Rbm3 Cas9-CKO Strategy

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Reviewer: Huimin Su

Design Date: 2020-2-19

Project Overview



Project Name

Rbm3

Project type

Cas9-CKO

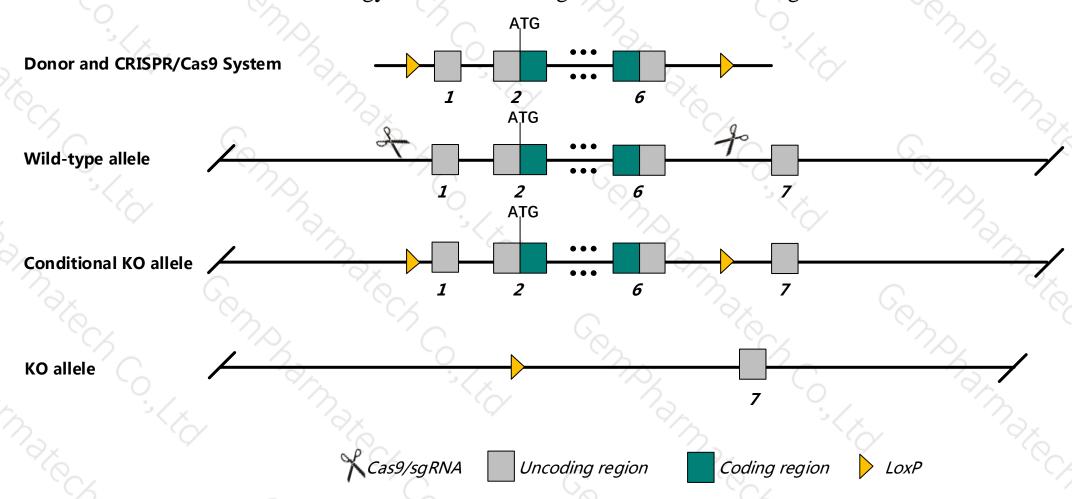
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rbm3* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Rbm3* gene has 11 transcripts. According to the structure of *Rbm3* gene, exon1-exon6 of *Rbm3*-201 (
- ➤ ENSMUST00000040010.9) 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 *Rbm3* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.

Notice



- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit decreased cell proliferation in mouse embryonic fibroblasts. This strategy may affect the 5-terminal regulation of the *Mdc1* gene.
- The KO region contains functional region of the *Rbm3os* gene. Knockout the region may affect the function of *Rbm3os* gene.
- The *Rbm3* gene is located on the ChrX. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Rbm3 RNA binding motif (RNP1, RRM) protein 3 [Mus musculus (house mouse)]

Gene ID: 19652, updated on 4-Jan-2020

Summary

☆ ?

Official Symbol Rbm3 provided by MGI

Official Full Name RNA binding motif (RNP1, RRM) protein 3 provided by MGI

Primary source MGI:MGI:1099460

See related Ensembl:ENSMUSG00000031167

Gene type protein coding
RefSeq status VALIDATED
Organism <u>Mus musculus</u>

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as 2600016C11Rik

Expression Broad expression in CNS E11.5 (RPKM 50.1), liver E14 (RPKM 45.8) and 22 other tissues See more

Orthologs human all

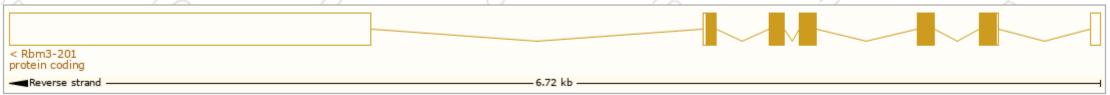
Transcript information (Ensembl)



The gene has 11 transcripts, and all transcripts are shown below:

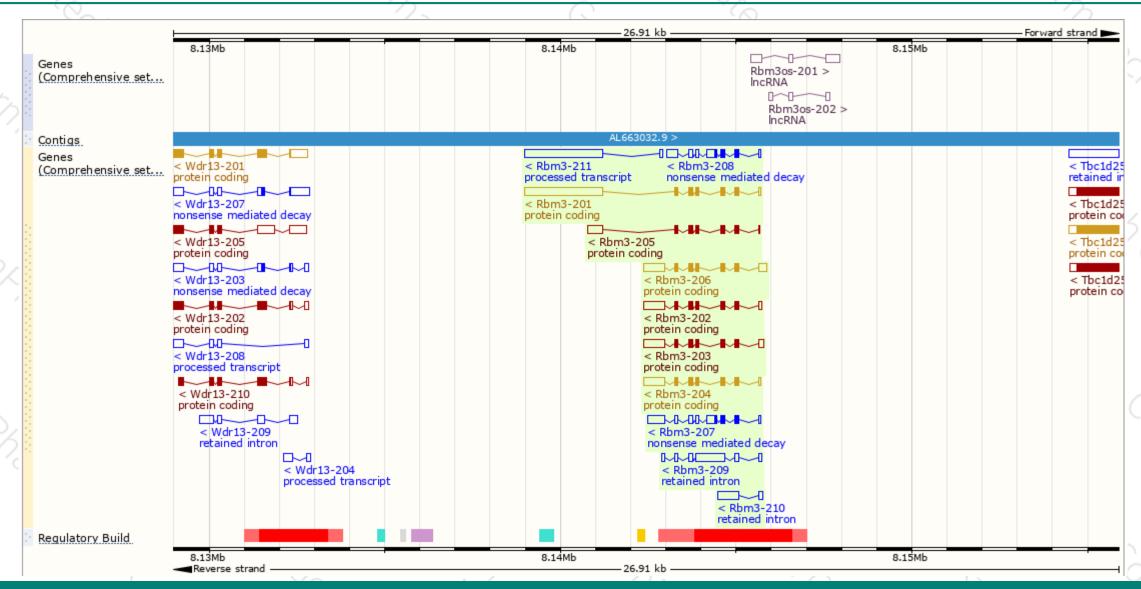
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Name 🍦	Transcript ID	bp 🌲	Protein	Biotype	CCDS	UniProt	Flags
Rbm3-201	ENSMUST00000040010.9	2792	<u>154aa</u>	Protein coding	CCDS29987 ₽	Q8BG13 ⁶	TSL:1 GENCODE basic APPRIS P3
Rbm3-206	ENSMUST00000115621.8	1338	<u>153aa</u>	Protein coding	CCDS52988 ₽	<u>089086</u> & <u>Q545K5</u> &	TSL:1 GENCODE basic APPRIS ALT1
Rbm3-203	ENSMUST00000115616.7	1225	<u>154aa</u>	Protein coding	CCDS29987 ₽	Q8BG13 ₽	TSL:1 GENCODE basic APPRIS P3
Rbm3-202	ENSMUST00000115615.8	1183	<u>153aa</u>	Protein coding	CCDS52988 ₽	<u>089086</u> & <u>Q545K5</u> &	TSL:1 GENCODE basic APPRIS ALT1
Rbm3-204	ENSMUST00000115617.9	1152	<u>154aa</u>	Protein coding	CCDS29987 ₽	Q8BG13 ₽	TSL:1 GENCODE basic APPRIS P3
Rbm3-205	ENSMUST00000115619.7	969	<u>153aa</u>	Protein coding	CCDS52988 ₽	<u>089086</u> & <u>Q545K5</u> &	TSL:3 GENCODE basic APPRIS ALT1
Rbm3-207	ENSMUST00000129947.8	1319	<u>90aa</u>	Nonsense mediated decay	-	<u>S4R2M6</u> ₽	TSL:5
Rbm3-208	ENSMUST00000141629.1	1055	<u>90aa</u>	Nonsense mediated decay	-	<u>S4R2M6</u> ₽	TSL:1
Rbm3-211	ENSMUST00000155599.1	2326	No protein	Processed transcript	-	-	TSL:1
Rbm3-209	ENSMUST00000141925.7	1274	No protein	Retained intron	-	-	TSL:5
Rbm3-210	ENSMUST00000145450.1	721	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of Rbm3-201 transcript, The transcription is shown below



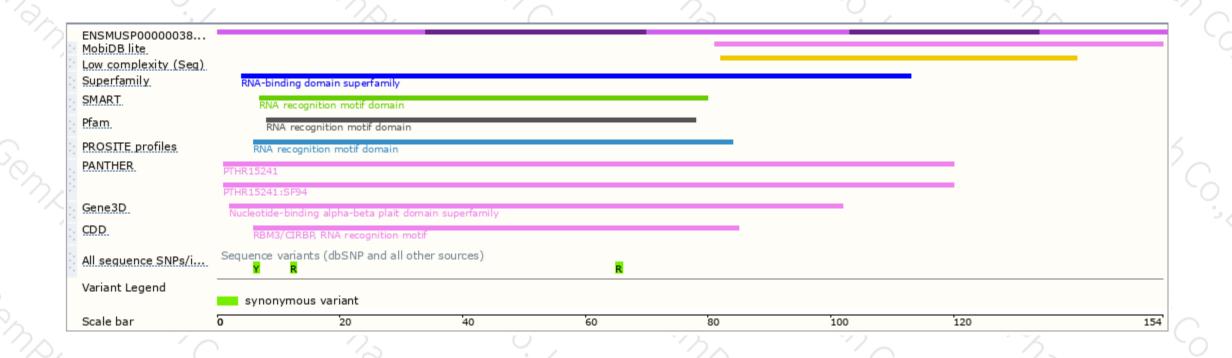
Genomic location distribution





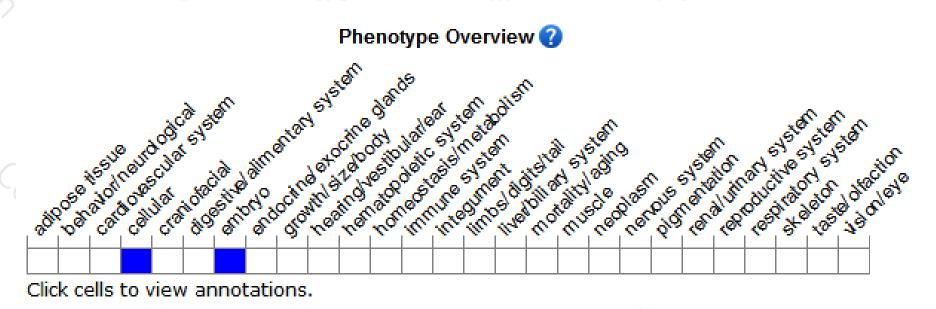
Protein domain





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 knock-out allele exhibit decreased cell proliferation in mouse embryonic fibroblasts.

If you have any questions, you are welcome to inquire. Tel: 025-5864 1534





