

Rbp7 Cas9-CKO Strategy

Designer:

Daohua Xu

Reviewer:

Huimin Su

Design Date:

2020-2-25

Project Overview



Project Name

Rbp7

Project type

Cas9-CKO

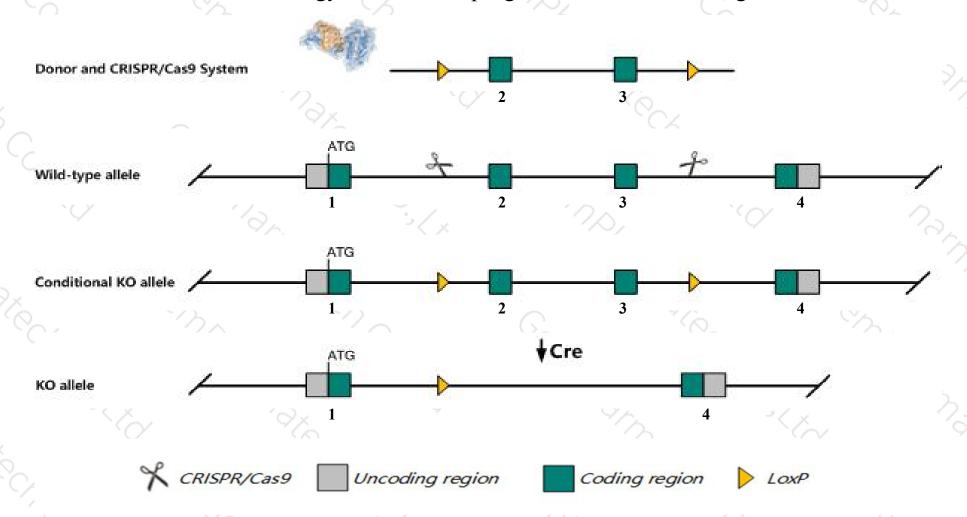
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rbp7* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Rbp7* gene has 3 transcripts. According to the structure of *Rbp7* gene, exon2-exon3 of *Rbp7-201*(ENSMUST00000030848.2) transcript is recommended as the knockout region. The region contains 281bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Rbp7* 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, Homozygous null mice have reduced retinyl ester levels in milk and other reductions in retinol and retinyl ester levels in a tissue and time dependent manner.
- ➤ This strategy may affect the 5-terminal regulation of the *Trmt112-ps2* gene.
- The *Rbp7* gene is located on the Chr4. 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)



Rbp7 retinol binding protein 7, cellular [Mus musculus (house mouse)]

Gene ID: 63954, updated on 31-Jan-2019

Summary

☆ ?

Official Symbol Rbp7 provided by MGI

Official Full Name retinol binding protein 7, cellular provided by MGI

Primary source MGI:MGI:1890409

See related Ensembl:ENSMUSG00000028996

Gene type protein coding
RefSeq status PROVISIONAL
Organism Mus musculus

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

Muroidea; Muridae; Murinae; Mus; Mus

Also known as 1110002J23Rik, CRBP-III

Expression Biased expression in subcutaneous fat pad adult (RPKM 3.4), heart adult (RPKM 2.0) and 13 other tissuesSee more

Orthologs <u>human</u> all

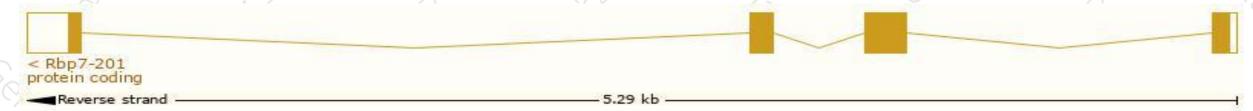
Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

