

Atg5 Cas9-CKO Strategy

Designer:

Huan Wang

Design Date:

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Project Overview

Project Name

Atg5

Project type

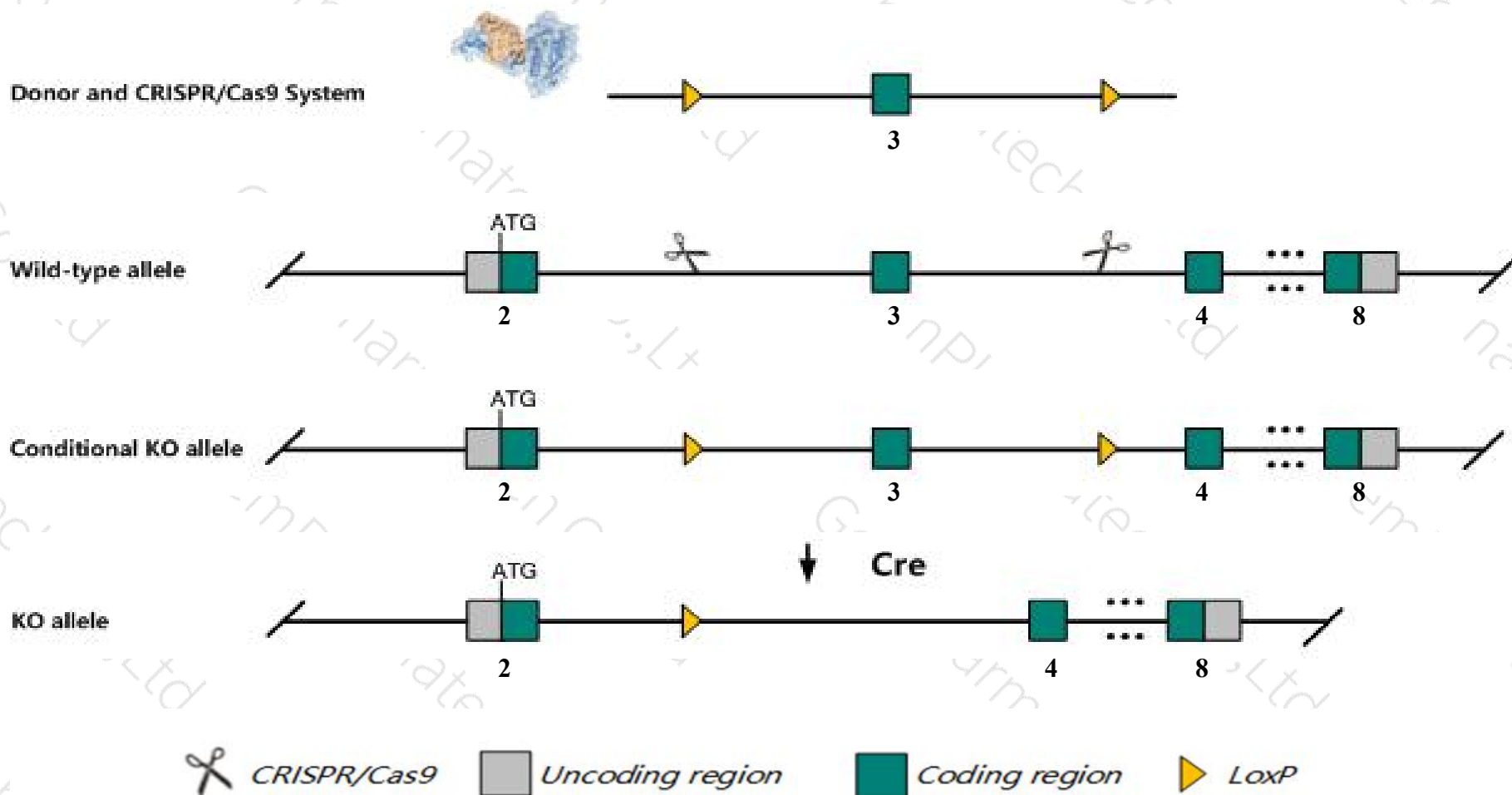
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Atg5* gene. The schematic diagram is as follows:



