

# Rbpj Cas9-KO Strategy

**Designer:** Huan Wang

**Design Date:** 2019-8-3

## **Project Overview**



Project Name Rbpj

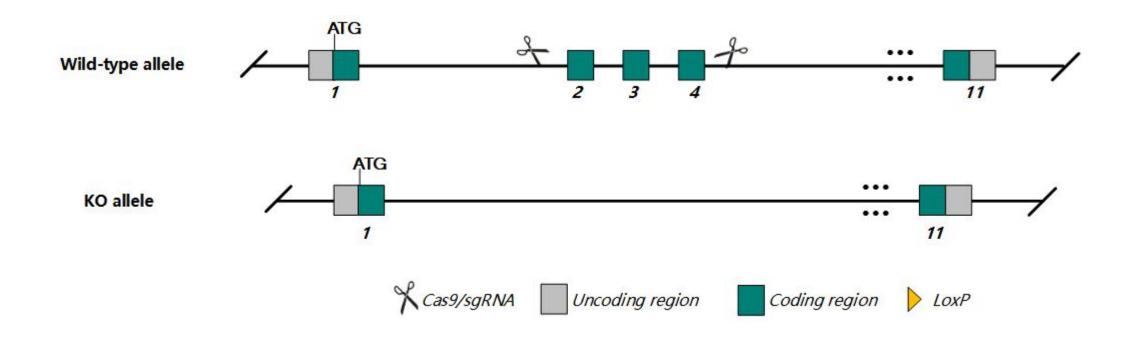
Project type Cas9-KO

Strain background C57BL/6JGpt

## **Knockout strategy**



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



### **Technical routes**



The *Rbpj* gene has 12 transcripts. According to the structure of *Rbpj* gene, exon2-exon4 of *Rbpj-201* ENSMUST00000037618.12) transcript is recommended as the knockout region. The region contains 80bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Rbpj* 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 null allele exhibit complete prenatal lethality.

The *Rbpj* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

## Gene information NCBI



#### Rbpj recombination signal binding protein for immunoglobulin kappa J region [Mus musculus (house mouse)]

Gene ID: 19664, updated on 12-Mar-2019

#### Summary



Official Symbol Rbpj provided by MGI

Official Full Name recombination signal binding protein for immunoglobulin kappa J region provided by MGI

Primary source MGI:MGI:96522

See related Ensembl:ENSMUSG00000039191

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 Al843960, CBF1, Igkirb, Igkrsbp, RBP-J, RBP-J kappa, RBP-Jkappa, RBPjk, Rbpsuh

Expression Ubiquitous expression in limb E14.5 (RPKM 12.9), CNS E11.5 (RPKM 12.5) and 28 other tissuesSee more

Orthologs <u>human all</u>

## Transcript information Ensembl



The gene has 12 transcripts, all transcripts are shown below:

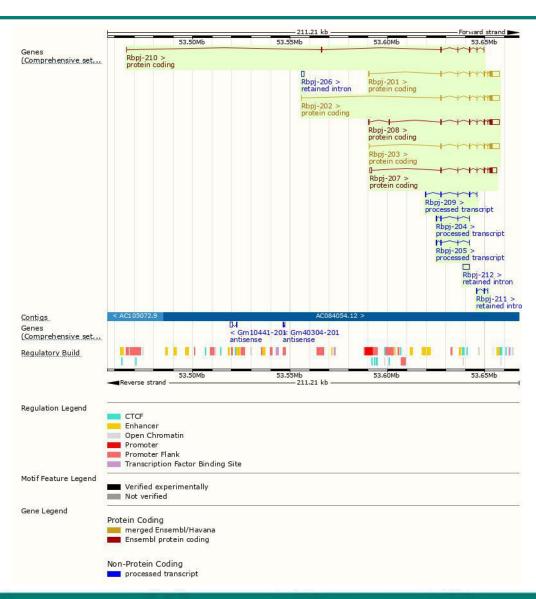
| Name 🍦   | Transcript ID A       | bp 🌲 | Protein 🍦    | Biotype              | CCDS 🍦         | UniProt 👙       | RefSeq 🍦                       | Flags                             |
|----------|-----------------------|------|--------------|----------------------|----------------|-----------------|--------------------------------|-----------------------------------|
| Rbpj-201 | ENSMUST00000037618.12 | 5459 | <u>526aa</u> | Protein coding       | CCDS19292₽     | P31266 @        | NM_009035 &<br>NP_033061 &     | TSL:1 GENCODE basic               |
| Rbpj-202 | ENSMUST00000087360.8  | 5252 | 485aa        | Protein coding       | CCDS51503₽     | <u>E9Q7W0</u> ₽ | NM_001080928&<br>NP_001074397& | TSL:5 GENCODE basic               |
| Rbpj-203 | ENSMUST00000113865.4  | 5440 | <u>487aa</u> | Protein coding       | CCDS51504₽     | P31266& Q3UM17& | NM_001080927@<br>NP_001074396@ | TSL:1   GENCODE basic   APPRIS P1 |
| Rbpj-204 | ENSMUST00000200856.3  | 499  | No protein   | Processed transcript | 82             | ( <u>)</u> 2()  | =                              | TSL:3                             |
| Rbpj-205 | ENSMUST00000201701.1  | 516  | No protein   | Processed transcript | 82             | (154)           | 8                              | TSL:2                             |
| Rbpj-206 | ENSMUST00000201721.1  | 1519 | No protein   | Retained intron      | 82             | (050)           | E .                            | TSL:NA                            |
| Rbpj-207 | ENSMUST00000201883.3  | 4961 | <u>465aa</u> | Protein coding       | 82             | Q3U6F1 &        | 9                              | TSL:1 GENCODE basic               |
| Rbpj-208 | ENSMUST00000201912.3  | 5485 | <u>507aa</u> | Protein coding       | CCDS80283 ₽    | A0A0J9YTV5@     | NM_001277116@<br>NP_001264045@ | TSL:1 GENCODE basic               |
| Rbpj-209 | ENSMUST00000201928.3  | 548  | No protein   | Processed transcript | 27             | (72)            | -                              | TSL:3                             |
| Rbpj-210 | ENSMUST00000201991.3  | 687  | 180aa        | Protein coding       | 87             | A0A0J9YVE0₽     | -                              | CDS 3' incomplete TSL:5           |
| Rbpj-211 | ENSMUST00000202092.1  | 678  | No protein   | Retained intron      | 87             | (Tab)           | -                              | TSL:2                             |
| Rbpj-212 | ENSMUST00000202476.1  | 3436 | No protein   | Retained intron      | 8 <del>7</del> | (73)            | -                              | TSL:NA                            |