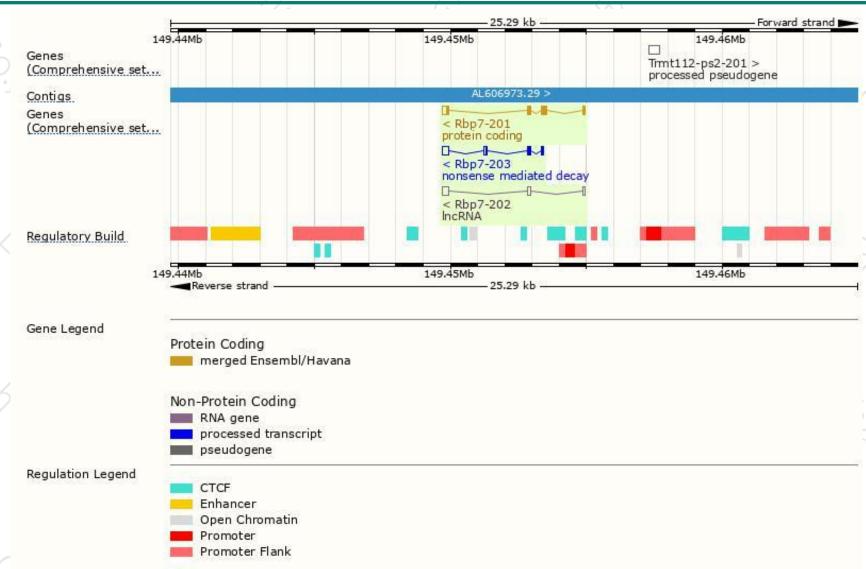
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rbp7-201	ENSMUST00000030848.2	619	<u>134aa</u>	Protein coding	CCDS18959	Q540P4 Q9EPC5	TSL:1 GENCODE basic APPRIS P1
Rbp7-203	ENSMUST00000148596.7	507	<u>68aa</u>	Nonsense mediated decay	-8	F6R1Y3	CDS 5' incomplete TSL:3
Rbp7-202	ENSMUST00000132103.1	416	No protein	IncRNA	-		TSL:3

The strategy is based on the design of *Rbp7-201* transcript, The transcription is shown below



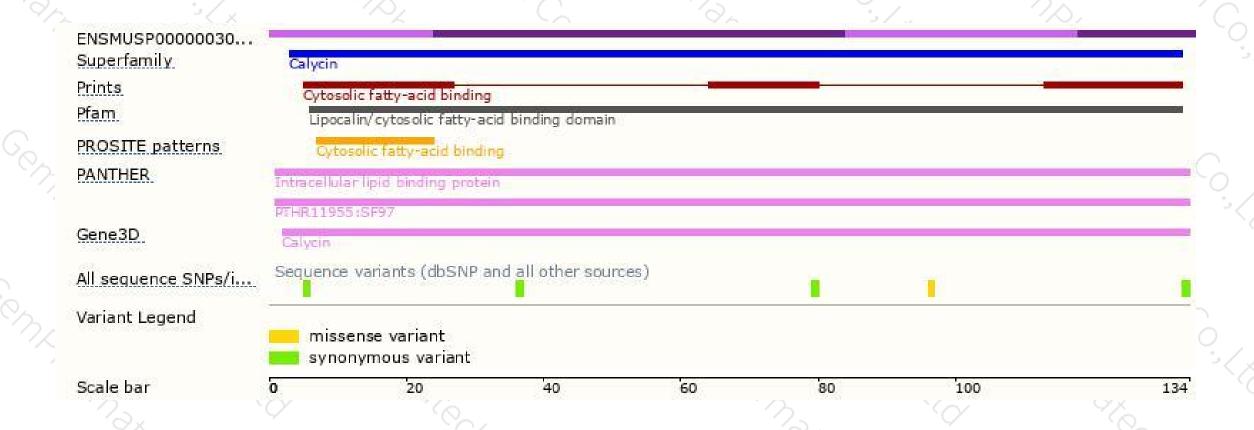
Genomic location distribution





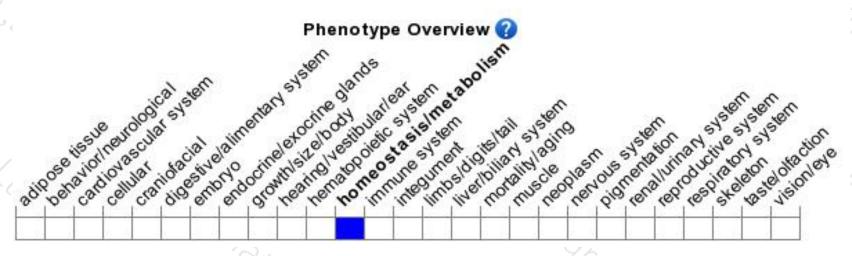
Protein domain





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, Homozygous null mice have reduced retinyl ester levels in milk and other reductions in retinol and retinyl ester levels in a tissue and time dependent manner.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





