

***Lrp5* Cas9-CKO Strategy**

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Design Date:2019-8-12

Project Overview

Project Name

Lrp5

Project type

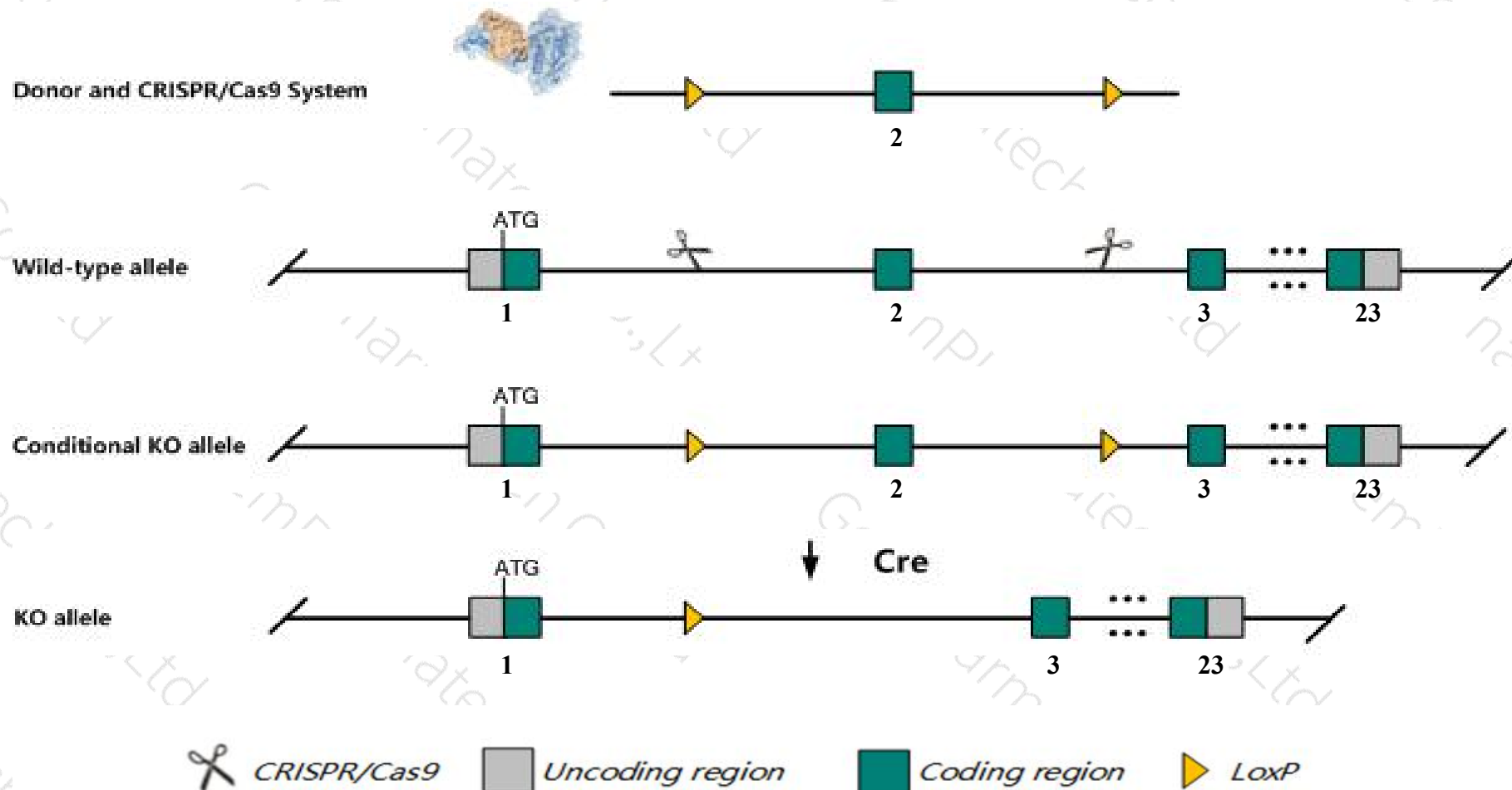
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Lrp5* gene. The schematic diagram is as follows:



Technical routes

- The *Lrp5* gene has 4 transcripts. According to the structure of *Lrp5* gene, exon2 of *Lrp5-201* (ENSMUST00000025856.16) transcript is recommended as the knockout region. The region contains 397bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Lrp5* 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.

