

Atg12 Cas9-KO Strategy

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Project Overview



Project Name Atg12

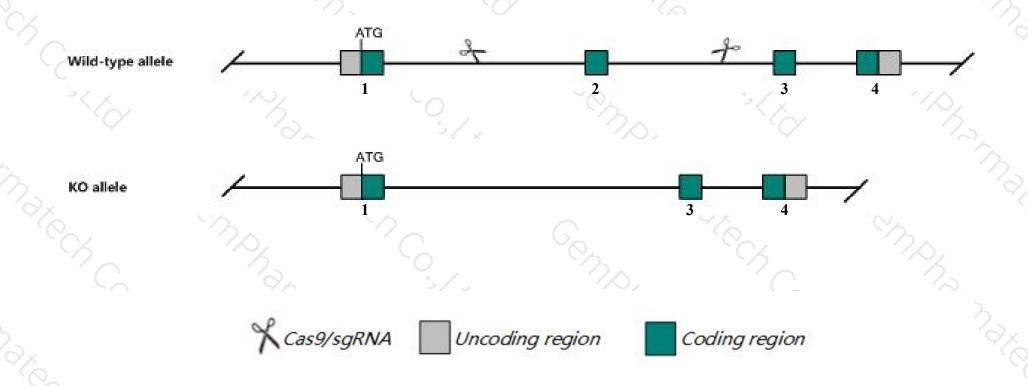
Project type Cas9-KO

Strain background C57BL/6J

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Atg12 gene. The schematic diagram is as follows:



Technical routes



- ➤ The Atg12 gene has 3 transcripts. According to the structure of Atg12 gene, exon2 of Atg12-201

 (ENSMUST00000035648.5) transcript is recommended as the knockout region. The region contains 137bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Atg12* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

Notice



- ➤ According to the existing MGI data, Mice homozygous for a knock-out allele exhibit neonatal lethality.

 Mice homozygous for a conditional allele activated in POMC neurons exhibit increased diet-induced obesity.
- ➤ The *Atg12* gene is located on the Chr18. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Atg12 autophagy related 12 [Mus musculus (house mouse)]

Gene ID: 67526, updated on 31-Jan-2019

Summary

☆ ?

Official Symbol Atg12 provided by MGI

Official Full Name autophagy related 12 provided by MGI

Primary source MGI:MGI:1914776

See related Ensembl: ENSMUSG00000032905

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 4931423H11Rik, A330058M13Rik, Apg12l, Atg12l

Expression Ubiquitous expression in CNS E18 (RPKM 28.2), placenta adult (RPKM 26.0) and 28 other tissuesSee more

Orthologs <u>human</u> all

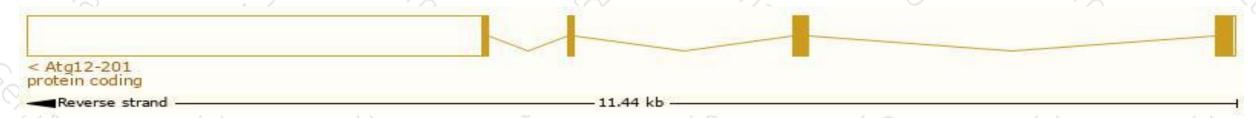
Transcript information (Ensembl)