Technical routes

- The *Atg5* gene has 2 transcripts. According to the structure of *Atg5* gene, exon3 of *Atg5-201* (ENSMUST00000039286.4) transcript is recommended as the knockout region. The region contains 128bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Atg5* 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, Mutation of this gene results in impaired autophagy due to absence of autolysosomes. Homozygotes die within 1 day of birth, have shorter survival times and reduced amino acid levels under fasting conditions. Homozygotes for a gene trap insertion mutation in this gene show no abnormal phenotype.
- The *Atg5* gene is located on the Chr10. 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)

Atg5 autophagy related 5 [Mus musculus (house mouse)]

Gene ID: 11793, updated on 2-Apr-2019

Summary



Official Symbol Atg5 provided by [MGI](#)

Official Full Name autophagy related 5 provided by [MGI](#)

Primary source [MGI:MGI:1277186](#)

See related [Ensembl:ENSMUSG000000038160](#)

Gene type protein coding

RefSeq status REVIEWED

Organism [Mus musculus](#)

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as 2010107M05Rik, 3110067M24Rik, AW319544, Apg5l, Atg5l, C88337, Paddy

Summary The protein encoded by this gene, in combination with autophagy protein 12, functions as an E1-like activating enzyme in a ubiquitin-like conjugating system. The encoded protein is involved in several cellular processes, including autophagic vesicle formation, mitochondrial quality control after oxidative damage, negative regulation of the innate antiviral immune response, lymphocyte development and proliferation, MHC II antigen presentation, adipocyte differentiation, and apoptosis. Two transcript variants encoding different protein isoforms have been found for this gene. [provided by RefSeq, Sep 2015]

Expression Ubiquitous expression in placenta adult (RPKM 9.7), bladder adult (RPKM 9.2) and 28 other tissues [See more](#)

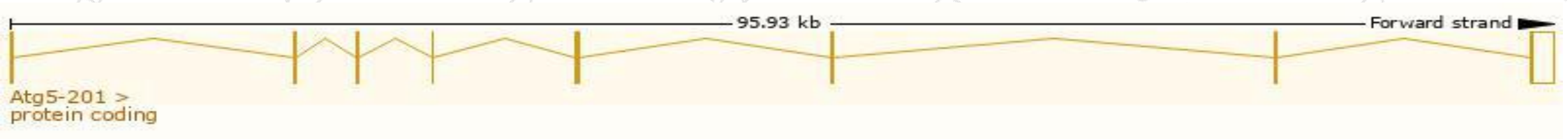
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

