

Kdm3a Cas9-KO Strategy Rohalanakoch Co.

Designer:Xiaojing Li

Project Overview



Project Name

Kdm3a

Project type

Cas9-KO

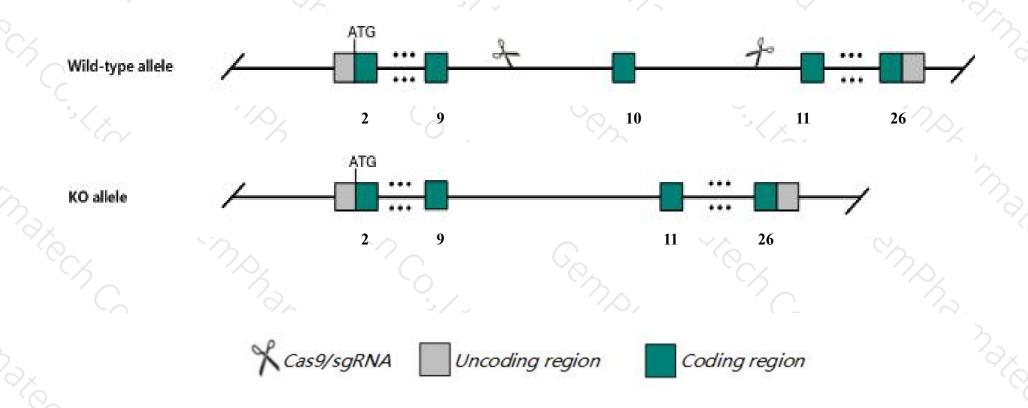
Strain background

C57BL/6J

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Kdm3a* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Kdm3a* gene has 14 transcripts. According to the structure of *Kdm3a* gene, exon10 of *Kdm3a-202* (
 ENSMUST00000167220.3) transcript is recommended as the knockout region. The region contains 512bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Kdm3a* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

Notice



- ➤ According to the existing MGI data, Male mice homozygous for a hypomorphic allele display infertility, oligoasthenoteratozoospermia, small testis, and impaired spermiogenesis. Mice homozygous for a null allele exhibit abnormal spermatogenesis and obesity associated with hyperlipidemia.
- ➤ The *Kdm3a* gene is located on the Chr6. 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information (NCBI)



Kdm3a lysine (K)-specific demethylase 3A [Mus musculus (house mouse)]

Gene ID: 104263, updated on 19-Feb-2019

Summary

☆ ?

Official Symbol Kdm3a provided by MGI

Official Full Name lysine (K)-specific demethylase 3A provided byMGI

Primary source MGI:MGI:98847

See related Ensembl:ENSMUSG00000053470

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 1700105C21Rik, C230043E16Rik, JHDM2a, Jmjd1, Jmjd1a, KDM2A, TGSA, Tsga

Expression Ubiquitous expression in CNS E11.5 (RPKM 18.6), testis adult (RPKM 15.8) and 25 other tissuesSee more

Orthologs <u>human</u> all

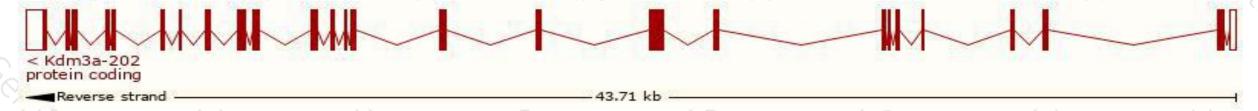
Transcript information (Ensembl)



