Kmt2a Cas9-CKO Strategy

Designer: Xiaojing Li
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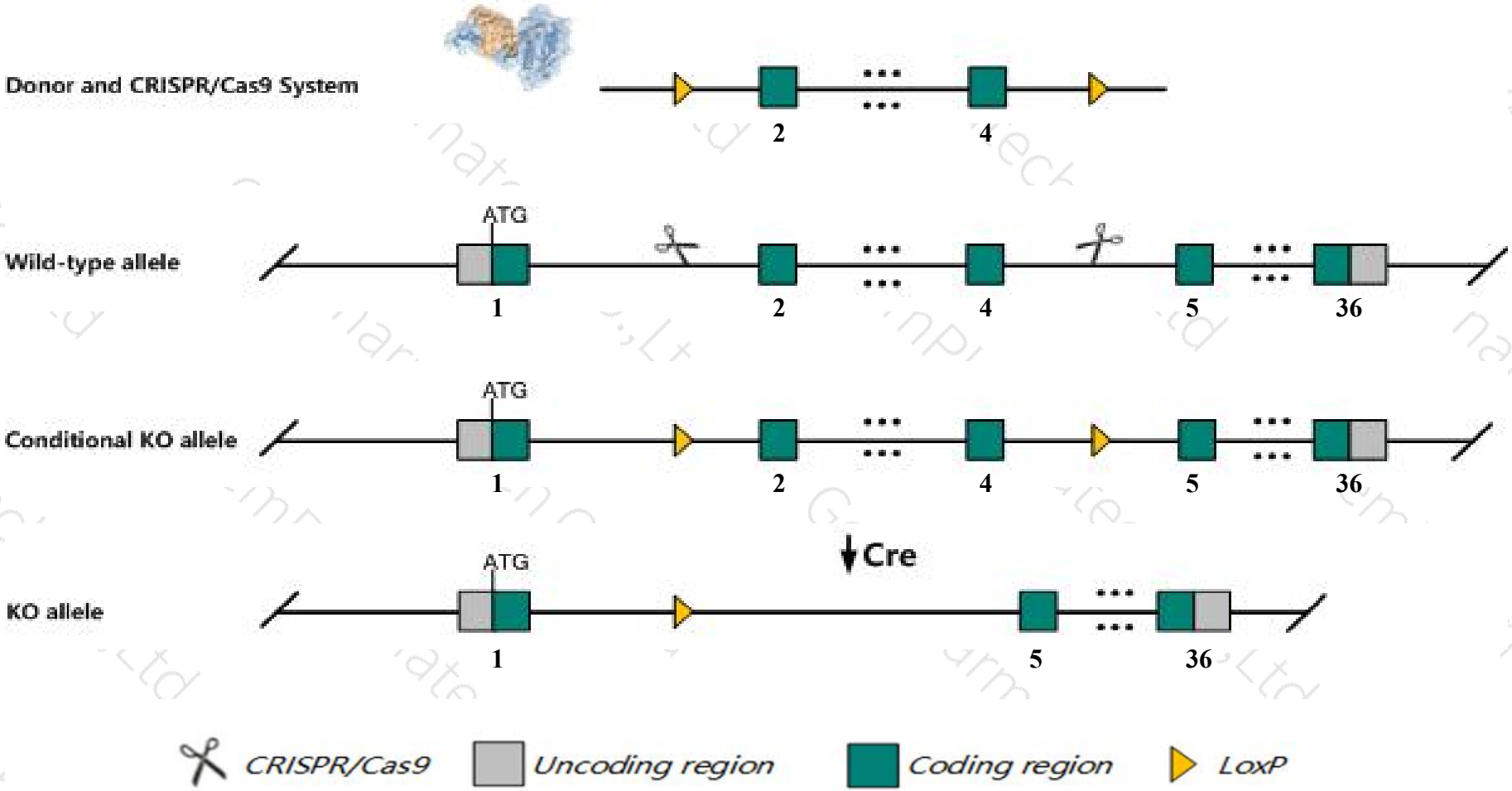
Reviewer: JiaYu

Project Overview

Project Name	***Kmt2a***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Kmt2a* gene. The schematic diagram is as follows:



Technical routes

- The *Kmt2a* gene has 6 transcripts. According to the structure of *Kmt2a* gene, exon2-exon4 of *Kmt2a-201* (ENSMUST00000002095.10) transcript is recommended as the knockout region.The region contains 2899bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Kmt2a* 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,Homozygotes for targeted null mutations die at embryonic day 11.5-14.5 with edematous bodies, petechiae, and hematopoietic insufficiency. Heterozygotes show reduced growth, hematopoietic abnormalities, and homeotic transformations of the axial skeleton.

