

Kdm4d Cas9-CKO Strategy

Designer: Xueting Zhang

Reviewer: Yanhua Shen

Date:2020-02-25

Project Overview



Project Name

Kdm4d

Project type

Cas9-CKO

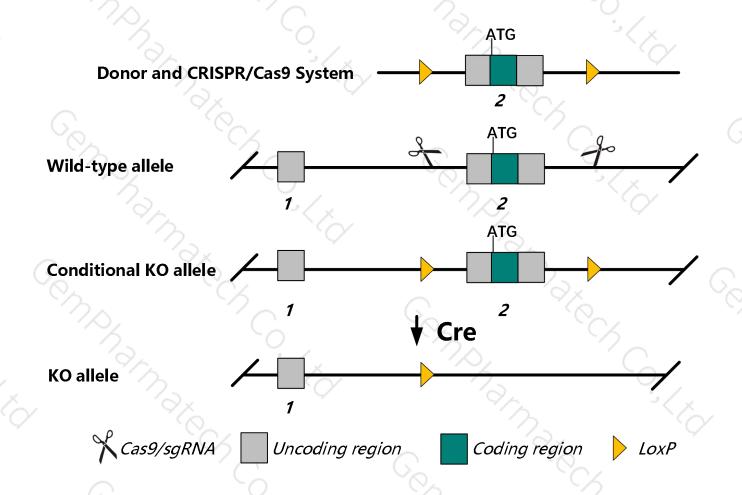
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Kdm4d* gene. The schematic diagram is as follows:



Technical routes



- The *Kdm4d* gene has 2 transcripts. According to the structure of *Kdm4d* gene, exon2 of *Kdm4d-201* (ENSMUST00000058796.6) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Kdm4d* 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, Mice homozygous for a knock-out allele exhibit accumulation of histone 3 methylation in spermatids, a transient increase in testes size, wider tubules, occasional male germ cell apoptosis, and decreased body weight. However, fertility is normal.
- > The *Kdm4d* gene is located on the Chr9. 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)



Kdm4d lysine (K)-specific demethylase 4D [Mus musculus (house mouse)]

Gene ID: 244694, updated on 16-Sep-2019

▲ Summary

☆ ?

Official Symbol Kdm4d provided by MGI

Official Full Name lysine (K)-specific demethylase 4D provided by MGI

Primary source MGI:MGI:3606484

See related Ensembl: ENSMUSG00000053914

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 Jmjd2d; 4932416A15

Expression Restricted expression toward testis adult (RPKM 31.3) See more

Orthologs human all

Genomic context



Location: 9; 9 A2

See Kdm4d in Genome Data Viewer

Exon count: 4

| Annotation release | Status | Assembly | Chr | Location | |
|--------------------|-------------------|------------------------------|-----|--|----|
| 108 | current | GRCm38.p6 (GCF_000001635.26) | 9 | NC_000075.6 (1446254814500522, complement) | 10 |
| Build 37.2 | previous assembly | MGSCv37 (GCF_000001635.18) | 9 | NC_000075.5 (1426702514304926, complement) | |

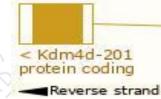
Transcript information (Ensembl)



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

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|-----------|----------------------|------|--------------|----------------|------------------|---------|-------------------------------|
| Kdm4d-201 | ENSMUST00000058796.6 | 2695 | 510aa | Protein coding | CCDS22822 | Q3U2K5 | TSL:1 GENCODE basic APPRIS P1 |
| Kdm4d-202 | ENSMUST00000115647.2 | 2117 | <u>450aa</u> | Protein coding | . . 8 | Z4YLE9 | CDS 3' incomplete TSL:1 |

