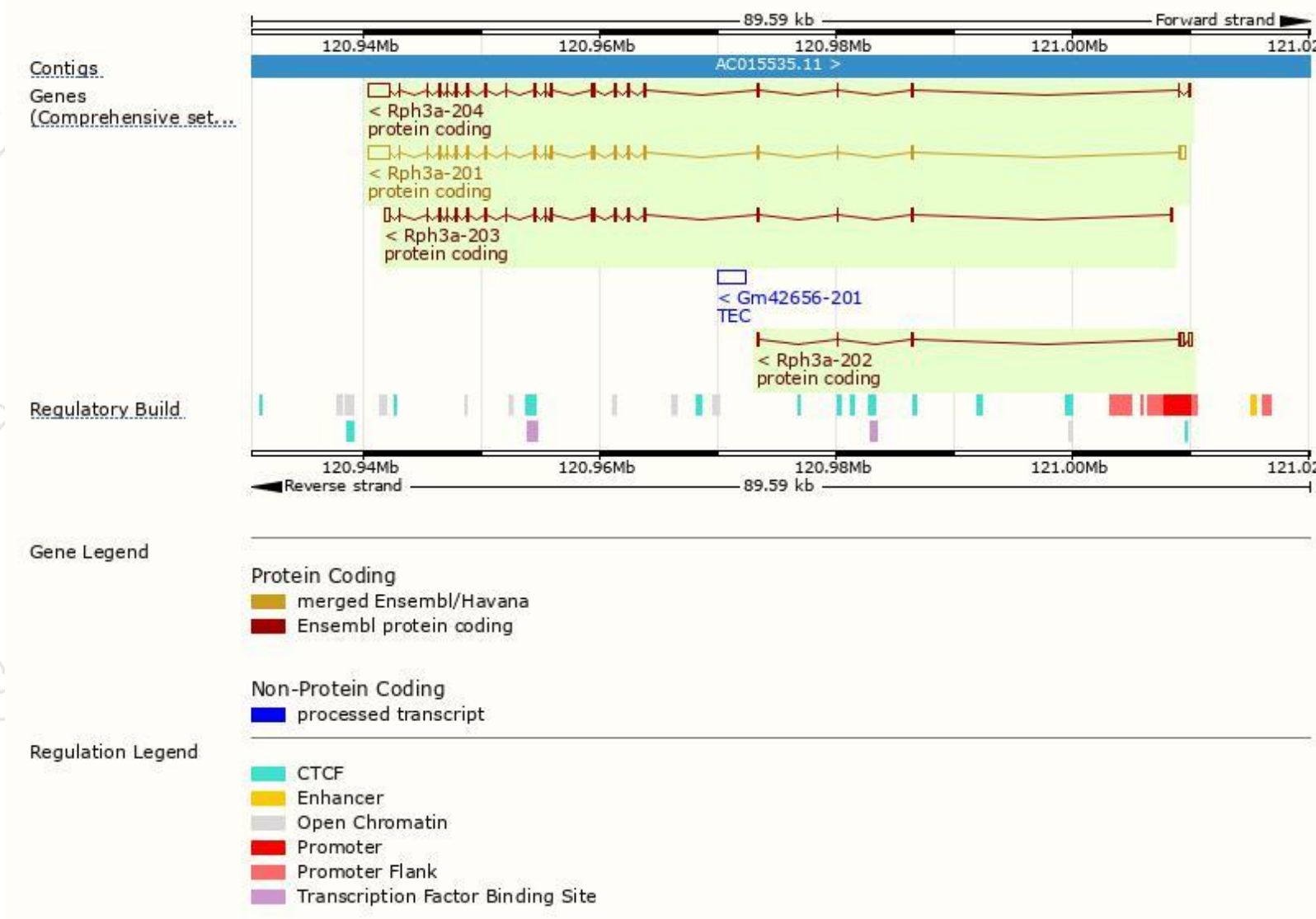
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rph3a-201	ENSMUST00000079204.8	4150	681aa	Protein coding	CCDS39246	P47708	TSL:1 GENCODE basic APPRIS P1
Rph3a-204	ENSMUST00000202406.3	4096	681aa	Protein coding	CCDS39246	P47708	TSL:1 GENCODE basic APPRIS P1
Rph3a-203	ENSMUST00000202326.3	2621	681aa	Protein coding	CCDS39246	P47708	TSL:1 GENCODE basic APPRIS P1
Rph3a-202	ENSMUST00000200792.1	750	49aa	Protein coding	-	A0A0J9YV12	CDS 3' incomplete TSL:3

The strategy is based on the design of *Rph3a-201* 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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