➢ The *Kmt2a* 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）

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Kmt2a lysine (K)-specific methyltransferase 2A [Mus musculus (house mouse)]

Gene ID: 214162, updated on 2-Apr-2019

Summary

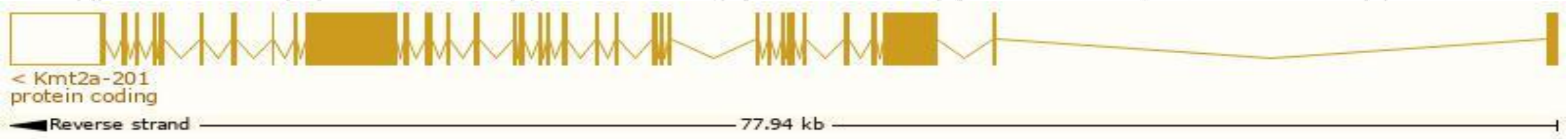
Official Symbol	Kmt2a provided by MGI
Official Full Name	lysine (K)-specific methyltransferase 2A provided by MGI
Primary source	MGI:MGI:96995
See related	Ensembl:ENSMUSG00000002028
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	6430520K01, ALL-1, All1, Cxxc7, HRX, HTRX1, Mll, Mll1, mKIAA4050
Expression	Ubiquitous expression in thymus adult (RPKM 13.7), spleen adult (RPKM 10.1) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

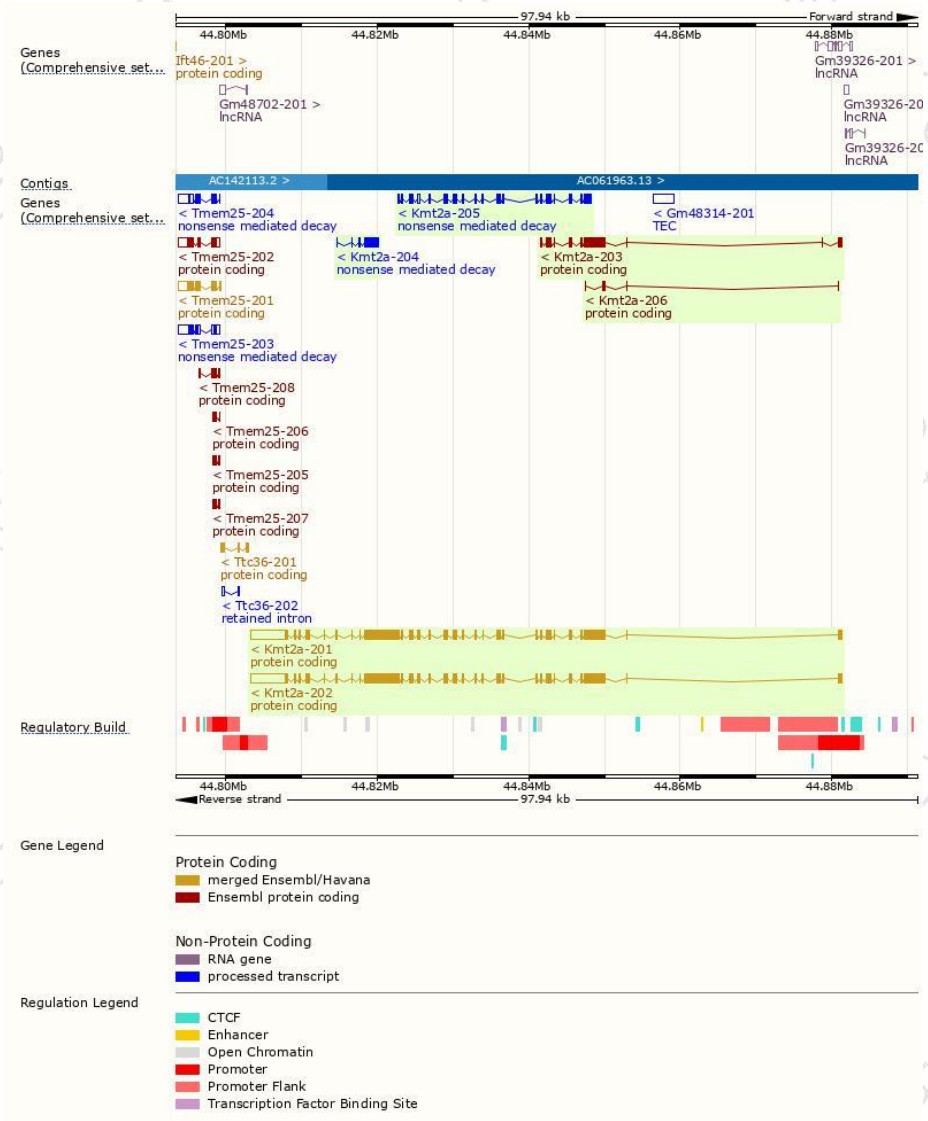
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kmt2a-201	ENSMUST00000002095.10	16461	3963aa	Protein coding	CCDS40603	P55200	TSL:5 GENCODE basic APPRIS P2
Kmt2a-202	ENSMUST00000114689.7	16470	3966aa	Protein coding	-	P55200	TSL:5 GENCODE basic APPRIS ALT2
Kmt2a-203	ENSMUST00000128768.2	4336	1438aa	Protein coding	-	D3Z5H5	CDS 3' incomplete TSL:5
Kmt2a-206	ENSMUST00000215489.1	521	173aa	Protein coding	-	A0A1L1SQS9	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Kmt2a-205	ENSMUST00000152241.7	4081	629aa	Nonsense mediated decay	-	S4R2S5	CDS 5' incomplete TSL:5
Kmt2a-204	ENSMUST00000138119.1	2146	639aa	Nonsense mediated decay	-	S4R199	CDS 5' incomplete TSL:1

