

Acad8 Cas9-CKO Strategy

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Date:2020-03-16

Project Overview



Project Name

Acad8

Project type

Cas9-CKO

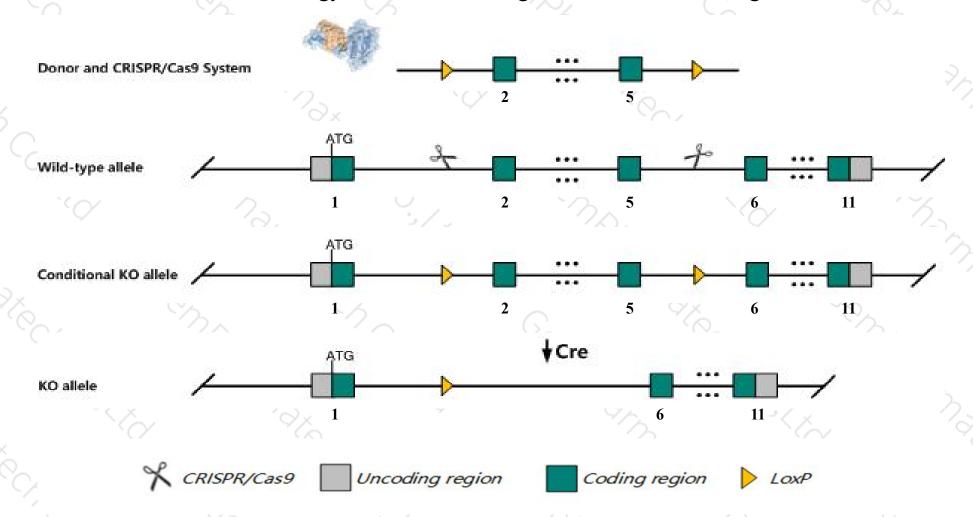
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Acad8 gene. The schematic diagram is as follows:



Technical routes



- The Acad8 gene has 8 transcripts. According to the structure of Acad8 gene, exon2-exon5 of Acad8-201 (ENSMUST00000060513.7) transcript is recommended as the knockout region. The region contains 458bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Acad8* 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, Mice homozygous for an ENU-induced allele exhibit cold intolerance at young age with a progressive hepatic steatosis and abnormal mitochondria.
- The floxed region is near to the N-terminal of *Thyn1* gene, this strategy may influence the regulatory function of the N-terminal of *Thyn1* gene.
- > Transcript *Acad8*-207 may not be affected.
- > The Acad8 gene is located on the Chr9. 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)



Acad8 acyl-Coenzyme A dehydrogenase family, member 8 [Mus musculus (house mouse)]

Gene ID: 66948, updated on 13-Mar-2020

Summary



Official Symbol Acad8 provided by MGI

Official Full Name acyl-Coenzyme A dehydrogenase family, member 8 provided by MGI

Primary source MGI:MGI:1914198

See related Ensembl: ENSMUSG00000031969

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 Al786953; 2310016C19Rik

Expression Ubiquitous expression in kidney adult (RPKM 8.3), genital fat pad adult (RPKM 7.1) and 28 other tissues <u>See more</u>

Orthologs human all

Genomic context



Location: 9; 9 A4

See Acad8 in Genome Data Viewer

Exon count: 11

Annotation release	Status	Assembly	Chr	Location	
108	current	GRCm38.p6 (GCF_000001635.26)	9	NC_000075.6 (2697413826999571, complement)	
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	9	NC_000075.5 (2678172326807134, complement)	

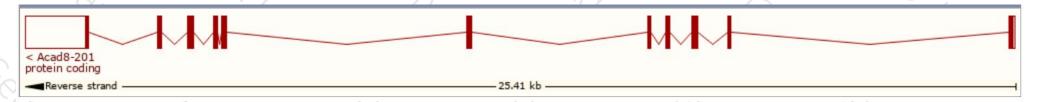
Transcript information (Ensembl)



The gene has 8 transcripts, all transcripts are shown below:

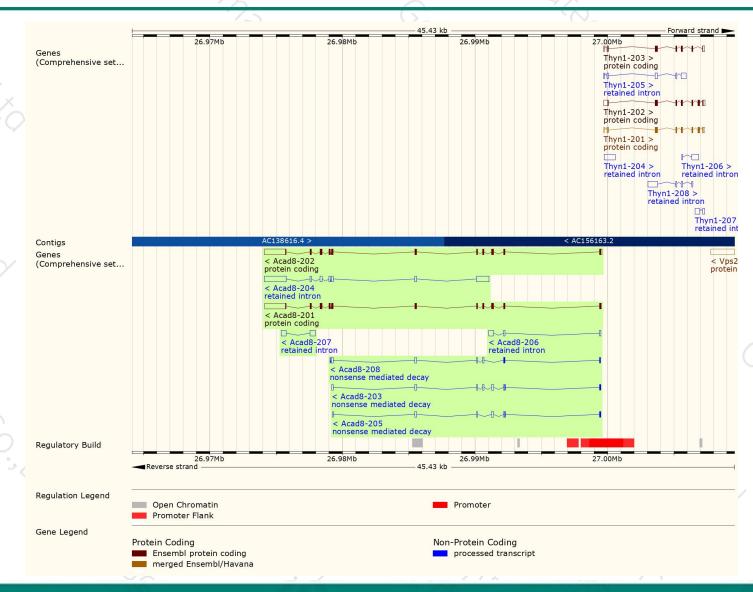
Name	Transcript ID 🔷	bp 🍦	Protein	Biotype	CCDS	UniProt 🝦	Flags
Acad8-201	ENSMUST00000060513.7	2859	<u>413aa</u>	Protein coding	CCDS40567 &	A0A0R4J0P1&	TSL:1 GENCODE basic APPRIS P2
Acad8-202	ENSMUST00000120367.7	2885	413aa	Protein coding	. 	D3YTT4 密	TSL:5 GENCODE basic APPRIS ALT2
Acad8-203	ENSMUST00000128923.7	852	49aa	Nonsense mediated decay	-	D6RDD5&	TSL:5
Acad8-205	ENSMUST00000132293.1	679	49aa	Nonsense mediated decay	(E)	D6RDD5@	TSL:3
Acad8-208	ENSMUST00000215693.1	661	<u>66aa</u>	Nonsense mediated decay	(=)	A0A1L1SUG2귣	CDS 5' incomplete TSL:5
Acad8-204	ENSMUST00000129490.7	3188	No protein	Retained intron	-	-	TSL:5
Acad8-207	ENSMUST00000151075.1	785	No protein	Retained intron	-	-	TSL:1
Acad8-206	ENSMUST00000138102.1	600	No protein	Retained intron	A 0	0 0 0	TSL:2

The strategy is based on the design of Acad8-201 transcript, The transcription is shown below



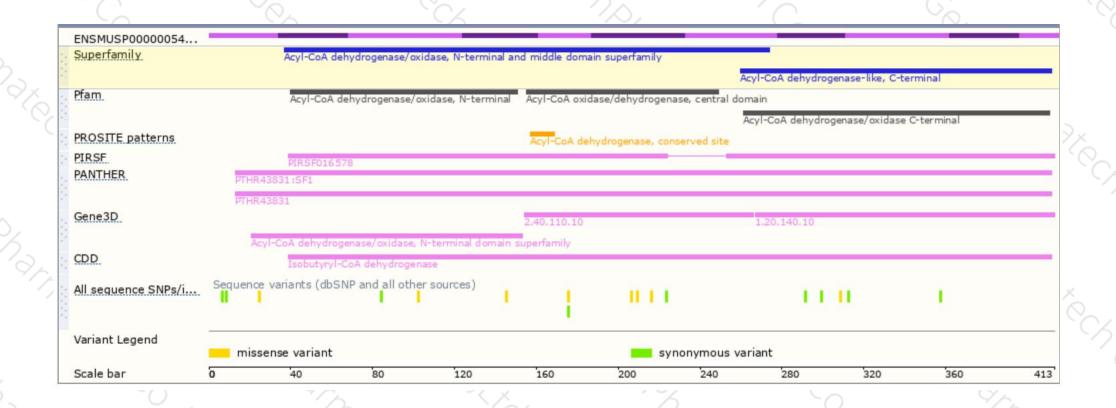
Genomic location distribution





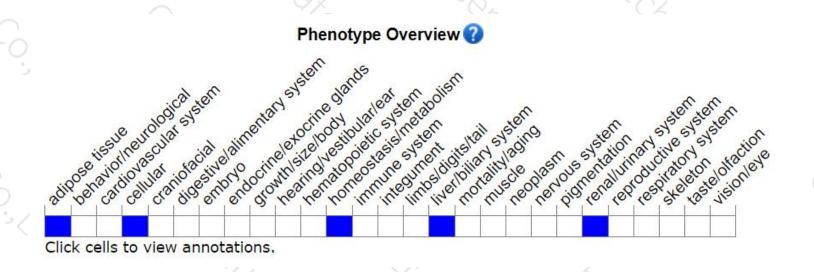
Protein domain





Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

Mice homozygous for an ENU-induced allele exhibit cold intolerance at young age with a progressive hepatic steatosis and abnormal mitochondria.



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





