

Rnf31 Cas9-KO Strategy

Designer:Xueting Zhang Reviewer:Yanhua Shen Design Date:2019-8-28

Project Overview



Project Name

Rnf31

Project type

Cas9-KO

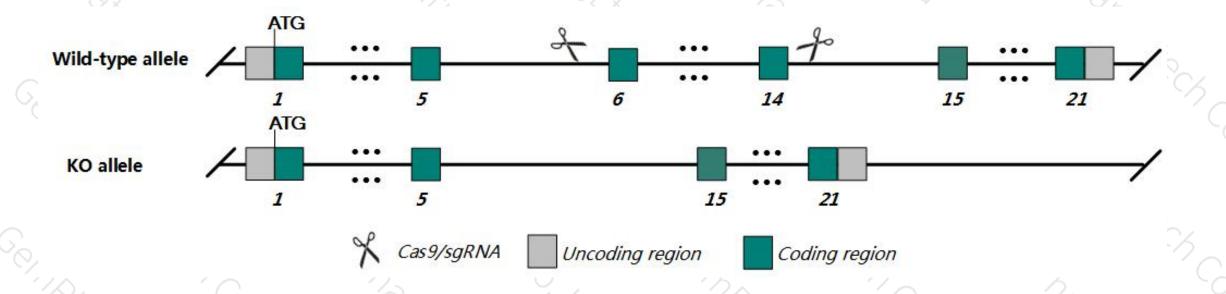
Strain background

C57BL/6JGpt

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rnf31* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Rnf31* gene has 7 transcripts. According to the structure of *Rnf31* gene, exon6-exon14 of *Rnf31-201* (ENSMUST00000019443.14) transcript is recommended as the knockout region. The region contains 1853bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Rnf31* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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.

Notice



- ➤ According to the existing MGI data, Mice homozygous for a knock-out allele exhibit complete embryonic lethality. Mice homozygous for a conditional allele activated in B cells exhibit severely impaired B1 B cell development and impaired antibody responses to both T cell-dependent and T cell-independent type 2 antigens.
- > Transcript *Rnf31-207* may not be affected.
- The knockout region is near to the N-terminal of *Psme2/Emc9/Irf9* gene, this strategy may influence the regulatory function of the N-terminal of these genes.
- > The N-terminal of *Rnf31* gene will remain 207aa, it may remain the partial function of *Rnf31* gene.
- The *Rnf31* gene is located on the Chr14. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Rnf31 ring finger protein 31 [Mus musculus (house mouse)]

Gene ID: 268749, updated on 12-Aug-2019

Summary

△ ?

Official Symbol Rnf31 provided by MGI

Official Full Name ring finger protein 31 provided by MGI

Primary source MGI:MGI:1934704

See related Ensembl: ENSMUSG00000047098

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 HOIP; Paul; AL033293; BC031509; Flj10111; mFLJ00217

Expression Ubiquitous expression in thymus adult (RPKM 23.8), spleen adult (RPKM 23.0) and 28 other tissues See more

Orthologs human all

Genomic context

☆ ?

Location: 14; 14 C3

See Rnf31 in Genome Data Viewer

Exon count: 21

Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	14	NC_000080.6 (5559171355603674)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	14	NC_000080.5 (5621062756222508)

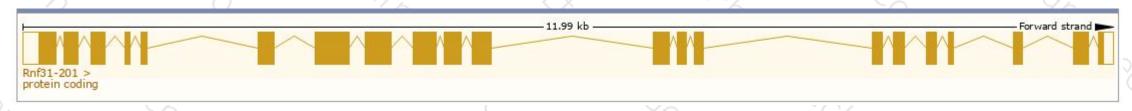
Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

