

***Rnf31* Cas9-CKO Strategy**

Designer: Xueting Zhang
Reviewer: Yanhua Shen
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Project Overview

Project Name

Rnf31

Project type

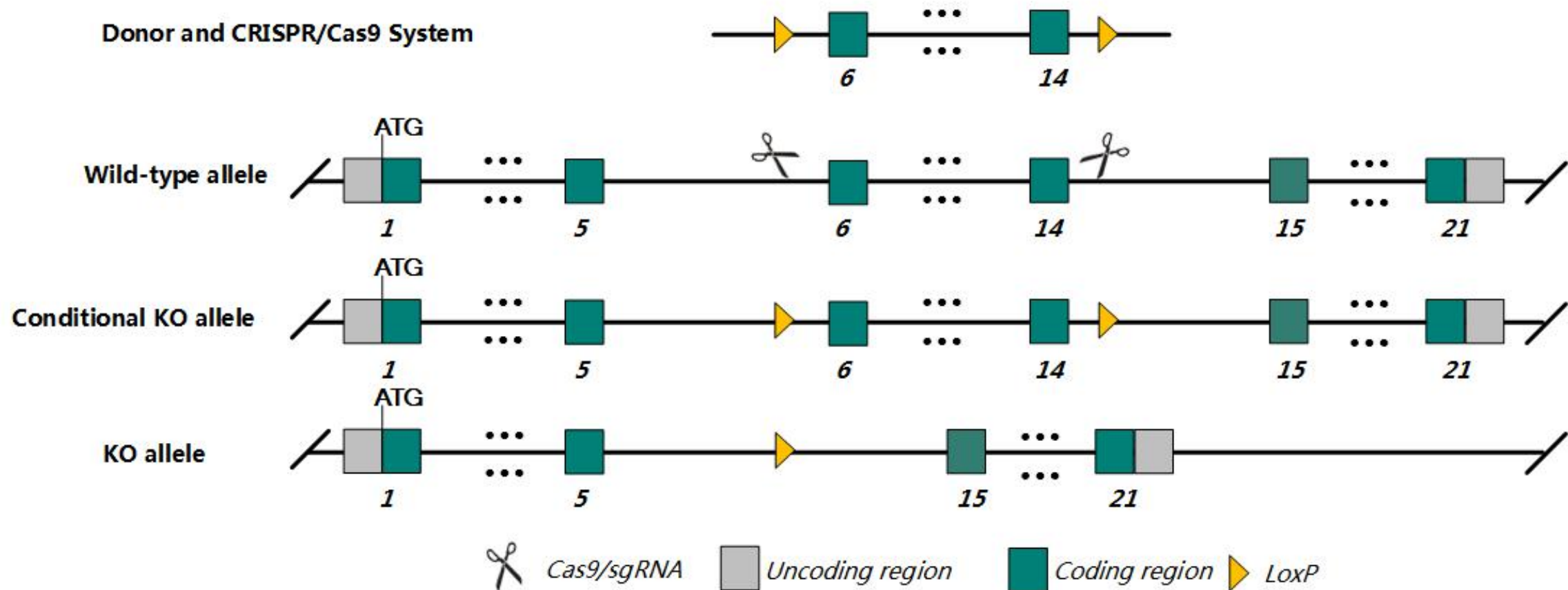
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



- 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, donor was constructed. Cas9, gRNA 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 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 floxed 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological 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

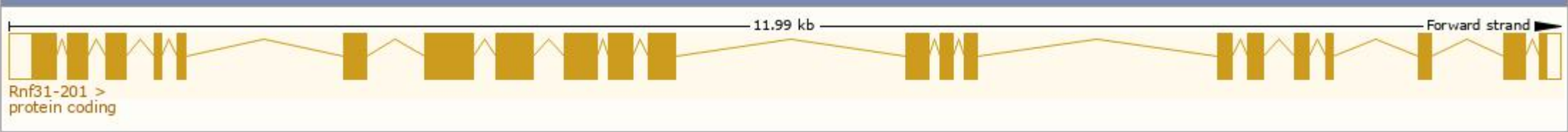
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	14	NC_000080.6 (55591713..55603674)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	14	NC_000080.5 (56210627..56222508)

Transcript information (Ensembl)

