

Lrrc8a Cas9-CKO Strategy

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Project Overview

Project Name

Lrrc8a

Project type

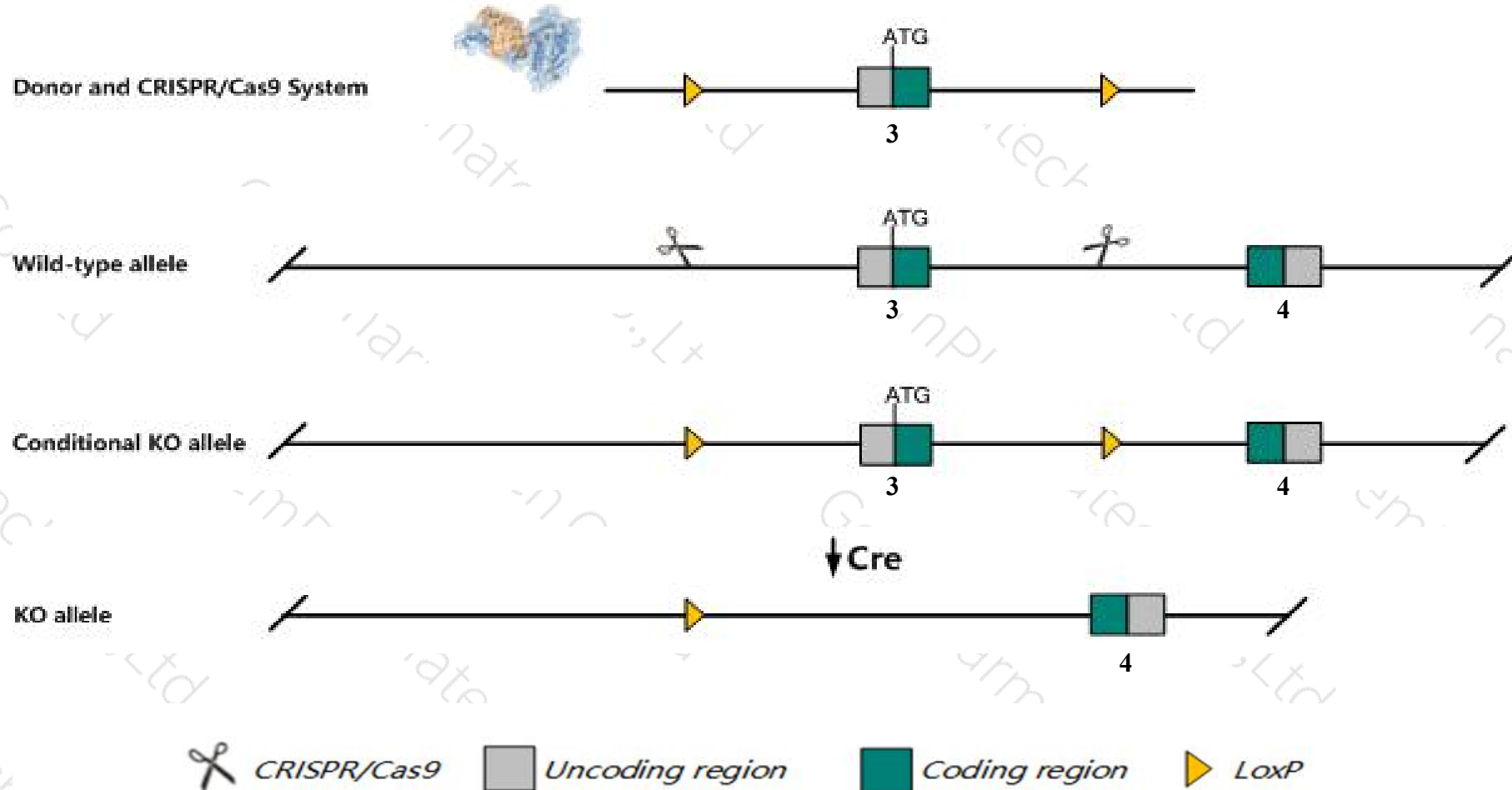
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Lrrc8a* gene. The schematic diagram is as follows:



Technical routes

- The *Lrrc8a* gene has 2 transcripts. According to the structure of *Lrrc8a* gene, exon3 of *Lrrc8a-201* (ENSMUST00000095078.2) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Lrrc8a* 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.