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

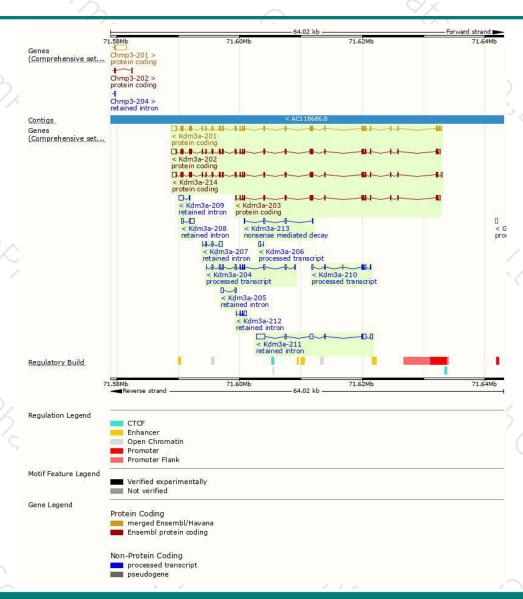
Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
ENSMUST00000167220.3	4853	1323aa	Protein coding	CCDS20233	Q6PCM1	TSL:1 GENCODE basic APPRIS P
ENSMUST00000065509.10	4816	<u>1323aa</u>	Protein coding	CCDS20233	Q6PCM1	TSL:1 GENCODE basic APPRIS P
ENSMUST00000207023.1	4753	<u>1323aa</u>	Protein coding	CCDS20233	Q6PCM1	TSL:5 GENCODE basic APPRIS P
ENSMUST00000205289.1	2723	834aa	Protein coding	-	A0A0U1RNV6	CDS 3' incomplete TSL:1
ENSMUST00000206916.1	679	<u>43aa</u>	Nonsense mediated decay	-5	A0A0U1RPI3	CDS 5' incomplete TSL:3
ENSMUST00000205470.1	1718	No protein	Processed transcript	-		TSL:5
ENSMUST00000206597.1	828	No protein	Processed transcript	-	(2)	TSL:3
ENSMUST00000206050.1	341	No protein	Processed transcript		128	TSL:3
ENSMUST00000206704.1	3137	No protein	Retained intron	-5	1/5/	TSL:1
ENSMUST00000206357.1	917	No protein	Retained intron	-		TSL:2
ENSMUST00000206339.1	815	No protein	Retained intron	-	(2)	TSL:2
ENSMUST00000206582.1	785	No protein	Retained intron		727	TSL:3
ENSMUST00000206798.1	743	No protein	Retained intron	-5	1/51	TSL:2
ENSMUST00000205505.1	541	No protein	Retained intron	-	393	TSL:2
	ENSMUST00000167220.3 ENSMUST00000167220.3 ENSMUST00000065509.10 ENSMUST00000207023.1 ENSMUST00000205289.1 ENSMUST00000206916.1 ENSMUST00000206597.1 ENSMUST00000206597.1 ENSMUST00000206704.1 ENSMUST00000206339.1 ENSMUST00000206339.1 ENSMUST00000206582.1 ENSMUST00000206798.1	ENSMUST00000167220.3 4853 ENSMUST00000065509.10 4816 ENSMUST00000207023.1 4753 ENSMUST00000205289.1 2723 ENSMUST00000206916.1 679 ENSMUST00000205470.1 1718 ENSMUST00000206597.1 828 ENSMUST00000206050.1 341 ENSMUST00000206704.1 3137 ENSMUST00000206357.1 917 ENSMUST00000206339.1 815 ENSMUST00000206582.1 785 ENSMUST00000206798.1 743	ENSMUST00000167220.3 4853 1323aa ENSMUST00000065509.10 4816 1323aa ENSMUST00000207023.1 4753 1323aa ENSMUST00000205289.1 2723 834aa ENSMUST00000206916.1 679 43aa ENSMUST00000205470.1 1718 No protein ENSMUST00000206597.1 828 No protein ENSMUST00000206050.1 341 No protein ENSMUST00000206704.1 3137 No protein ENSMUST00000206704.1 917 No protein ENSMUST00000206357.1 917 No protein ENSMUST0000020639.1 815 No protein ENSMUST00000206582.1 785 No protein ENSMUST00000206798.1 743 No protein	ENSMUST00000167220.3 4853 1323aa Protein coding ENSMUST000000665509.10 4816 1323aa Protein coding ENSMUST00000207023.1 4753 1323aa Protein coding ENSMUST00000205289.1 2723 834aa Protein coding ENSMUST00000206916.1 679 43aa Nonsense mediated decay ENSMUST00000205470.1 1718 No protein Processed transcript ENSMUST00000206597.1 828 No protein Processed transcript ENSMUST00000206050.1 341 No protein Processed transcript ENSMUST00000206704.1 3137 No protein Retained intron ENSMUST00000206339.1 815 No protein Retained intron ENSMUST00000206582.1 785 No protein Retained intron ENSMUST00000206798.1 743 No protein Retained intron	ENSMUST00000167220.3 4853 1323aa Protein coding CCDS20233 ENSMUST000000665509.10 4816 1323aa Protein coding CCDS20233 ENSMUST00000207023.1 4753 1323aa Protein coding CCDS20233 ENSMUST00000205289.1 2723 834aa Protein coding - ENSMUST00000206916.1 679 43aa Nonsense mediated decay - ENSMUST00000205470.1 1718 No protein Processed transcript - ENSMUST00000206597.1 828 No protein Processed transcript - ENSMUST00000206050.1 341 No protein Retained intron - ENSMUST00000206357.1 917 No protein Retained intron - ENSMUST00000206339.1 815 No protein Retained intron - ENSMUST00000206582.1 785 No protein Retained intron - ENSMUST00000206798.1 743 No protein Retained intron -	ENSMUST00000167220.3 4853 1323aa Protein coding CCDS20233 Q6PCM1 ENSMUST00000065509.10 4816 1323aa Protein coding CCDS20233 Q6PCM1 ENSMUST00000207023.1 4753 1323aa Protein coding CCDS20233 Q6PCM1 ENSMUST00000205289.1 2723 834aa Protein coding - A0A0U1RNV6 ENSMUST00000206916.1 679 43aa Nonsense mediated decay - A0A0U1RPI3 ENSMUST00000205470.1 1718 No protein Processed transcript - - ENSMUST00000206597.1 828 No protein Processed transcript - - ENSMUST00000206050.1 341 No protein Retained intron - - ENSMUST00000206357.1 917 No protein Retained intron - - ENSMUST00000206339.1 815 No protein Retained intron - - ENSMUST00000206582.1 785 No protein Retained intron - - ENSMUST00000206798.1 </td

