

Atp8a1 Cas9-CKO Strategy

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Reviewer:JiaYu

Design Date:2020-2-12

Project Overview



Project Name

Atp8a1

Project type

Cas9-CKO

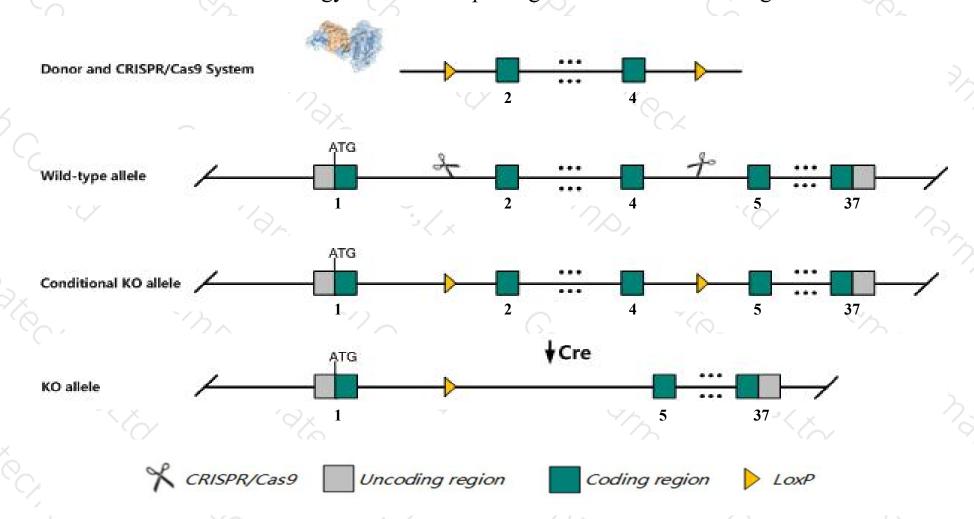
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Atp8a1 gene. The schematic diagram is as follows:



Technical routes



- The *Atp8a1* gene has 14 transcripts. According to the structure of *Atp8a1* gene, exon2-exon4 of *Atp8a1-201* (ENSMUST00000037380.14) transcript is recommended as the knockout region. The region contains 314bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Atp8a1* 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 mutant mice are viable, fertile and phenotypically normal.
- The *Atp8a1* gene is located on the Chr5. 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)



Atp8a1 ATPase, aminophospholipid transporter (APLT), class I, type 8A, member 1 [Mus musculus (house mouse)]

Gene ID: 11980, updated on 4-Dec-2019

Summary

Official Symbol Atp8a1 provided by MGI

Official Full Name ATPase, aminophospholipid transporter (APLT), class I, type 8A, member 1 provided by MGI

Primary source MGI:MGI:1330848

See related Ensembl: ENSMUSG00000037685

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 APLT; Atp3a2; Classl; Al481521; Al853962; AW743152; AW822227; B230107D19Rik

Expression Broad expression in lung adult (RPKM 16.4), cortex adult (RPKM 15.1) and 26 other tissues See more

Orthologs human all

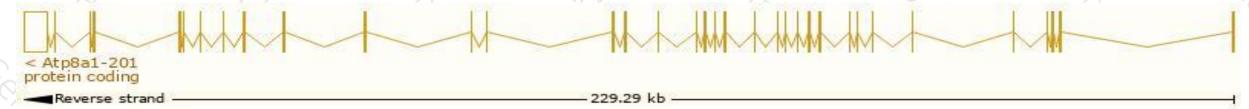
Transcript information (Ensembl)



The gene has 14 transcripts, all transcripts are shown below:

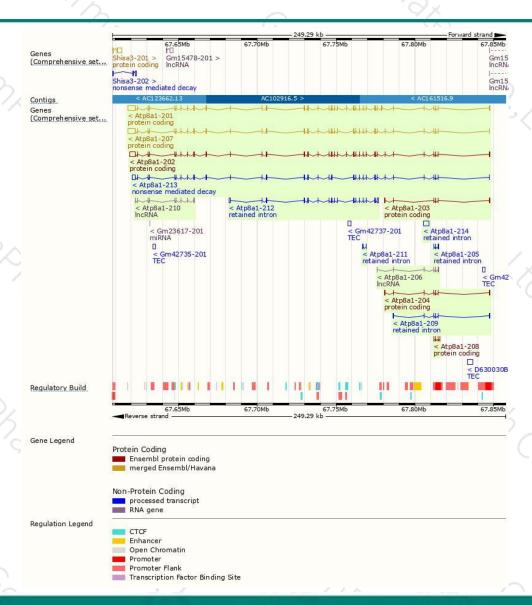
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Atp8a1-201	ENSMUST00000037380.14	8178	1164aa	Protein coding	CCDS39104	P70704	TSL:1 GENCODE basic APPRIS P4
Atp8a1-207	ENSMUST00000135930.7	8069	<u>1149aa</u>	Protein coding	CCDS39105	P70704	TSL:1 GENCODE basic APPRIS ALT1
Atp8a1-202	ENSMUST00000072971.12	7154	<u>1164aa</u>	Protein coding	CCDS80295	A0A0M3HEP7	TSL:1 GENCODE basic APPRIS ALT2
Atp8a1-204	ENSMUST00000113652.7	854	<u>175aa</u>	Protein coding	92	<u>Q3U9N5</u>	TSL:1 GENCODE basic
Atp8a1-203	ENSMUST00000113651.7	808	<u>122aa</u>	Protein coding	-	E9Q4J5	TSL:5 GENCODE basic
Atp8a1-208	ENSMUST00000141443.1	368	106aa	Protein coding) to	F6Q8P7	CDS 3' incomplete TSL:5
Atp8a1-213	ENSMUST00000200955,3	5373	441aa	Nonsense mediated decay	-	A0A0J9YV34	TSL:5
Atp8a1-214	ENSMUST00000202248.1	3360	No protein	Retained intron	100	20	TSL:NA
Atp8a1-212	ENSMUST00000155911.1	2932	No protein	Retained intron	-	-	TSL:1
Atp8a1-209	ENSMUST00000143013.7	1014	No protein	Retained intron	8-		TSL:2
Atp8a1-205	ENSMUST00000128726.1	750	No protein	Retained intron	-	2	TSL:5
Atp8a1-211	ENSMUST00000152433.1	617	No protein	Retained intron	100	20	TSL:3
Atp8a1-210	ENSMUST00000149501.1	762	No protein	IncRNA	-	-	TSL:3
Atp8a1-206	ENSMUST00000130652.7	469	No protein	IncRNA	-		TSL:5
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The strategy is based on the design of Atp8a1-201 transcript, The transcription is shown below



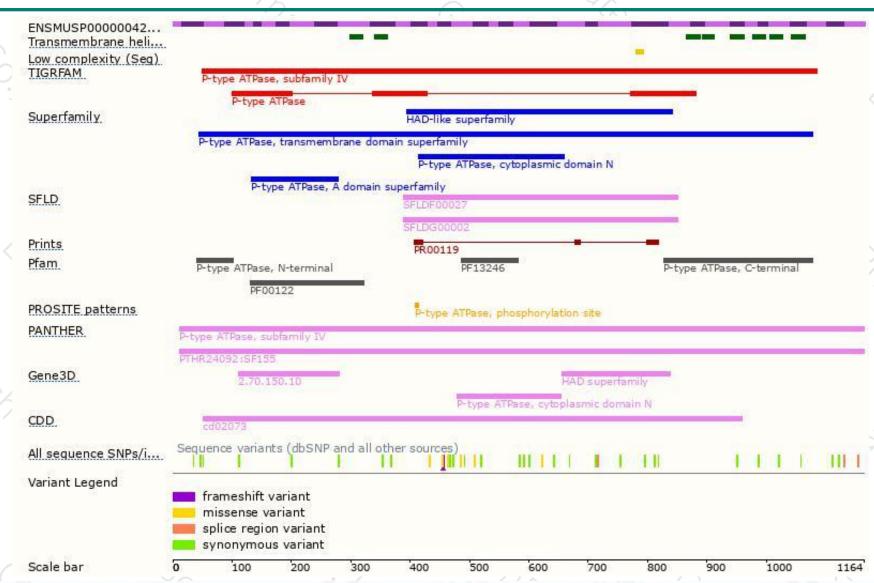
Genomic location distribution





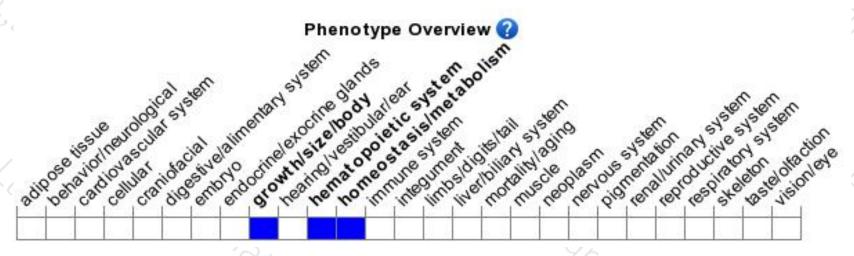
Protein domain





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 mutant mice are viable, fertile and phenotypically normal.



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





