

Acsl5 Cas9-CKO Strategy

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Overview

Target Gene Name

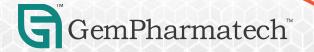
• Acsl5

Project Type

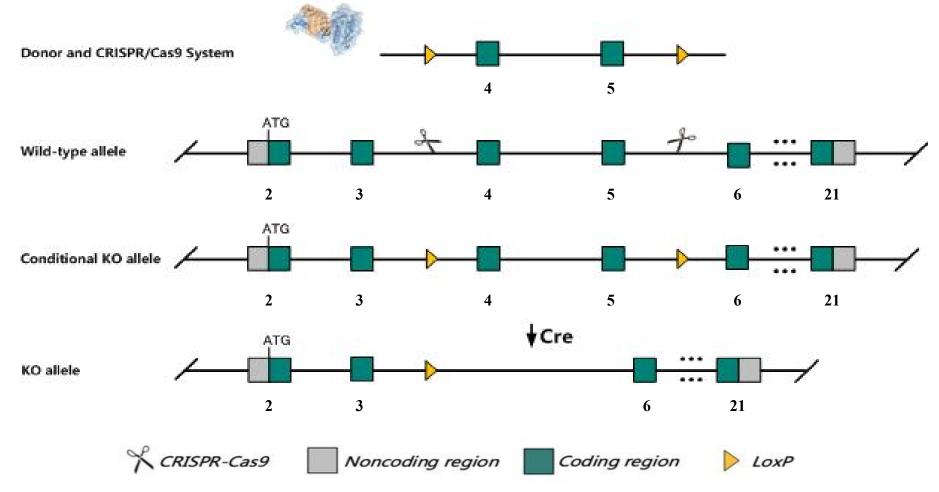
• Cas9-CKO

Genetic Background

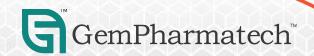
• C57BL/6JGpt



Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the Acsl5 gene.



Technical Information

- The *Acsl5* gene has 6 transcripts. According to the structure of *Acsl5* gene, exon4-exon5 of *Acsl5*-201 (ENSMUST00000043150.6) transcript is recommended as the knockout region. The region contains 167bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Acsl5* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.



Gene Information

Acsl5 acyl-CoA synthetase long-chain family member 5 [Mus musculus (house mouse)]

5). [provided by Alliance of Genome Resources, Apr 2022]

Gene ID: 433256, updated on 18-May-2023

Orthologs human all

Expression Broad expression in liver E18 (RPKM 62.0), large intestine adult (RPKM 51.5) and 23 other tissuesSee more

Source: https://www.ncbi.nlm.nih.gov/



Transcript Information

The gene has 6 transcripts, all transcripts are shown below:

Transcript ID	Name 🍦	bp 🌲	Protein 🍦	Biotype	CCDS	UniProt Match	Flags
ENSMUST00000043150.6	Acsl5-201	4243	<u>683aa</u>	Protein coding	CCDS29908 ₽	Q3UC67 ₢ Q8JZR0 ₢	Ensembl Canonical GENCODE basic APPRIS P1 TSL:1
ENSMUST00000225551.2	AcsI5-205	520	<u>117aa</u>	Protein coding		A0A286YD68 ₽	CDS 3' incomplete
ENSMUST00000224414.2	AcsI5-203	455	<u>96aa</u>	Protein coding		A0A286YCG4₺	CDS 5' incomplete
ENSMUST00000225454.2	AcsI5-204	432	<u>62aa</u>	Protein coding		A0A286YCF3₽	CDS 5' incomplete
ENSMUST00000224337.2	Acsl5-202	529	<u>95aa</u>	Nonsense mediated decay		A0A286YE29₽	
ENSMUST00000226019.2	Acsl5-206	358	No protein	Retained intron		(2)	2

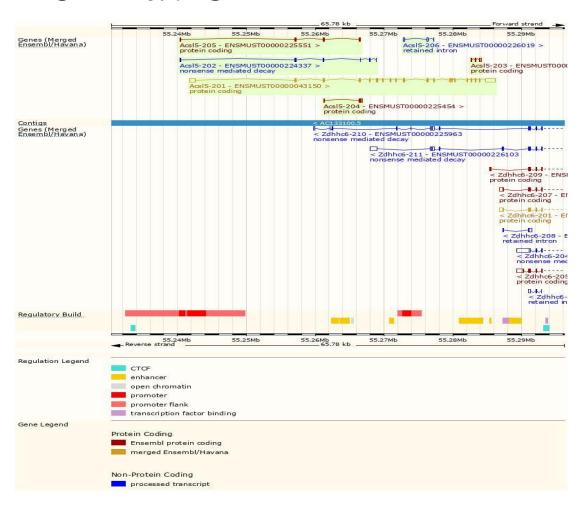
The strategy is based on the design of *Acsl5*-201 transcript, the transcription is shown below:



Source: https://www.ensembl.org



Genomic Information





Source:: https://www.ensembl.org

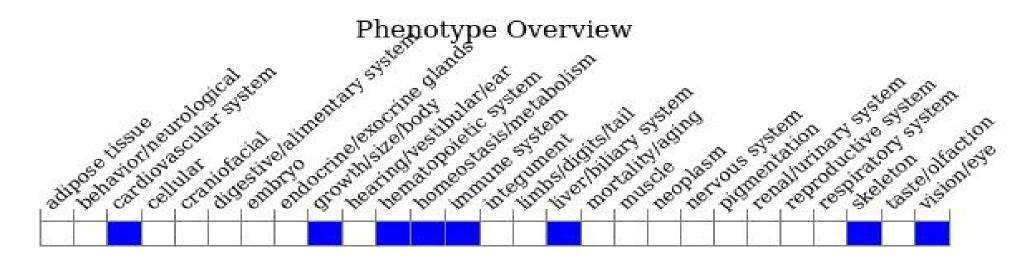
Protein Information





Source: : https://www.ensembl.org

Mouse Phenotype Information (MGI)



• Homozygous mutant mice exhibit decreased mean bone mineral content and density measurements when compared with controls. A notably decreased mean platelet count is also observed.



Important Information

- According to MGI, homozygous mutant mice exhibit decreased mean bone mineral content and density measurements when compared with controls. A notably decreased mean platelet count is also observed.
- The effect of this strategy on the transcripts of this gene *Acsl5*-203, *Acsl5*-204 and *Acsl5*-205 are unknown.
- The introns of the gene Zdhhc6 overlap in the knockout region, and the effect on the gene Zdhhc6 is unknown.
- *Acsl5* is located on Chr19. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.

