

***Kmt2d* Cas9-CKO Strategy**

Designer:

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Project Overview

Project Name

Kmt2d

Project type

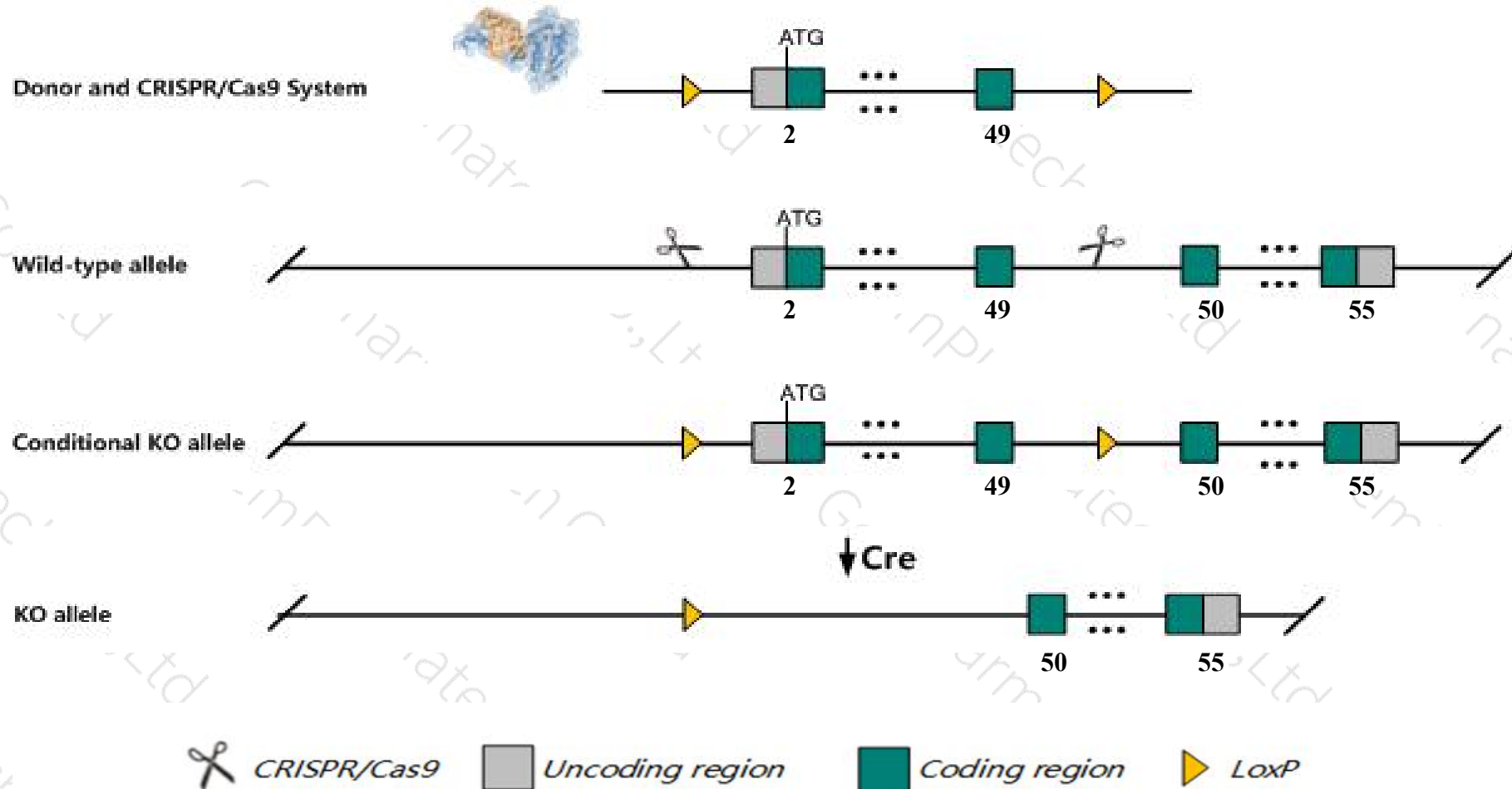
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Kmt2d* gene. The schematic diagram is as follows:



- The *Kmt2d* gene has 5 transcripts. According to the structure of *Kmt2d* gene, exon2-exon49 of *Kmt2d-201* (ENSMUST00000023741.15) 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 *Kmt2d* 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, Mice homozygous for a gene trap allele exhibit embryonic lethality around E9.5. Mice homozygous for a conditional allele activated in different cell-types exhibit impaired adipogenesis, impaired myogenesis, perturbed germinal B cell development and promotion of lymphomagenesis.
- The *Kmt2d* gene is located on the Chr15. 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)