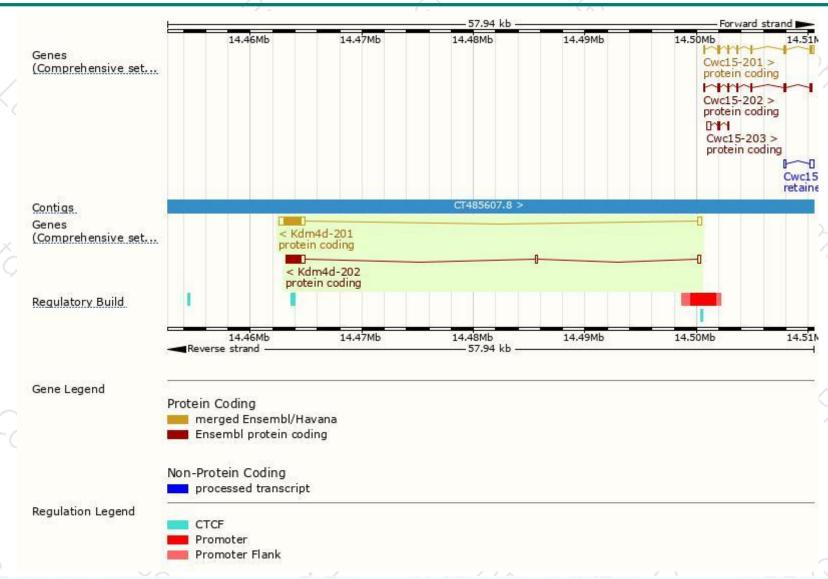
The strategy is based on the design of *Kdm4d-201* transcript, The transcription is shown below



37.94 kb

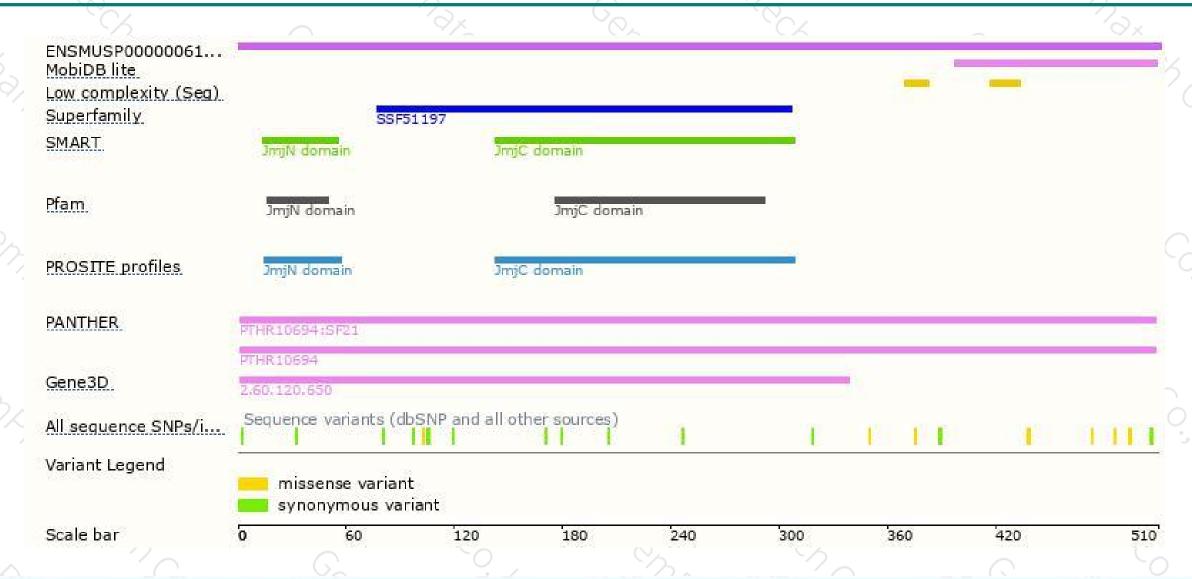
Genomic location distribution





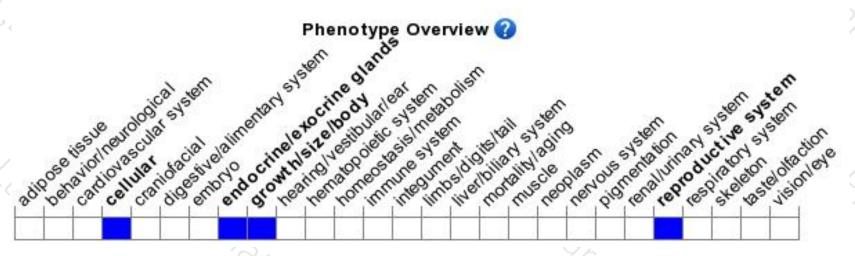
Protein domain





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 knock-out allele exhibit accumulation of histone 3 methylation in spermatids, a transient increase in testes size, wider tubules, occasional male germ cell apoptosis, and decreased body weight. However, fertility is normal.



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