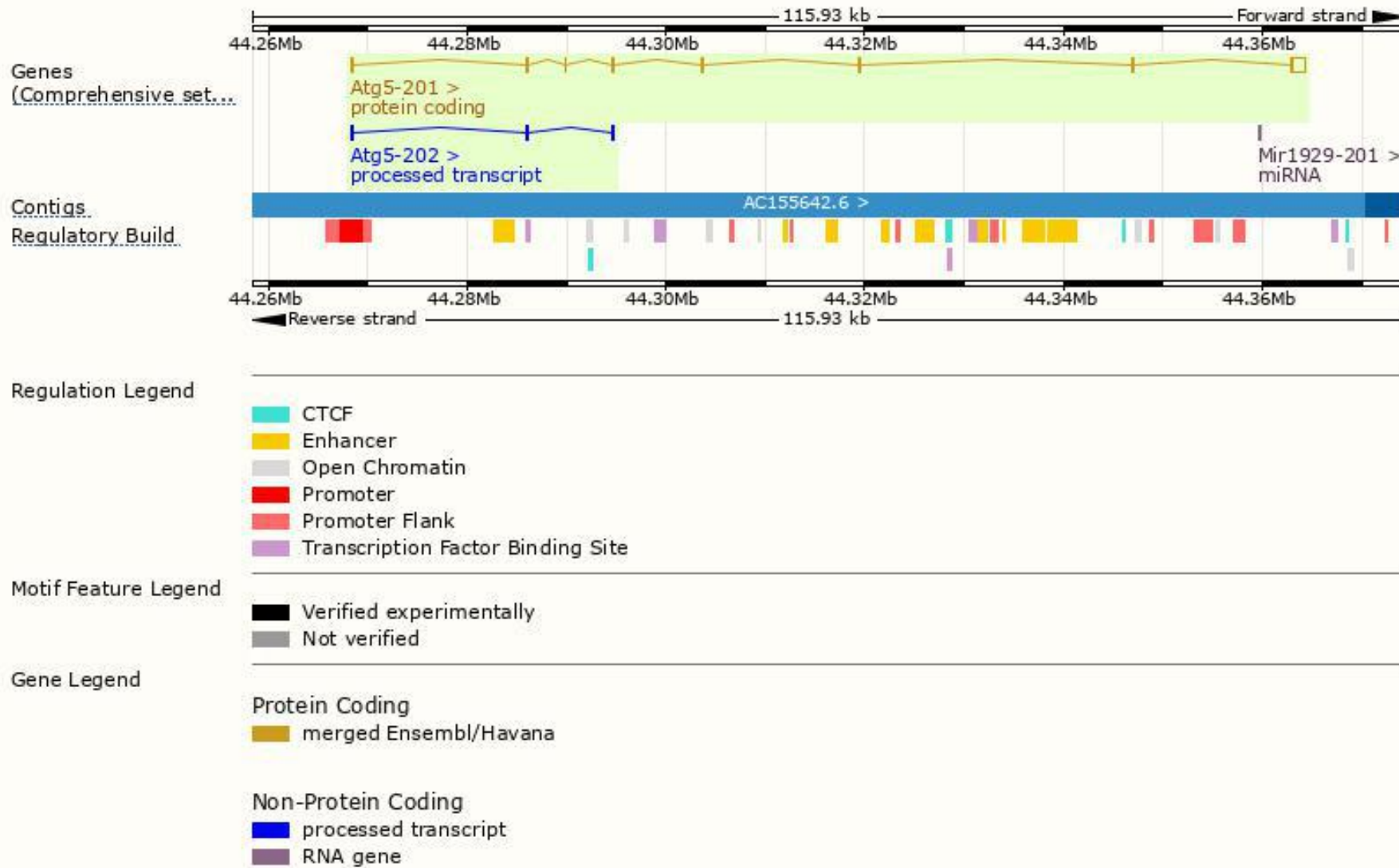
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Atg5-201	ENSMUST00000039286.4	2352	275aa	Protein coding	CCDS23824	Q99J83	TSL:1 GENCODE basic APPRIS P1
Atg5-202	ENSMUST00000217412.1	415	No protein	Processed transcript	-	-	TSL:3

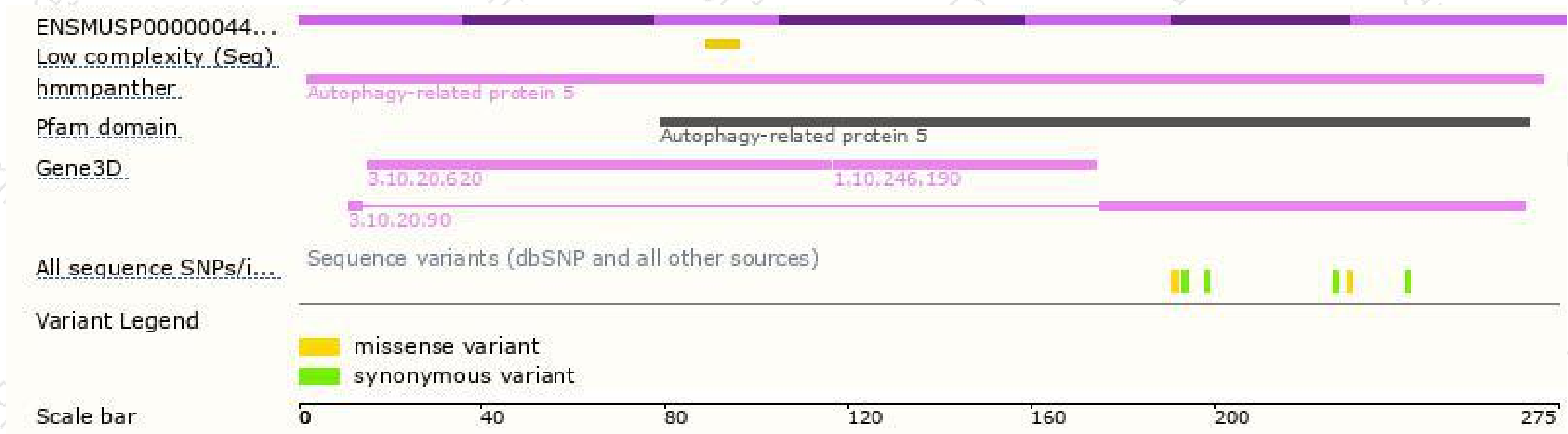
The strategy is based on the design of *Atg5-201* transcript,The transcription is shown below