- According to the existing MGI data, Homozygous mutants show variable bone loss, decreased osteoblast proliferation, impaired glucose tolerance, increased plasma cholesterol on high-fat diet and persistent embryonic eye vascularization, depending on allelic combination and strain background.
- Transcript *Lrp5*-203 may not be affected.
- The *Lrp5* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Lrp5 low density lipoprotein receptor-related protein 5 [Mus musculus (house mouse)]

Gene ID: 16973, updated on 9-Apr-2019

Summary



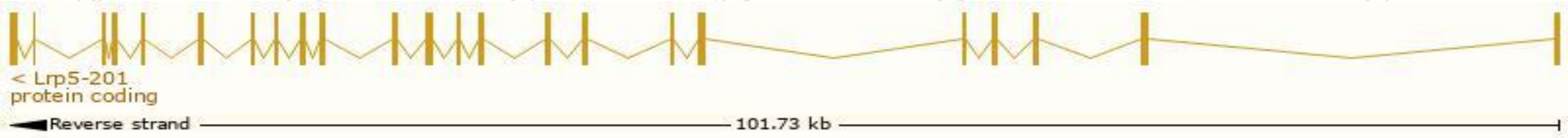
Official Symbol	Lrp5 provided by MGI
Official Full Name	low density lipoprotein receptor-related protein 5 provided by MGI
Primary source	MGI:MGI:1278315
See related	Ensembl:ENSMUSG00000024913
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BMND1, HBM, LR3, LRP7, OPPG, mKIAA4142
Expression	Ubiquitous expression in adrenal adult (RPKM 38.3), lung adult (RPKM 30.1) and 24 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

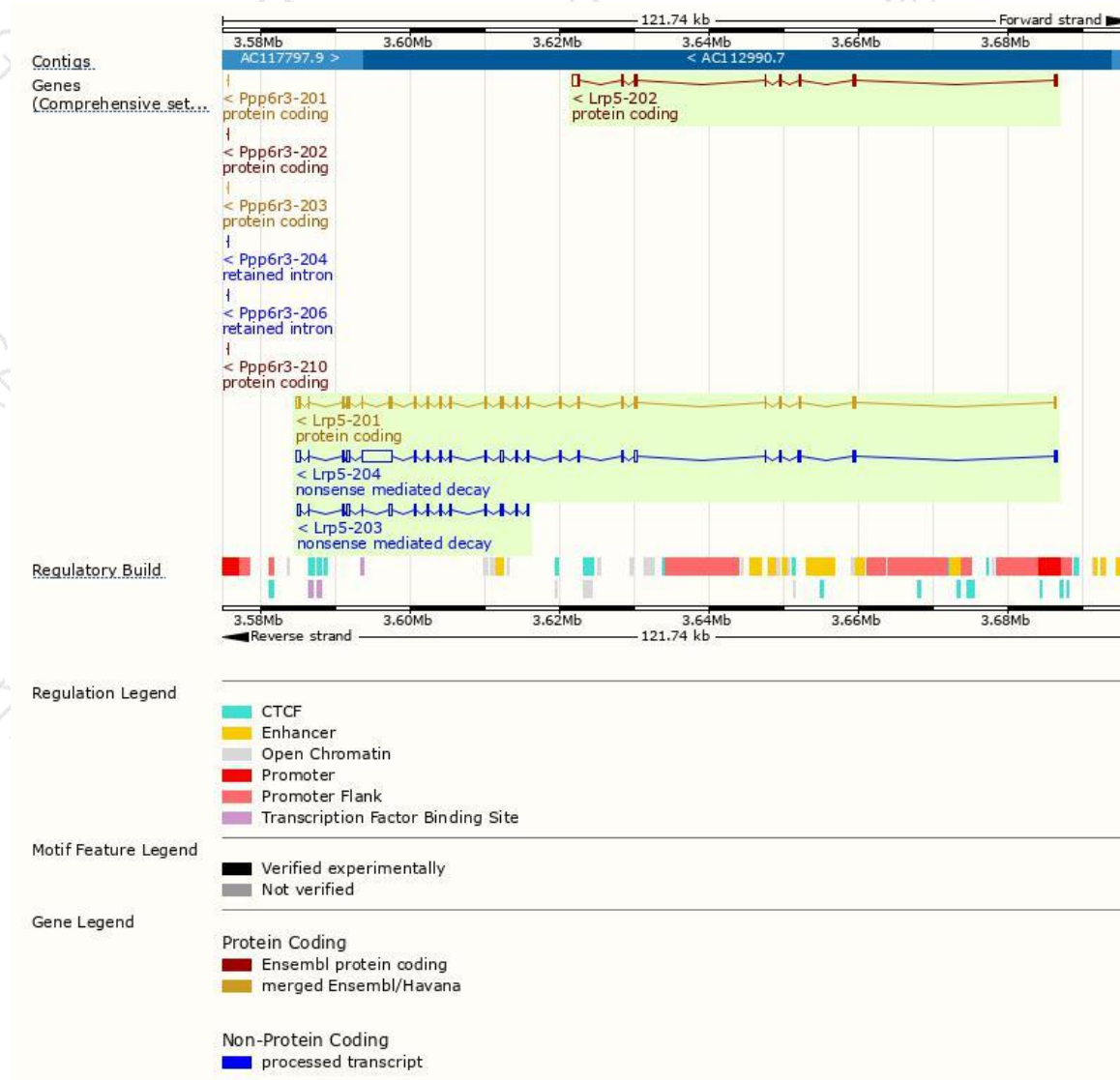
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Lrp5-201	ENSMUST00000025856.16	5161	1614aa	Protein coding	CCDS37881	Q91VN0	TSL:1 GENCODE basic APPRIS P1
Lrp5-202	ENSMUST00000176867.1	2759	618aa	Protein coding	-	H3BL52	TSL:1 GENCODE basic
Lrp5-204	ENSMUST00000177330.7	8893	279aa	Nonsense mediated decay	-	H3BJH0	TSL:2
Lrp5-203	ENSMUST00000177294.1	2835	226aa	Nonsense mediated decay	-	H3BIY3	CDS 5' incomplete TSL:1