Kmt2d lysine (K)-specific methyltransferase 2D [Mus musculus (house mouse)]

Gene ID: 381022, updated on 2-Apr-2019

Summary



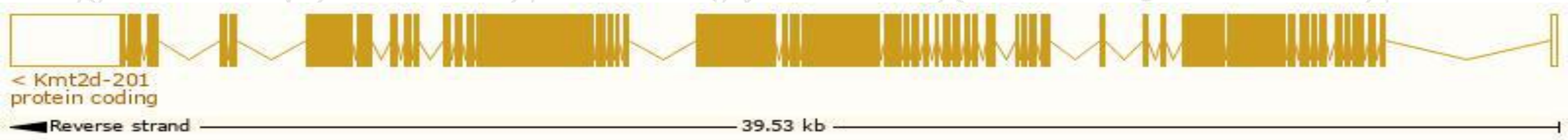
Official Symbol	Kmt2d provided by MGI
Official Full Name	lysine (K)-specific methyltransferase 2D provided by MGI
Primary source	MGI:MGI:2682319
See related	Ensembl:ENSMUSG00000048154
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	ALR, BC032281, BC058659, C430014K11Rik, KMT2B, MII2, MII4
Expression	Ubiquitous expression in thymus adult (RPKM 25.1), spleen adult (RPKM 21.8) and 27 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

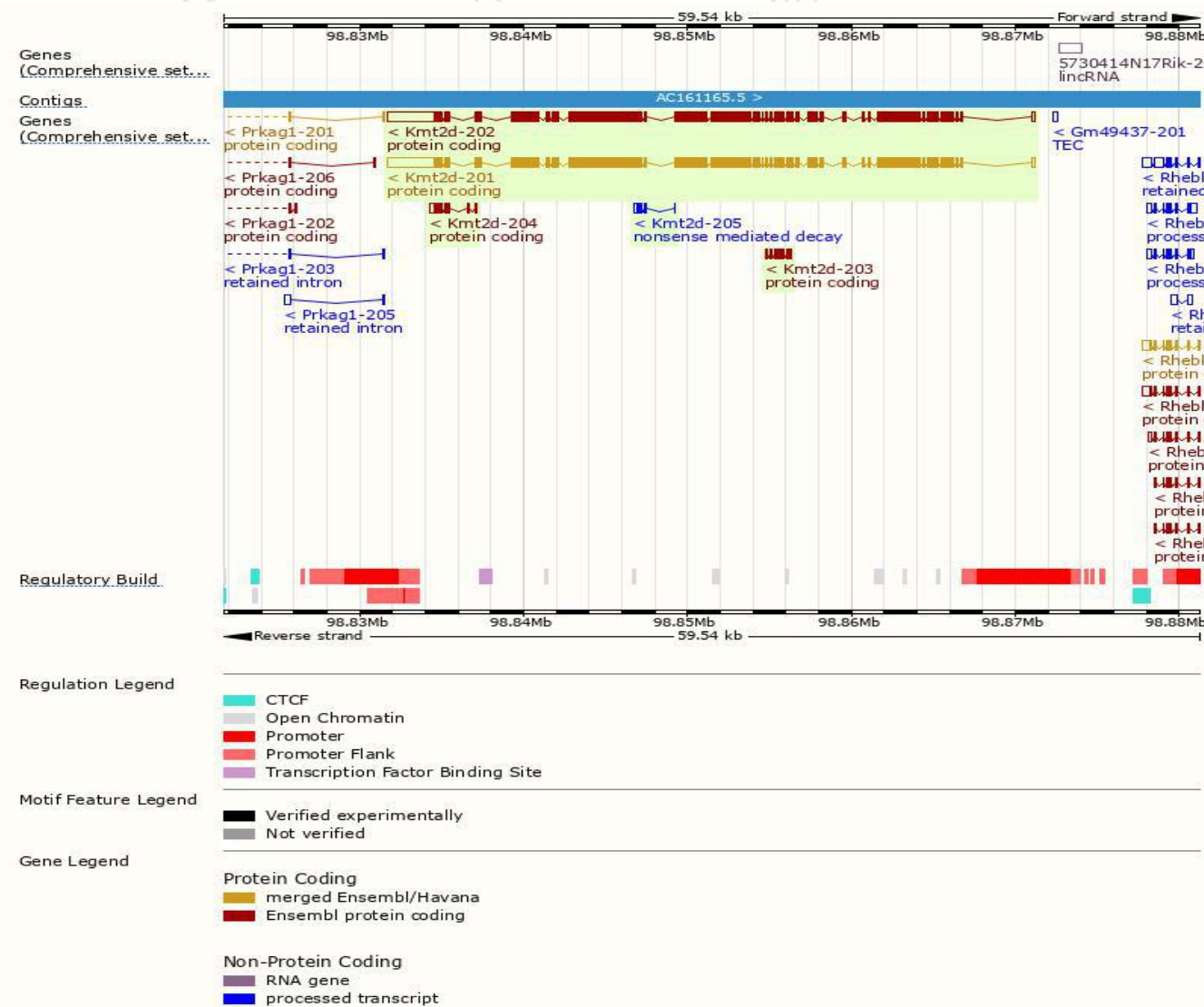
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kmt2d-201	ENSMUST00000023741.15	19823	5588aa	Protein coding	CCDS49725	A0A0A0MQ73	TSL:5 GENCODE basic APPRIS P2
Kmt2d-202	ENSMUST00000178486.8	19805	5588aa	Protein coding	-	Q6PDK2	TSL:5 GENCODE basic APPRIS ALT2
Kmt2d-204	ENSMUST00000191973.2	899	233aa	Protein coding	-	A0A0A6YY77	CDS 5' incomplete TSL:3
Kmt2d-203	ENSMUST00000184363.2	751	250aa	Protein coding	-	V9GX77	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Kmt2d-205	ENSMUST00000229651.1	470	22aa	Nonsense mediated decay	-	A0A2R8VHD4	CDS 5' incomplete