The strategy is based on the design of *Kmt2a-201* transcript,The transcription is shown below

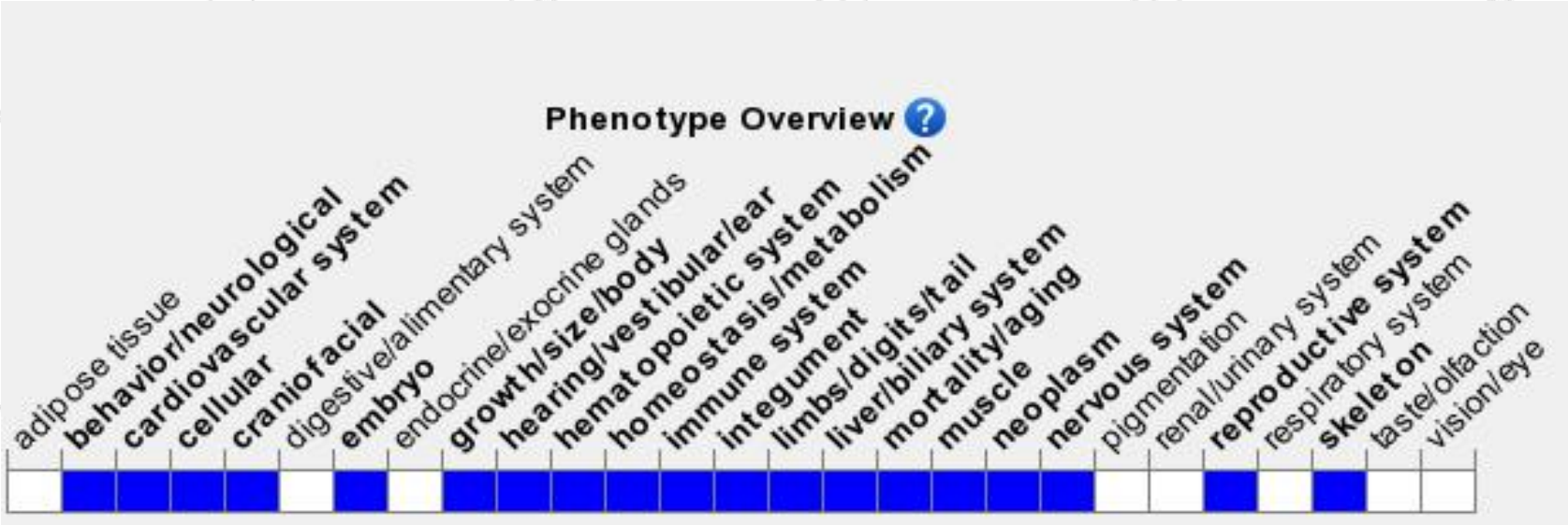


Genomic location distribution



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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集萃药康
GemPharmatech

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

Tel: 400-9660890

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GemPharmatech Co.,Ltd