

# Cd8b1 Cas9-CKO Strategy

Designer: Xueting Zhang

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# **Project Overview**



**Project Name** 

Cd8b1

**Project type** 

Cas9-CKO

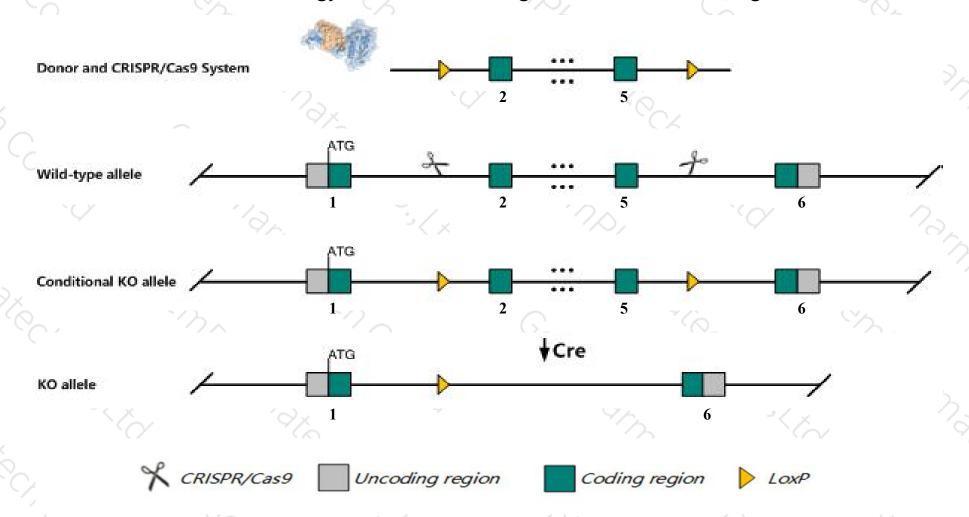
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Cd8b1* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Cd8b1* gene has 1 transcript. According to the structure of *Cd8b1* gene, exon2-exon5 of *Cd8b1-201*(ENSMUST00000065248.8) transcript is recommended as the knockout region. The region contains 586bp coding sequence.

  Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cd8b1* 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.

### **Notice**



- ➤ According to the existing MGI data, Animals homozygous for a mutation in this gene lack CD8+CD4- cytotoxic T cells in the thymus and spleen and do not mount a cytotoxic response to alloantigens.
- The floxed region is near to the N-terminal of *Symd1* gene, this strategy may influence the regulatory function of the N-terminal of *Symd1* gene.
- The *Cd8b1* gene is located on the Chr6. 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)



#### Cd8b1 CD8 antigen, beta chain 1 [Mus musculus (house mouse)]

Gene ID: 12526, updated on 31-Jan-2019

#### Summary

☆ ?

Official Symbol Cd8b1 provided by MGI

Official Full Name CD8 antigen, beta chain 1 provided by MGI

Primary source MGI:MGI:88347

See related Ensembl: ENSMUSG00000053044

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 Cd8b, Ly-3, Ly-C, Lyt-3

Expression Restricted expression toward thymus adult (RPKM 247.7)See more

Orthologs <u>human</u> all

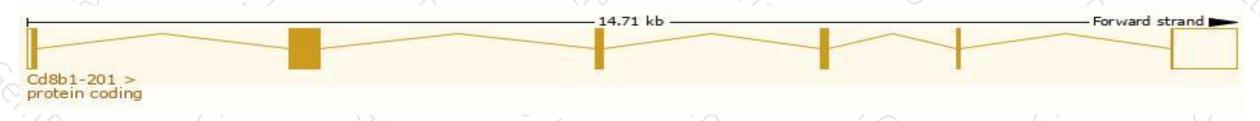
# Transcript information (Ensembl)



The gene has 1 transcript, and the transcript is shown below:

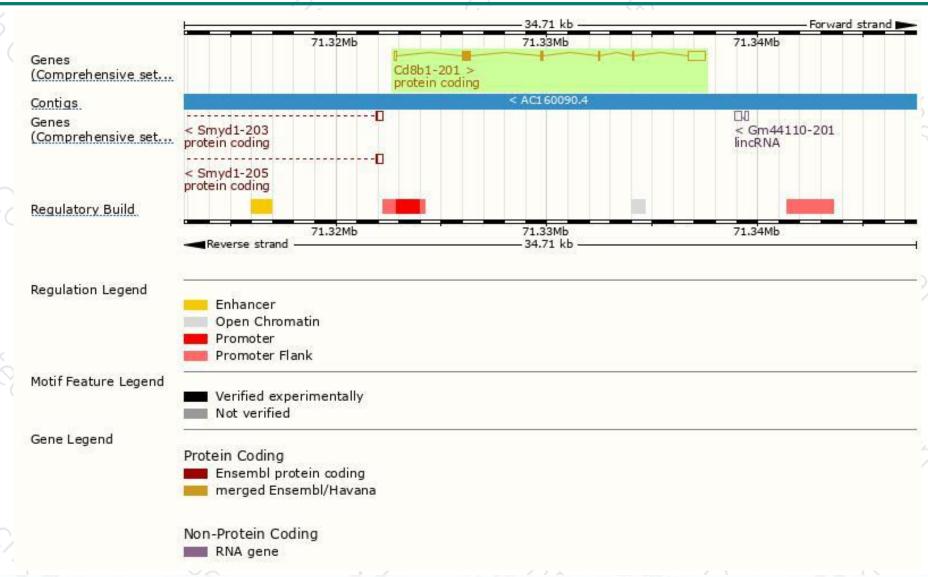
| Name      | Transcript ID        | bp   | Protein | Biotype        | CCDS      | UniProt       | Flags                         |
|-----------|----------------------|------|---------|----------------|-----------|---------------|-------------------------------|
| Cd8b1-201 | ENSMUST00000065248.8 | 1483 | 213aa   | Protein coding | CCDS39506 | P10300 Q3TEK8 | TSL:1 GENCODE basic APPRIS P1 |

The strategy is based on the design of Cd8b1-201 transcript, The transcription is shown below



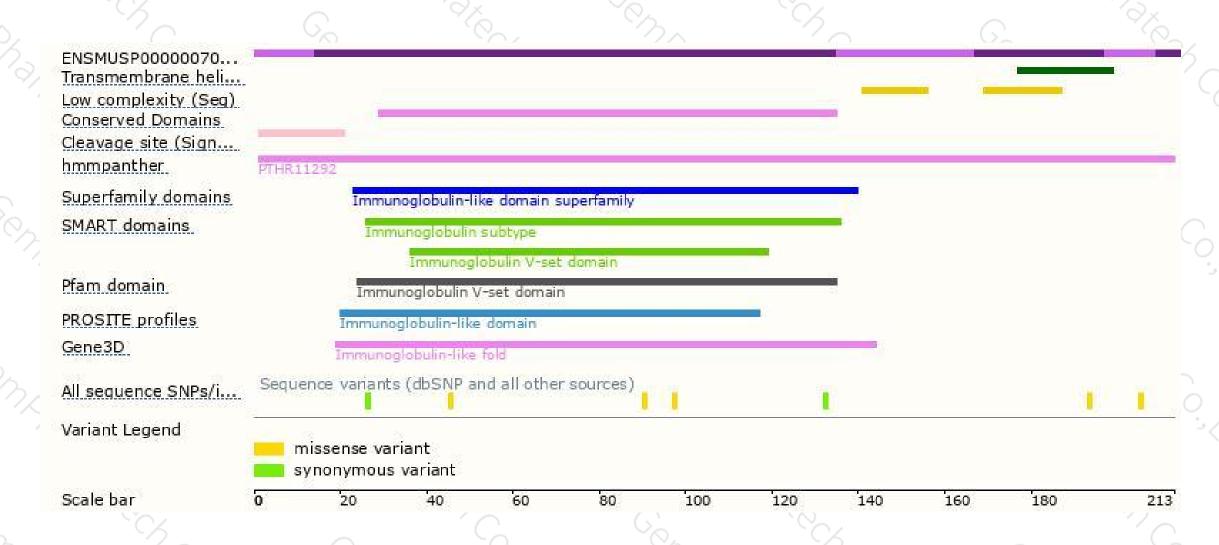
### Genomic location distribution





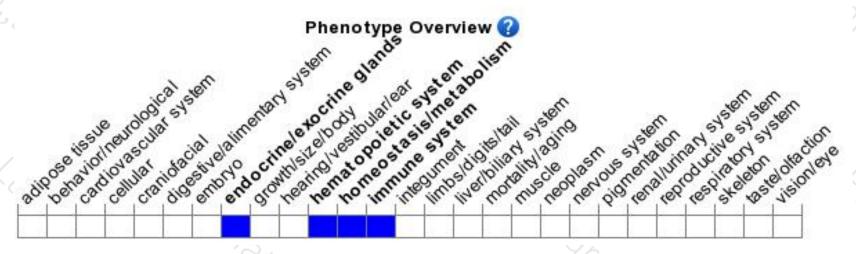
### Protein domain





# Mouse phenotype description(MGI)





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

According to the existing MGI data, Animals homozygous for a mutation in this gene lack CD8+CD4- cytotoxic T cells in the thymus and spleen and do not mount a cytotoxic response to alloantigens.



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