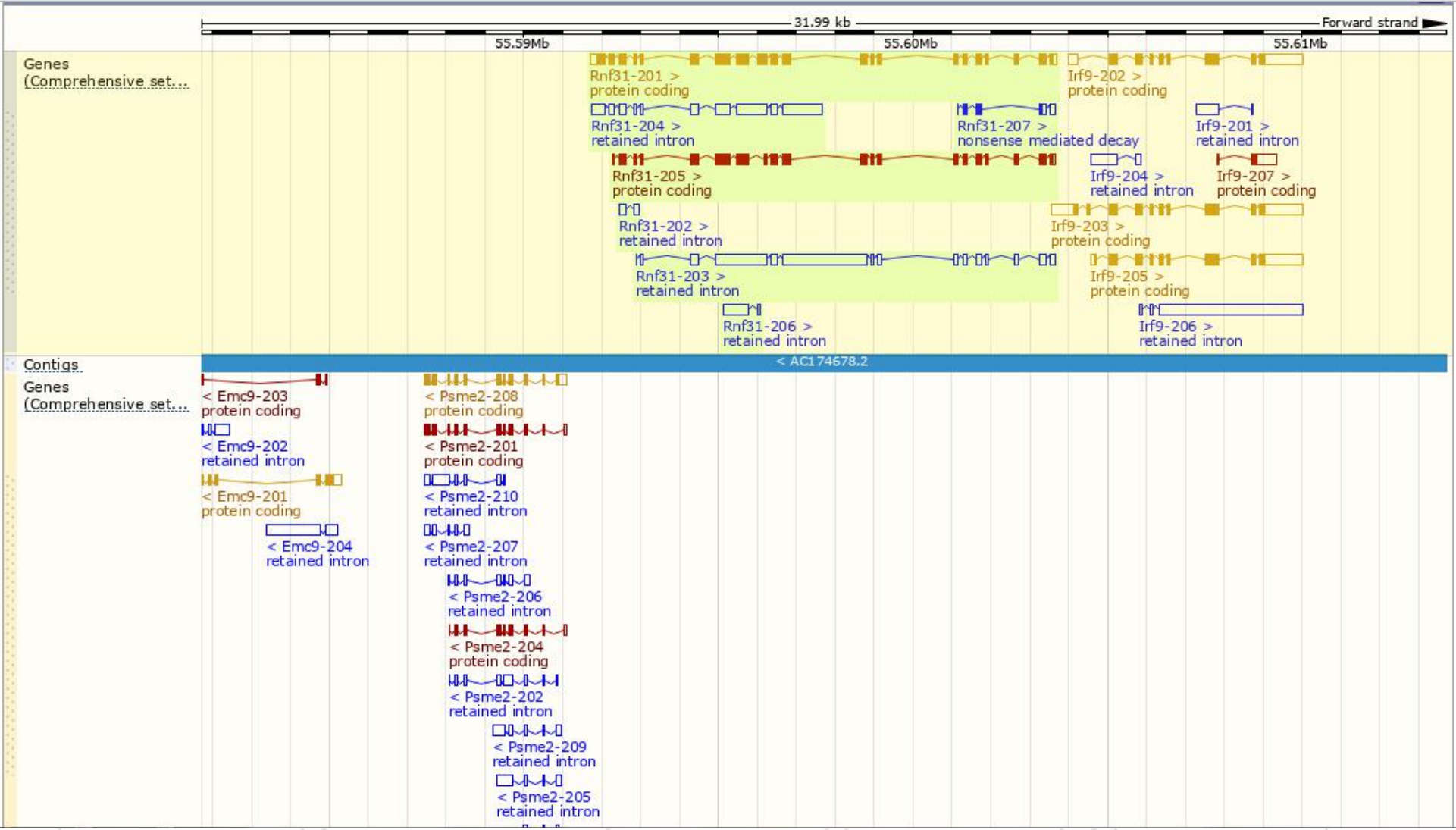
The gene has 7 transcripts,all transcripts are shown below:

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

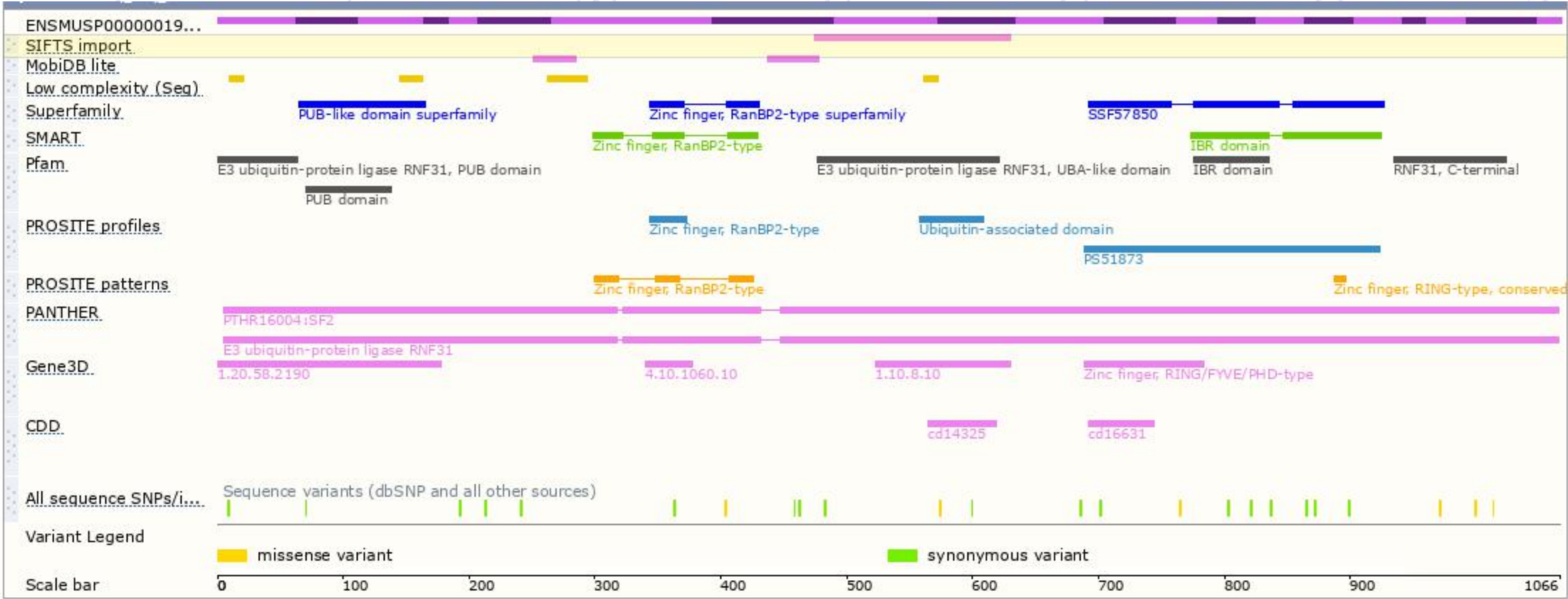
The strategy is based on the design of *Rnf31-201* transcript,The transcription is shown below



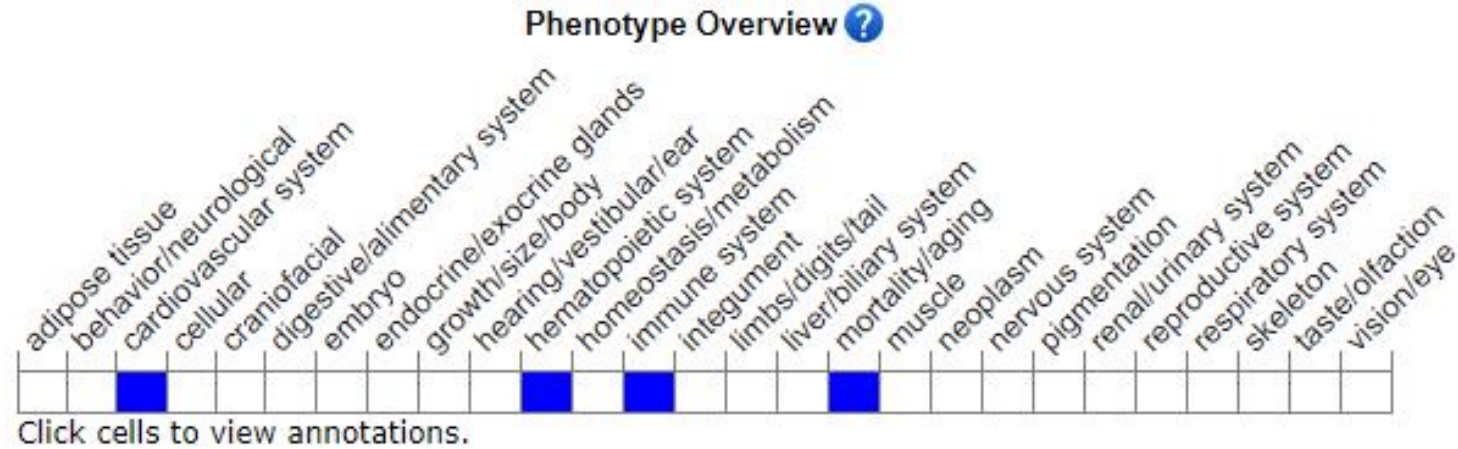
Genomic location distribution



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