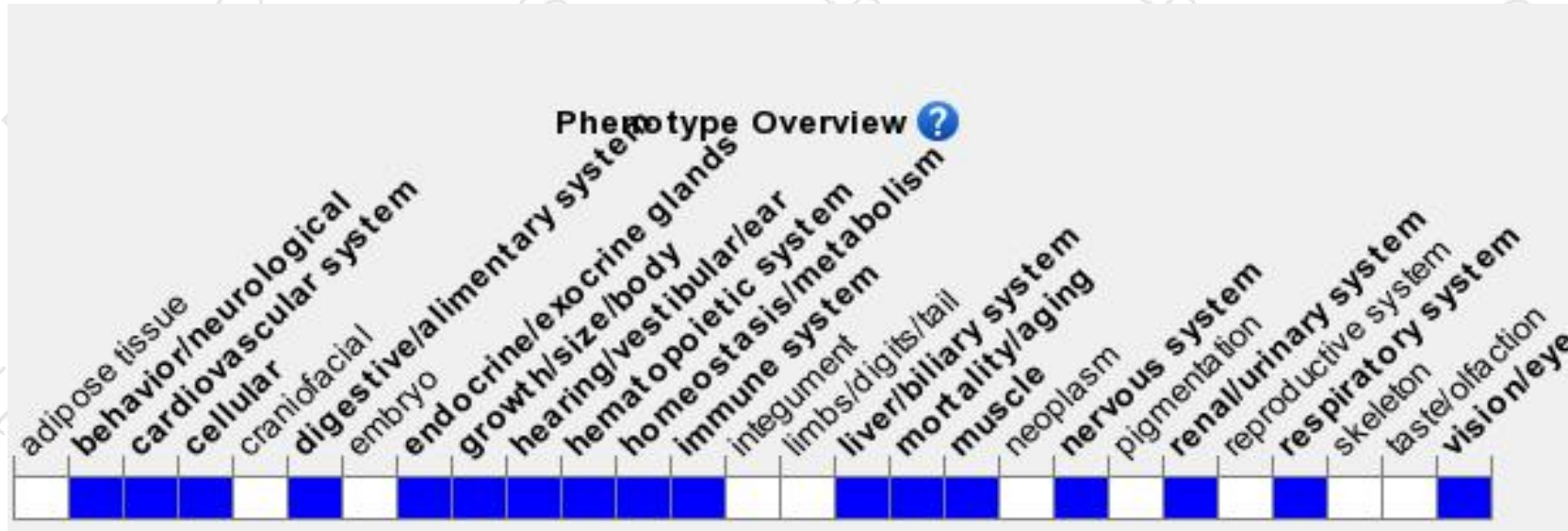
Genomic location distribution



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, Mutation of this gene results in impaired autophagy due to absence of autolysosomes.

Homozygotes die within 1 day of birth, have shorter survival times and reduced amino acid levels under fasting conditions. Homozygotes for a gene trap insertion mutation in this gene show no abnormal phenotype.

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

Tel: 400-9660890