- According to the existing MGI data, Homozygous for a knock-out allele exhibit prenatal lethality and premature death, growth retardation, sterility, multiple tissue abnormalities, a severe block in early thymic development, and impaired peripheral T cell function. B cell development is modestly impaired but B cell function is normal.
- Because of *Gm28035* gene is located in *Lrrc8a* gene, *Gm28035* gene will be deleted together in this strategy.
- The *Lrrc8a* gene is located on the Chr2. 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)

Lrrc8a leucine rich repeat containing 8A [Mus musculus (house mouse)]

Gene ID: 241296, updated on 17-Feb-2019

Summary



| | |
|---------------------------|---|
| Official Symbol | Lrrc8a provided by MGI |
| Official Full Name | leucine rich repeat containing 8A provided by MGI |
| Primary source | MGI:MGI:2652847 |
| See related | Ensembl:ENSMUSG000000007476 |
| 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 | Lrrc8, ebo, mKIAA1437 |
| Expression | Ubiquitous expression in lung adult (RPKM 34.6), ovary adult (RPKM 30.0) and 28 other tissues See more |
| Orthologs | human all |

Transcript information (Ensembl)

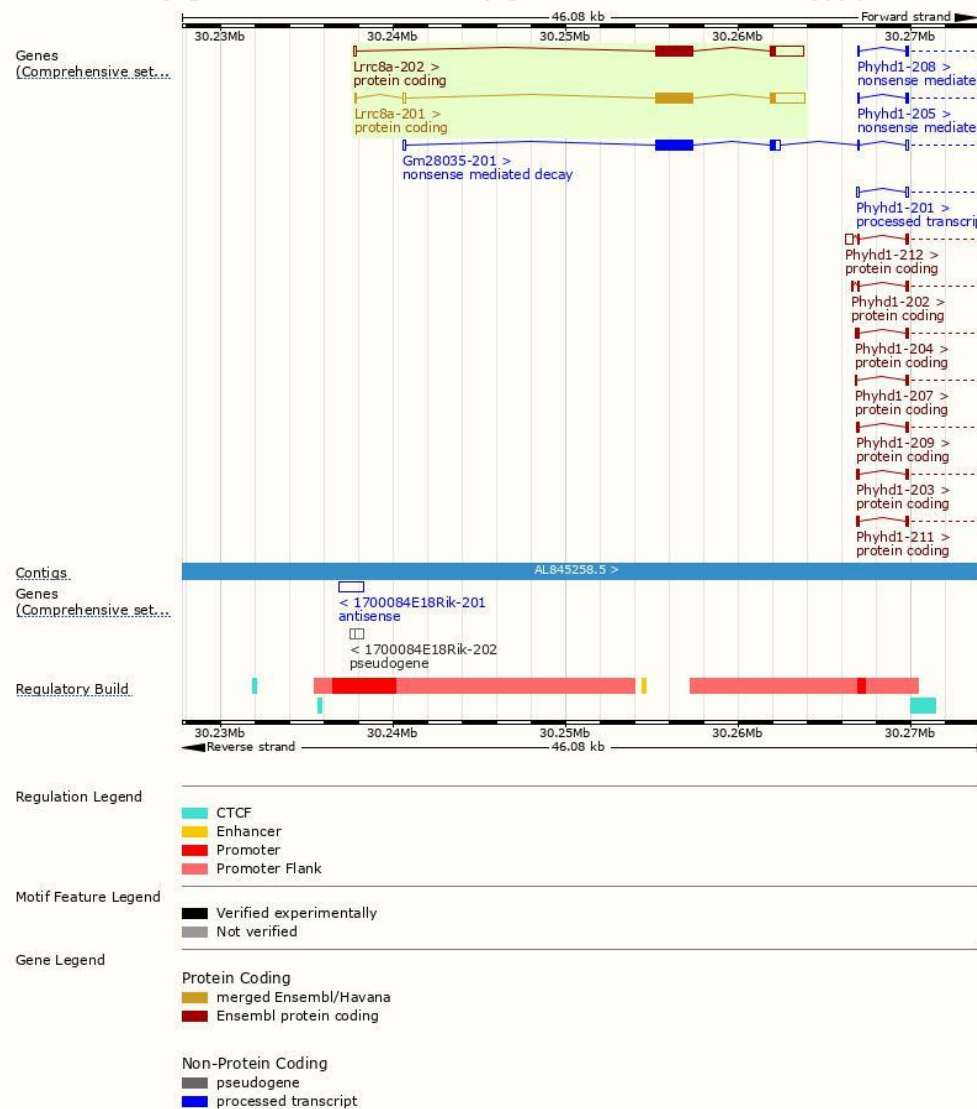
The gene has 2 transcripts,all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|------------|--------------------------------------|------|-----------------------|----------------|---------------------------|------------------------|-------------------------------|
| Lrrc8a-201 | ENSMUST00000095078.2 | 4301 | 810aa | Protein coding | CCDS15875 | Q80WG5 | TSL:5 GENCODE basic APPRIS P1 |
| Lrrc8a-202 | ENSMUST00000113654.7 | 4194 | 810aa | Protein coding | CCDS15875 | Q80WG5 | TSL:5 GENCODE basic APPRIS P1 |

