Rbbp8 Cas9-KO Strategy Rond Annakach Co. 1/4

Designer: Gensolatina Kech Co. (M.)

Daohua Xu

Project Overview



Project Name

Rbbp8

Project type

Cas9-KO

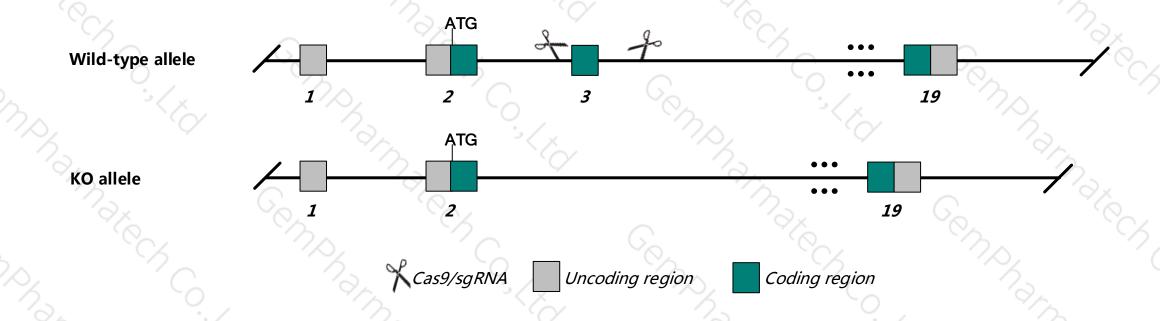
Strain background

C57BL/6JGpt

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rbbp8* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Rbbp8* gene has 11 transcripts. According to the structure of *Rbbp8* gene, exon3 of *Rbbp8*-202 transcript is recommended as the knockout region. The region contains 43bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Rbbp8* 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, Embryos homozygous for a knock-out allele die at E4.0 as blastocysts fail to enter S phase and arrest at G1, leading to elevated cell death. Heterozygous mutant mice display a shortened lifespan due to formation of multiple tumors, mostly large lymphomasof both B and T cells.
- The *Rbbp8* gene is located on the Chr18. 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)



Rbbp8 retinoblastoma binding protein 8, endonuclease [Mus musculus (house mouse)]

Gene ID: 225182, updated on 31-Jan-2019

Summary

☆ ?

Official Symbol Rbbp8 provided by MGI

Official Full Name retinoblastoma binding protein 8, endonuclease provided by MGI

Primary source MGI:MGI:2442995

See related Ensembl: ENSMUSG00000041238

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 RIM; CtIP; SAE2; RBBP-8; 9930104E21Rik

Expression Broad expression in CNS E11.5 (RPKM 6.2), placenta adult (RPKM 5.1) and 23 other tissues See more

Orthologs human all

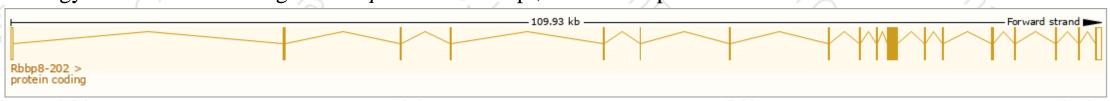
Transcript information (Ensembl)