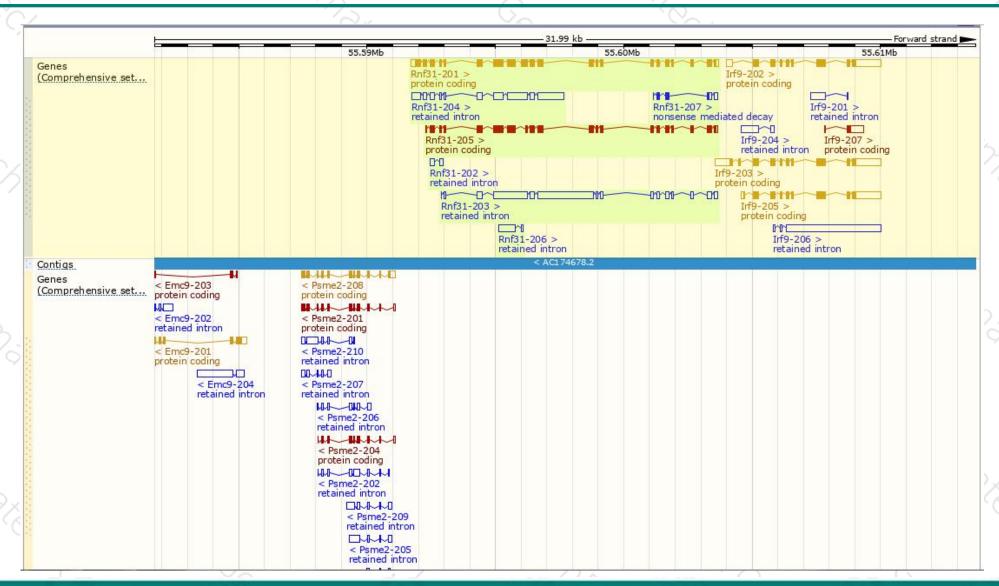
Name 🍦	Transcript ID A	bp 🍦	Protein 🍦	Biotype	CCDS	UniProt 🍦	Flags
Rnf31-201	ENSMUST00000019443.14	3485	<u>1066aa</u>	Protein coding	CCDS27118@	Q924T7₽	TSL:1 GENCODE basic APPRIS P1
Rnf31-202	ENSMUST00000126544.1	266	No protein	Retained intron	-8	-	TSL:1
Rnf31-203	ENSMUST00000133903.2	4939	No protein	Retained intron	-8	-	TSL:5
Rnf31-204	ENSMUST00000137296.7	3293	No protein	Retained intron	-8	-	TSL:1
Rnf31-205	ENSMUST00000140178.1	2817	<u>912aa</u>	Protein coding		F6XVP7₽	CDS 5' incomplete TSL:5
Rnf31-206	ENSMUST00000227664.1	720	No protein	Retained intron	-8	-	7-
Rnf31-207	ENSMUST00000227708.1	552	103aa	Nonsense mediated decay		A0A2I3BRY4₽	CDS 5' incomplete

The strategy is based on the design of Rnf31-201 transcript, The transcription is shown below



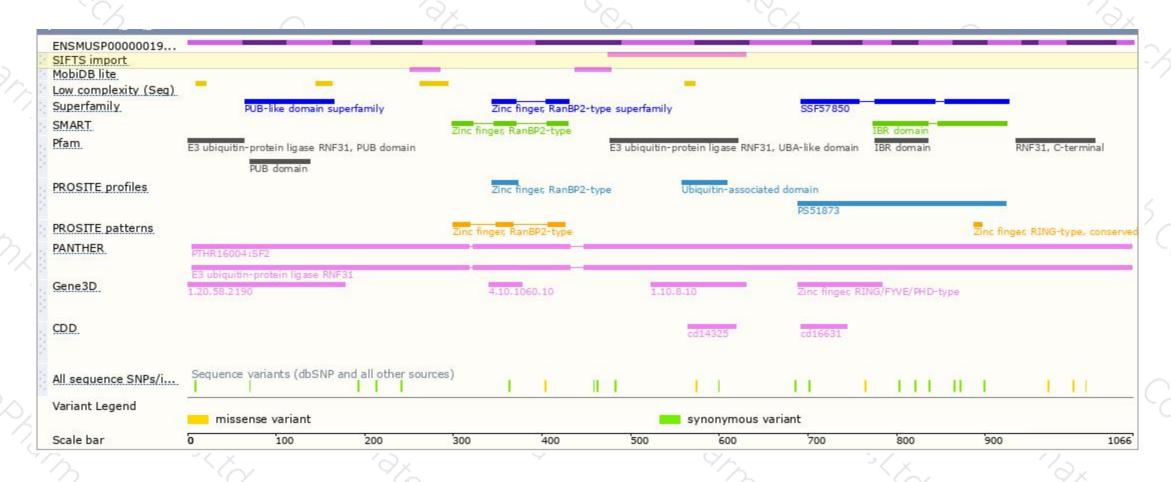
Genomic location distribution





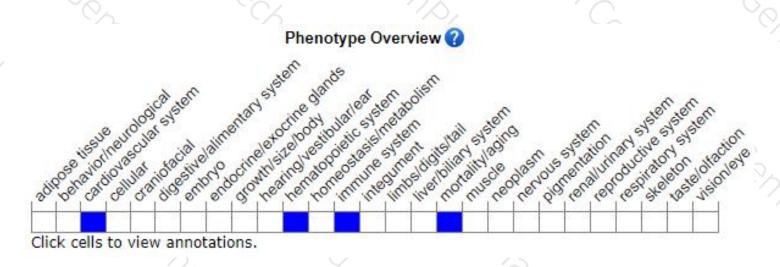
Protein domain





Mouse phenotype description(MGI)





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

Mice homozygous for a knock-out allele exhibit complete embryonic lethality. Mice homozygous for a conditional allele activated in B cells exhibit severely impaired B1 B cell development and impaired antibody responses to both T cell-dependent and T cell-independent type 2 antigens.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