The strategy is based on the design of *Lrp5-201* transcript,The transcription is shown below



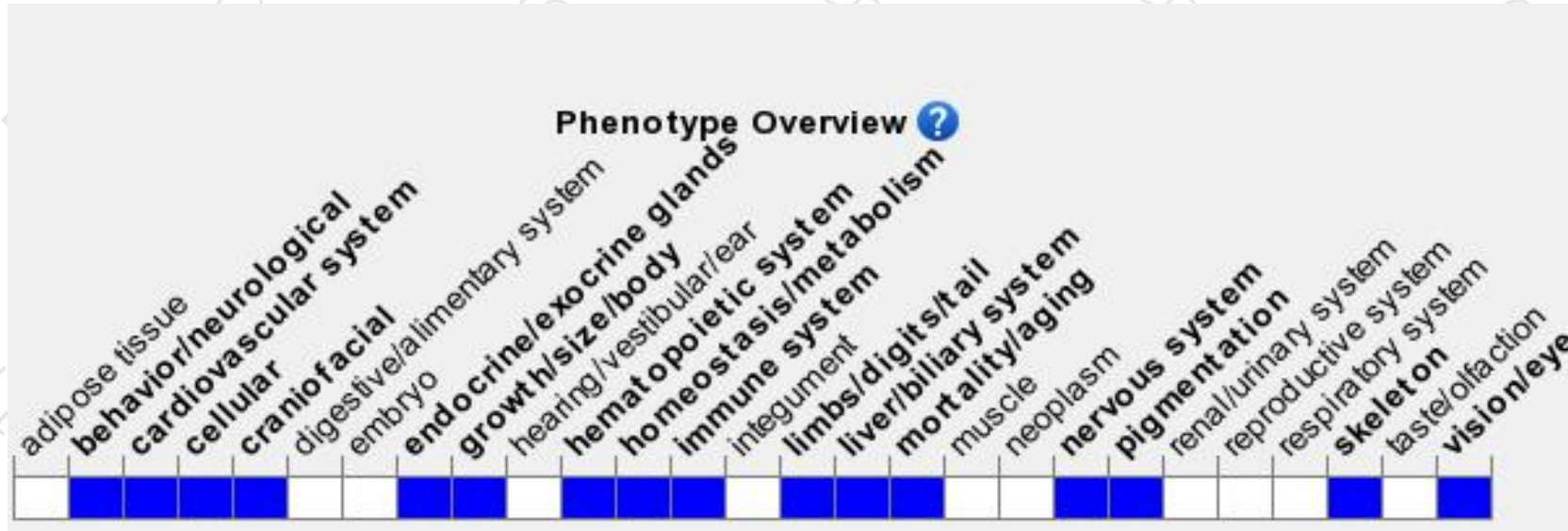
Genomic location distribution



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 mutants show variable bone loss, decreased osteoblast proliferation, impaired glucose tolerance, increased plasma cholesterol on high-fat diet and persistent embryonic eye vascularization, depending on allelic combination and strain background.

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

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