

Rmnd5a Cas9-CKO Strategy

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Overview

Target Gene Name

• *Rmnd5a*

Project Type

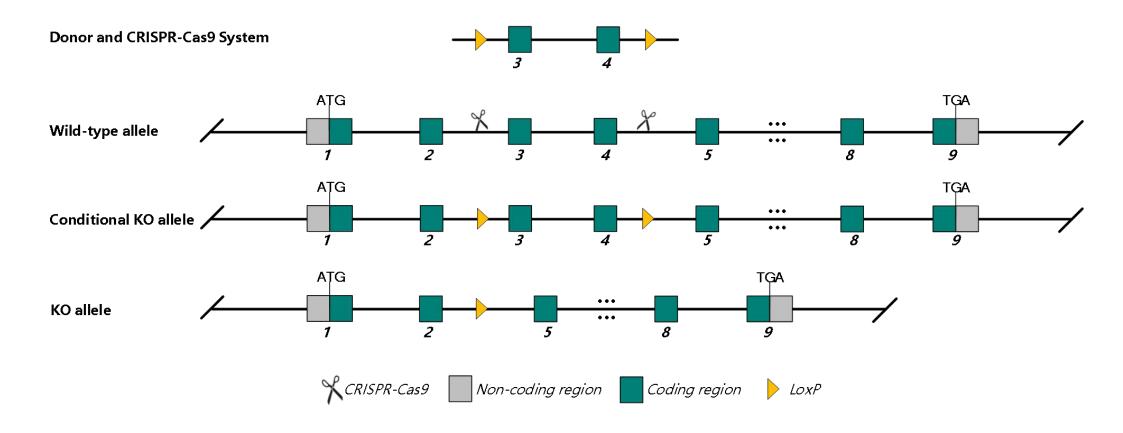
• Cas9-CKO

Genetic Background

• C57BL/6JGpt



Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the *Rmnd5a* gene.



Technical Information

- The *Rmnd5a* gene has 5 transcripts. According to the structure of *Rmnd5a* gene, exon 3-4 of *Rmnd5a*-201 (ENSMUST00000002292.15) is recommended as the knockout region. The region contains 236 bp of coding sequence. Knocking out the region will result in disruption of gene function.
- In this project we use CRISPR-Cas9 technology to modify *Rmnd5a* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.



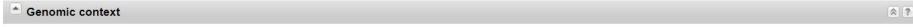
Gene Information

Rmnd5a required for meiotic nuclear division 5 homolog A [Mus musculus (house mouse)]

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Gene ID: 68477, updated on 7-Sep-2023





Location: 6 C1; 6 32.14 cM See Rmnd5a in Genome Data Viewer

Exon count: 9

https://www.ncbi.nlm.nih.gov/gene/68477

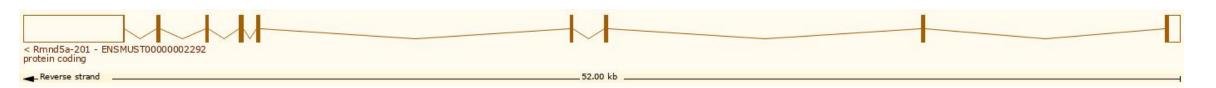


Transcript Information

The gene has 5 transcripts, all transcripts are shown below:

Show/hide columns (1 hidden)							Filter	
Transcript ID 👙	Name 🌲	bp 🌲	Protein ▼	Biotype	CCDS .	UniProt Match 🍦	Flags	
ENSMUST00000002292.15	Rmnd5a-201	6165	<u>391aa</u>	Protein coding	CCDS20231 €	Q80YQ8@	Ensembl Canonical GENCODE basic APPRIS P1 TSL:1	
ENSMUST00000149415.2	Rmnd5a-205	3768	218aa	Protein coding		F6V8A7₽	TSL:1 CDS 5' incomplete	
ENSMUST00000144081.8	Rmnd5a-204	3086	No protein	Protein coding CDS not defined		22	TSL:1	
ENSMUST00000140696.2	Rmnd5a-203	999	No protein	Protein coding CDS not defined		. 1551	TSL:3	
ENSMUST00000123947.2	Rmnd5a-202	667	No protein	Retained intron		(16)	TSL:2	

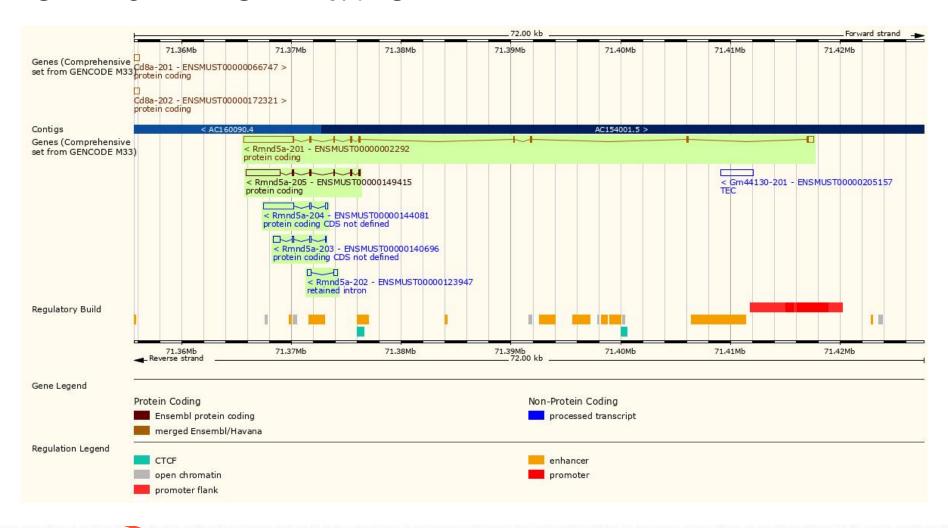
The strategy is based on the design of *Rmnd5a*-201 transcript, the transcription is shown below:





Source: http://asia.ensembl.org/

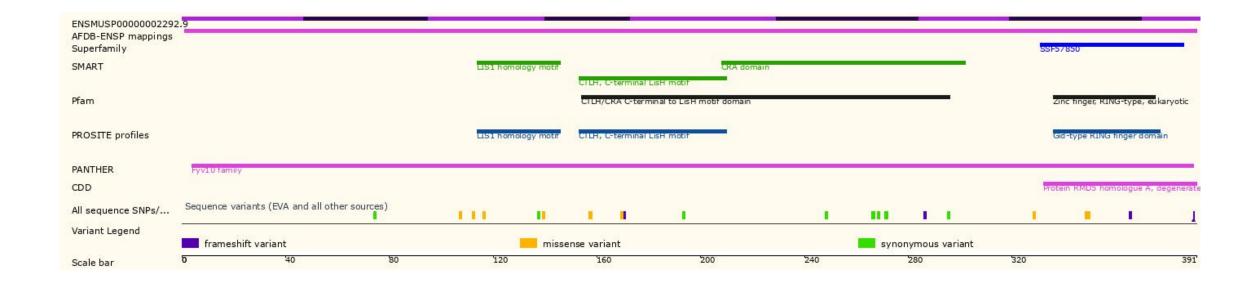
Genomic Information





Source: http://asia.ensembl.org/

Protein Information





Source: https://www.ensembl.org

Important Information

- Since the *Rmnd5a*-205 transcript 5' is incomplete, the effect of this stratergy on *Rmnd5a*-205 transcript is unknown.
- This stratergy may not affect Rmnd5a-202, Rmnd5a-203 and Rmnd5a-204 transcript.
- *Rmnd5a* is located on Chr 6. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.