The strategy is based on the design of *Rbpj-208* transcript, The transcription is shown below



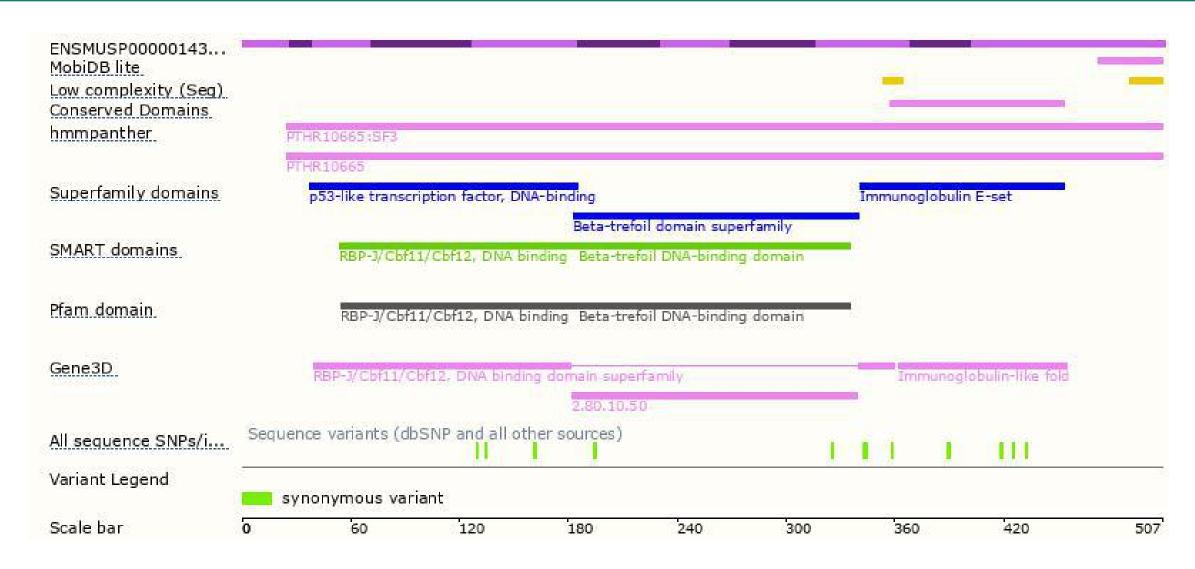
### Genomic location distribution





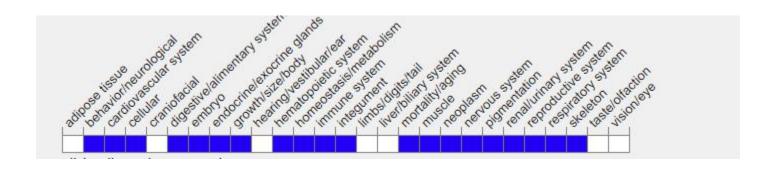
### Protein domain





## Mouse phenotype description(MGI)





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

According to the existing MGI data, Mice homozygous for a null allele exhibit complete prenatal lethality.



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