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

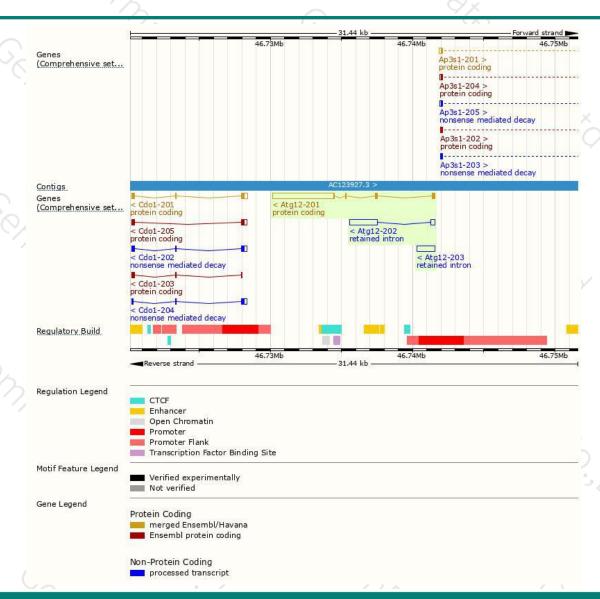
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Atg12-201	ENSMUST00000035648.5	4765	<u>141aa</u>	Protein coding	CCDS37811	Q9CQY1	TSL:1 GENCODE basic APPRIS P1
Atg12-202	ENSMUST00000234430.1	2289	No protein	Retained intron	188		
Atg12-203	ENSMUST00000234830.1	1274	No protein	Retained intron	(2)	(4)	

The strategy is based on the design of Atg12-201 transcript, The transcription is shown below



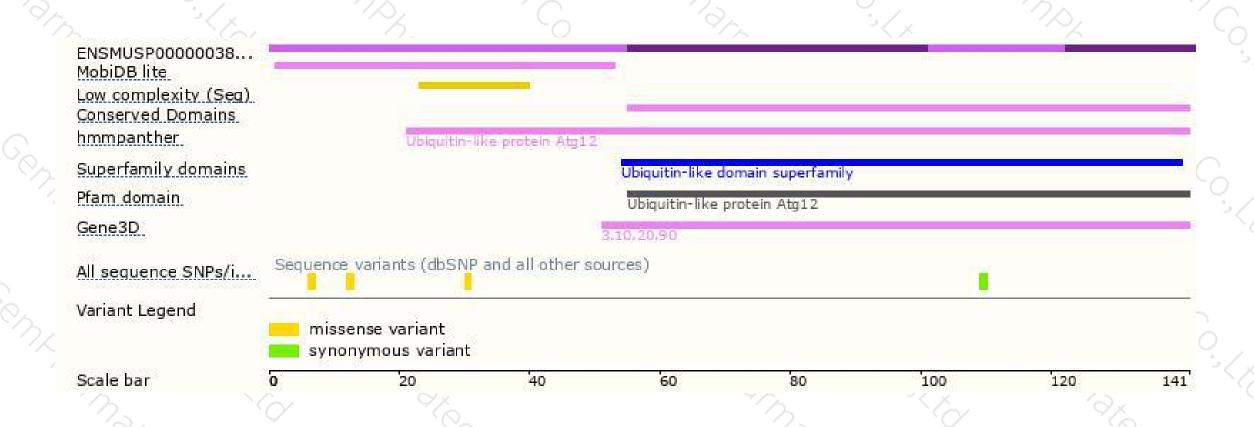
Genomic location distribution





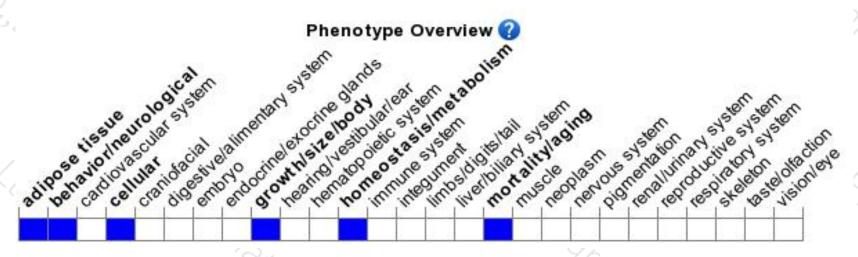
Protein domain





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, Mice homozygous for a knock-out allele exhibit neonatal lethality. Mice homozygous for a conditional allele activated in POMC neurons exhibit increased diet-induced obesity.



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

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