The strategy is based on the design of *Lrrc8a-201* transcript,The transcription is shown below



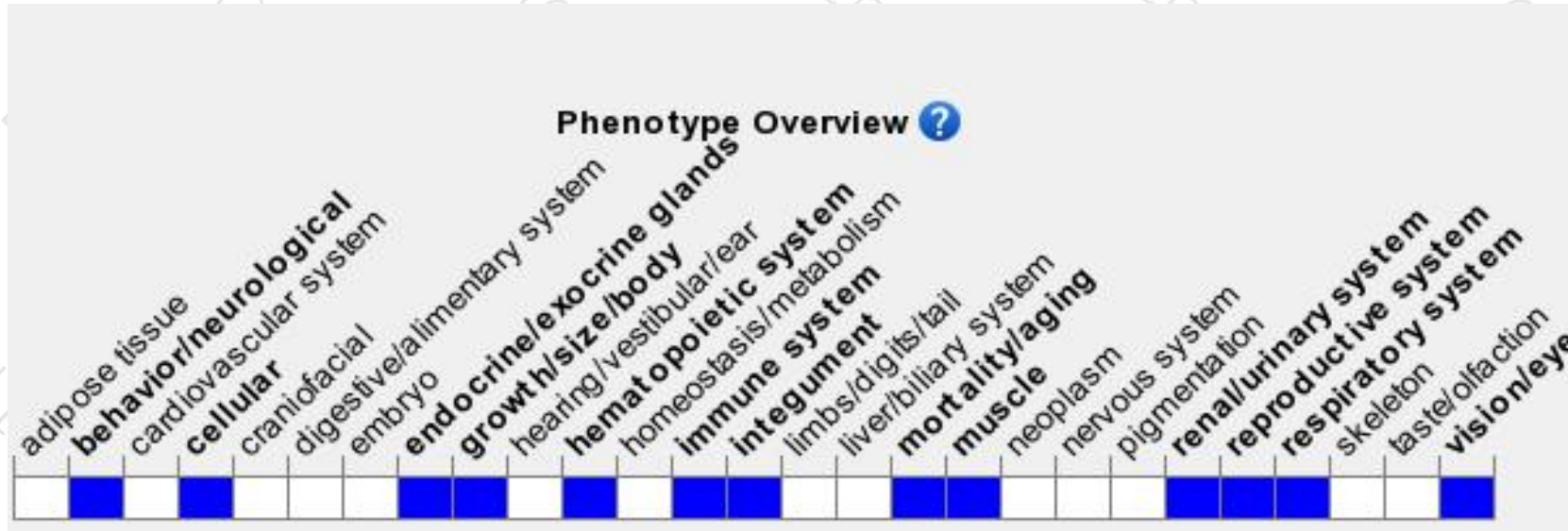
Genomic location distribution



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/>).

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If you have any questions, you are welcome to inquire.

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