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

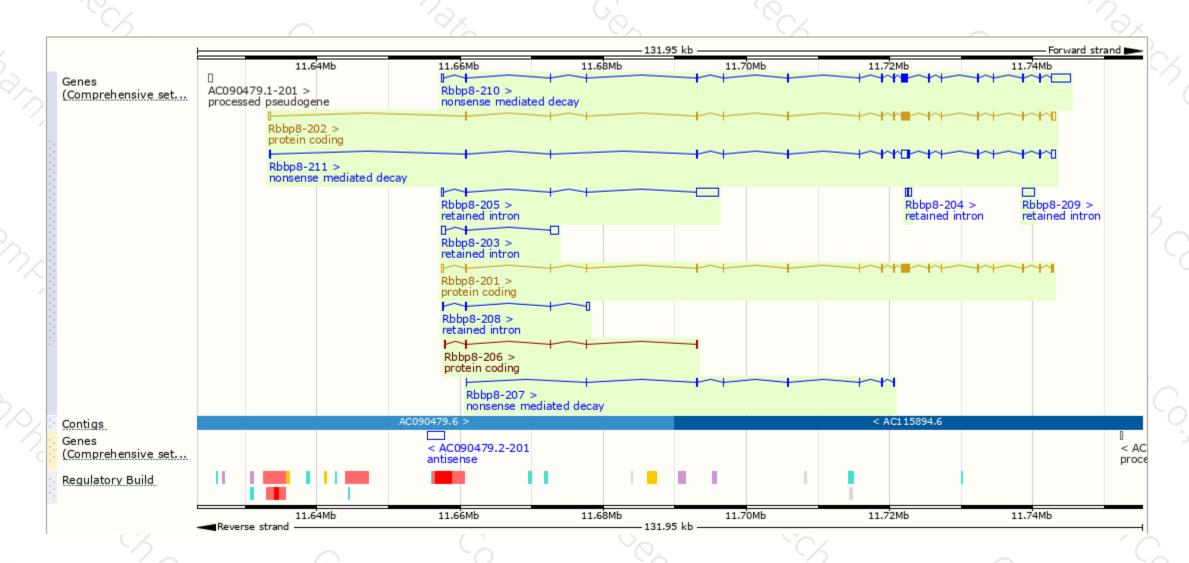
					7777			2 (
	Show/hide columns (1 hidden)							Filter
2	Name 🍦	Transcript ID	bp 🌲	Protein 🍦	Biotype	CCDS	UniProt	Flags 🝦
	Rbbp8-202	ENSMUST00000115861.8	3523	<u>893aa</u>	Protein coding	CCDS37738 ₽	<u>Q80YR6</u> ₽	TSL:1 GENCODE basic APPRIS P1
	Rbbp8-201	ENSMUST00000047322.7	3209	<u>893aa</u>	Protein coding	CCDS37738 ₽	<u>Q80YR6</u> ₽	TSL:1 GENCODE basic APPRIS P1
	Rbbp8-206	ENSMUST00000234499.1	586	<u>118aa</u>	Protein coding	-	-	CDS 3' incomplete
	Rbbp8-210	ENSMUST00000234984.1	5471	<u>612aa</u>	Nonsense mediated decay	-	-	-
	Rbbp8-211	ENSMUST00000235039.1	3360	<u>247aa</u>	Nonsense mediated decay	-	-	-
	Rbbp8-207	ENSMUST00000234616.1	781	<u>26aa</u>	Nonsense mediated decay	-	-	CDS 5' incomplete
	Rbbp8-205	ENSMUST00000234184.1	3714	No protein	Retained intron	-	-	-
	Rbbp8-203	ENSMUST00000234074.1	1904	No protein	Retained intron	-	-	-
	Rbbp8-209	ENSMUST00000234766.1	1736	No protein	Retained intron	-	-	-
	Rbbp8-208	ENSMUST00000234744.1	857	No protein	Retained intron	-	-	-
	Rbbp8-204	ENSMUST00000234161.1	679	No protein	Retained intron	-	-	-

The strategy is based on the design of *Rbbp8*-202 transcript, The transcription is shown below



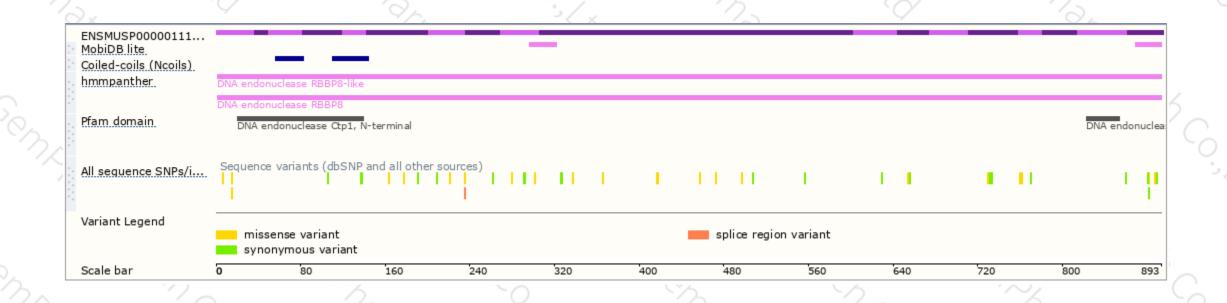
Genomic location distribution





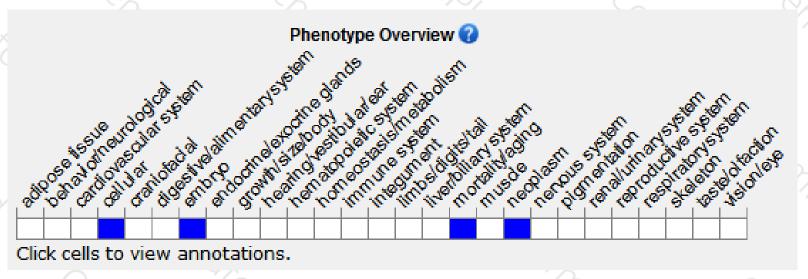
Protein domain





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, Embryos homozygous for a knock-out allele die at E4.0 as blastocysts fail to enter S phase and arrest at G1, leading to elevated cell death. Heterozygous mutant mice display a shortened lifespan due to formation of multiple tumors, mostly large lymphomasof both B and T cells.

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