The strategy is based on the design of Kdm3a-202 transcript, The transcription is shown below



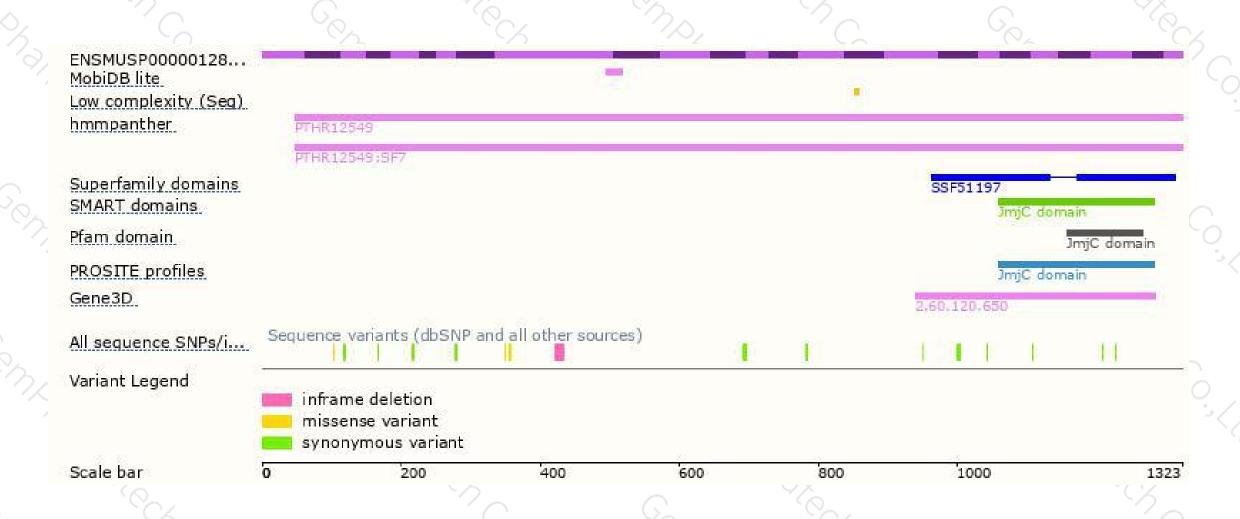
Genomic location distribution





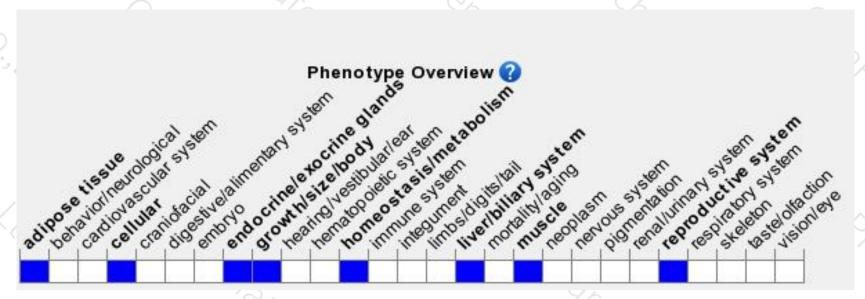
Protein domain





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, Male mice homozygous for a hypomorphic allele display infertility, oligoasthenoteratozoospermia, small testis, and impaired spermiogenesis. Mice homozygous for a null allele exhibit abnorma spermatogenesis and obesity associated with hyperlipidemia.



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

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