The strategy is based on the design of *Kmt2d-201* transcript,The transcription is shown below



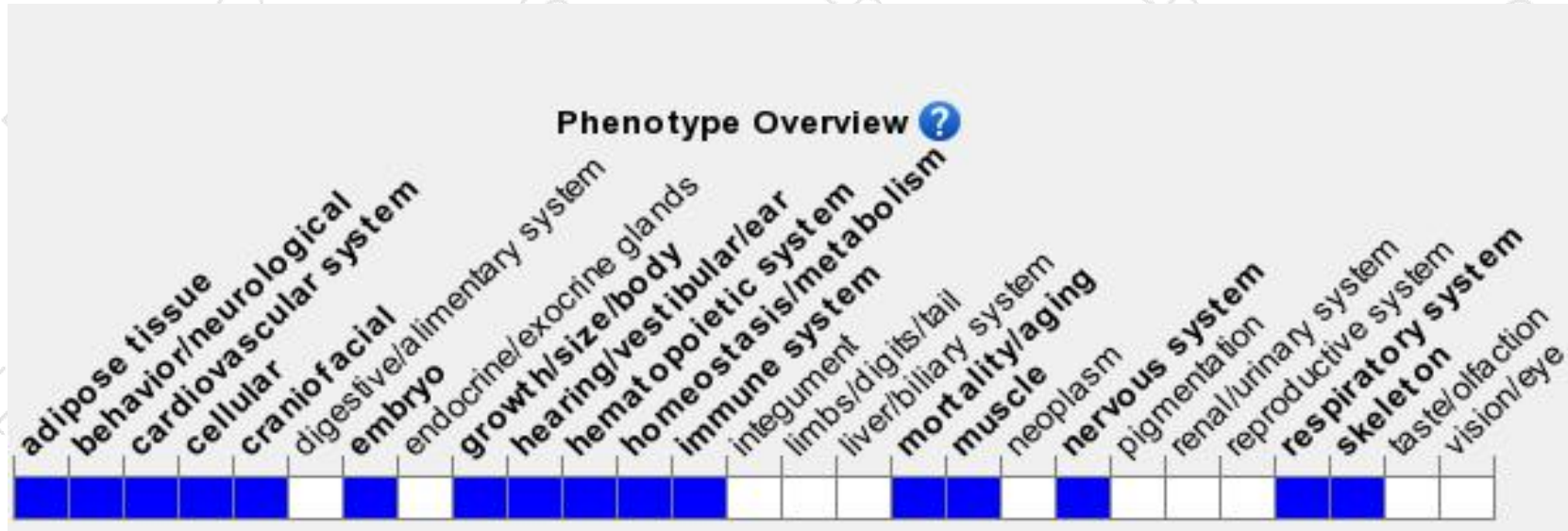
Genomic location distribution



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, Mice homozygous for a gene trap allele exhibit embryonic lethality around E9.5. Mice homozygous for a conditional allele activated in different cell-types exhibit impaired adipogenesis, impaired myogenesis, perturbed germinal B cell development and promotion of lymphomagenesis.

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

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