

# Kdm2a Cas9-KO Strategy

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Design Date: 2019-08-05

# **Project Overview**



Project Name Kdm2a

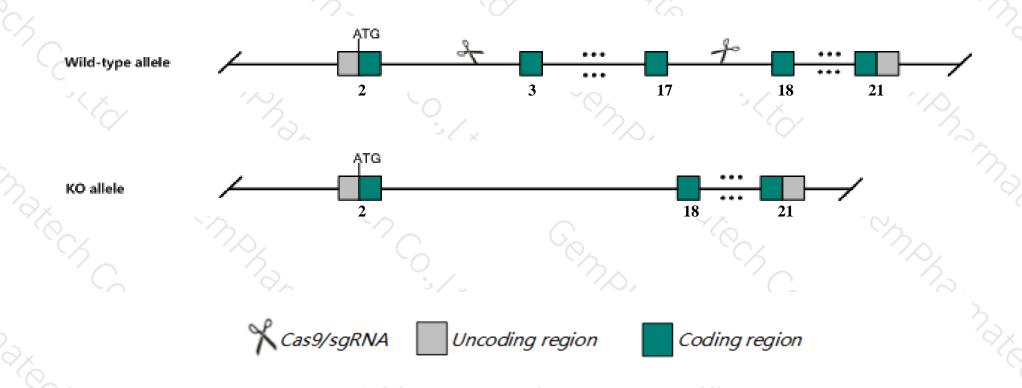
Project type Cas9-KO

Strain background C57BL/6JGpt

# **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Kdm2a* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Kdm2a* gene has 14 transcripts. According to the structure of *Kdm2a* gene, exon3-exon17 of *Kdm2a-201* (ENSMUST00000047898.13) transcript is recommended as the knockout region. The region contains 2723bp coding sequence Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Kdm2a* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

### **Notice**



- ➤ According to the existing MGI data, Mice homozygous for a null allele show embryonic lethality, severe growth retardation, reduced neuron proliferation, increased neuron apoptosis, impaired neuron differentiation, small hearts, abnormal cardiac looping and, in some cases, incomplete embryonic turning and neural tube closure defects.
- ➤ The *Kdm2a* gene is located on the Chr19. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

# Gene information (NCBI)



#### Kdm2a lysine (K)-specific demethylase 2A [Mus musculus (house mouse)]

Gene ID: 225876, updated on 31-Jan-2019

#### Summary

☆ ?

Official Symbol Kdm2a provided by MGI

Official Full Name lysine (K)-specific demethylase 2A provided by MGI

Primary source MGI:MGI:1354736

See related Ensembl:ENSMUSG00000054611

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 100043628, 5530401A10Rik, AA589516, AW536790, Cxxc8, Fbl11, Fbl7, Fbxl11, Gm4560, Jhdm1, Jhdm1a, Ialina

Expression Ubiquitous expression in thymus adult (RPKM 17.5), limb E14.5 (RPKM 16.0) and 28 other tissuesSee more

Orthologs <u>human</u> all

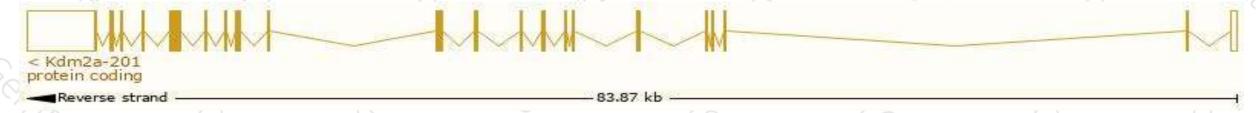
# Transcript information (Ensembl)



The gene has 14 transcripts, all transcripts are shown below:

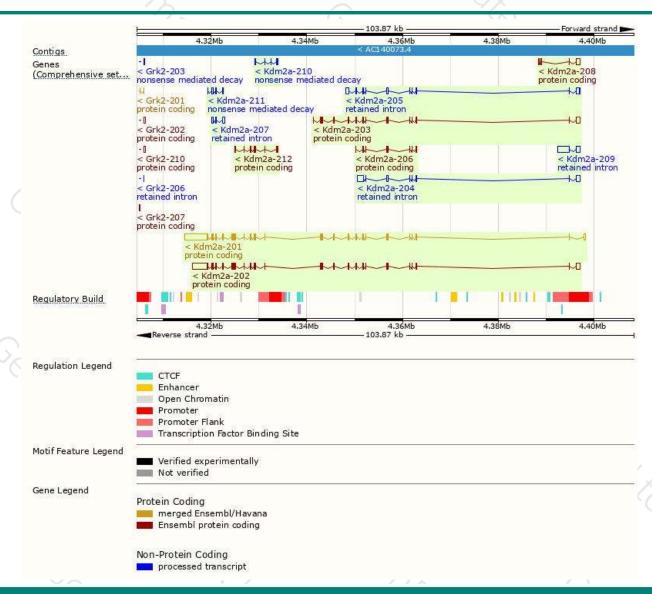
-	Name	Transcript ID A	bp 🛊	Protein	Biotype	CCDS	UniProt	Flags
2	Kdm2a-201	ENSMUST00000047898.13	8578	<u>1161aa</u>	Protein coding	CCDS37886 €	F6YRW4&	TSL:1 GENCODE basic APPRIS P1
	Kdm2a-202	ENSMUST00000075856.10	7242	<u>1161aa</u>	Protein coding	<u>CCDS37886</u> €	F6YRW4&	TSL:5 GENCODE basic APPRIS P1
	Kdm2a-203	ENSMUST00000116571.8	2413	<u>494aa</u>	Protein coding	-	<u>A0A087WP68</u> ₽	TSL:1 GENCODE basic
	Kdm2a-204	ENSMUST00000175682.1	2638	No protein	Retained intron	-	-	TSL:1
	Kdm2a-205	ENSMUST00000175777.8	2073	No protein	Retained intron	-	-	TSL:5
	Kdm2a-206	ENSMUST00000175959.1	738	246aa	Protein coding	-	H3BL82@	CDS 5' and 3' incomplete TSL:3
	Kdm2a-207	ENSMUST00000175978.1	767	No protein	Retained intron	-	-	TSL:2
	Kdm2a-208	ENSMUST00000176483.2	1128	<u>35aa</u>	Protein coding	-	A0A087WQL9@	TSL:1 GENCODE basic
	Kdm2a-209	ENSMUST00000176495.1	3207	No protein	Retained intron	-	-	TSL:1
	Kdm2a-210	ENSMUST00000176497.1	438	No protein	IncRNA	-	-	TSL:3
	Kdm2a-211	ENSMUST00000176532.1	570	<u>84aa</u>	Nonsense mediated decay	-	H3BKJ9©	CDS 5' incomplete TSL:3
	Kdm2a-212	ENSMUST00000176653.1	1076	<u>292aa</u>	Protein coding	-	H3BLD4₽	CDS 3' incomplete TSL:3
	Kdm2a-213	ENSMUST00000235335.1	2195	<u>29aa</u>	Nonsense mediated decay	-	-	CDS 5' incomplete
	Kdm2a-214	ENSMUST00000237268.1	894	No protein	Retained intron	-	-	-

The strategy is based on the design of *Kdm2a-201* transcript, The transcription is shown below



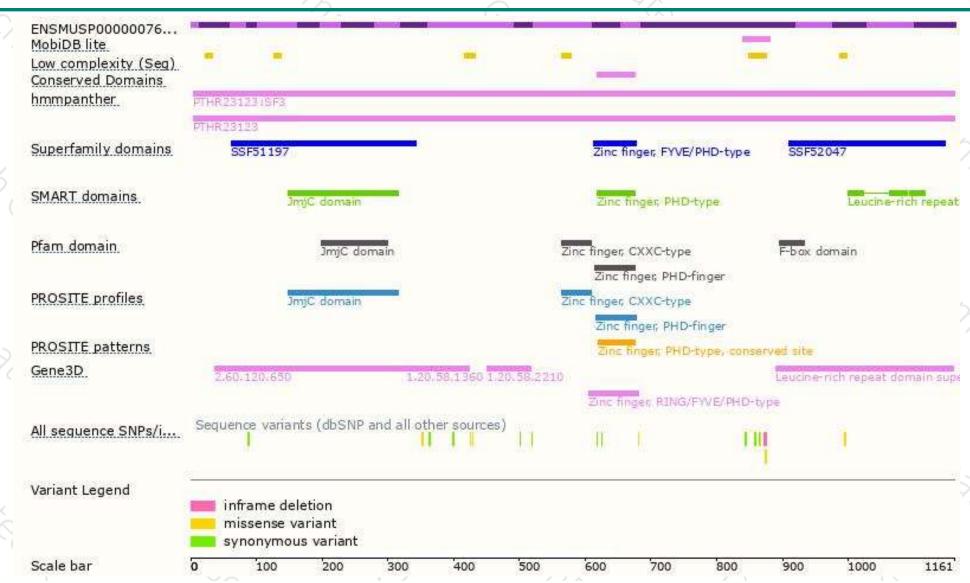
### Genomic location distribution





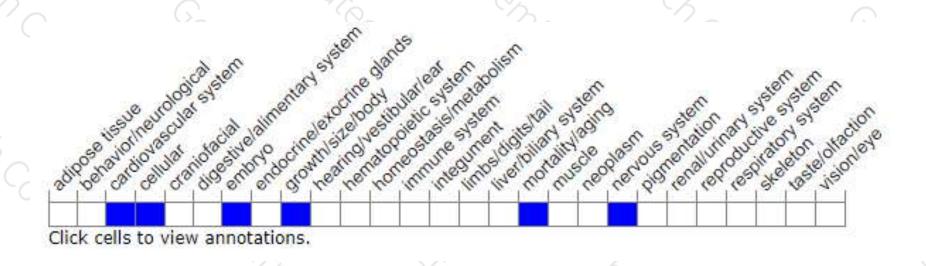
### Protein domain





## 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 null allele show embryonic lethality, severe growth retardation, reduced neuron proliferation, increased neuron apoptosis, impaired neuron differentiation, small hearts, abnormal cardiac looping and, in some cases, incomplete embryonic turning and neural tube closure defects.



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

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