Atg16l1 Cas9-CKO *Strategy*

Designer: Jinling Wang

Design Date: 2019-7-24

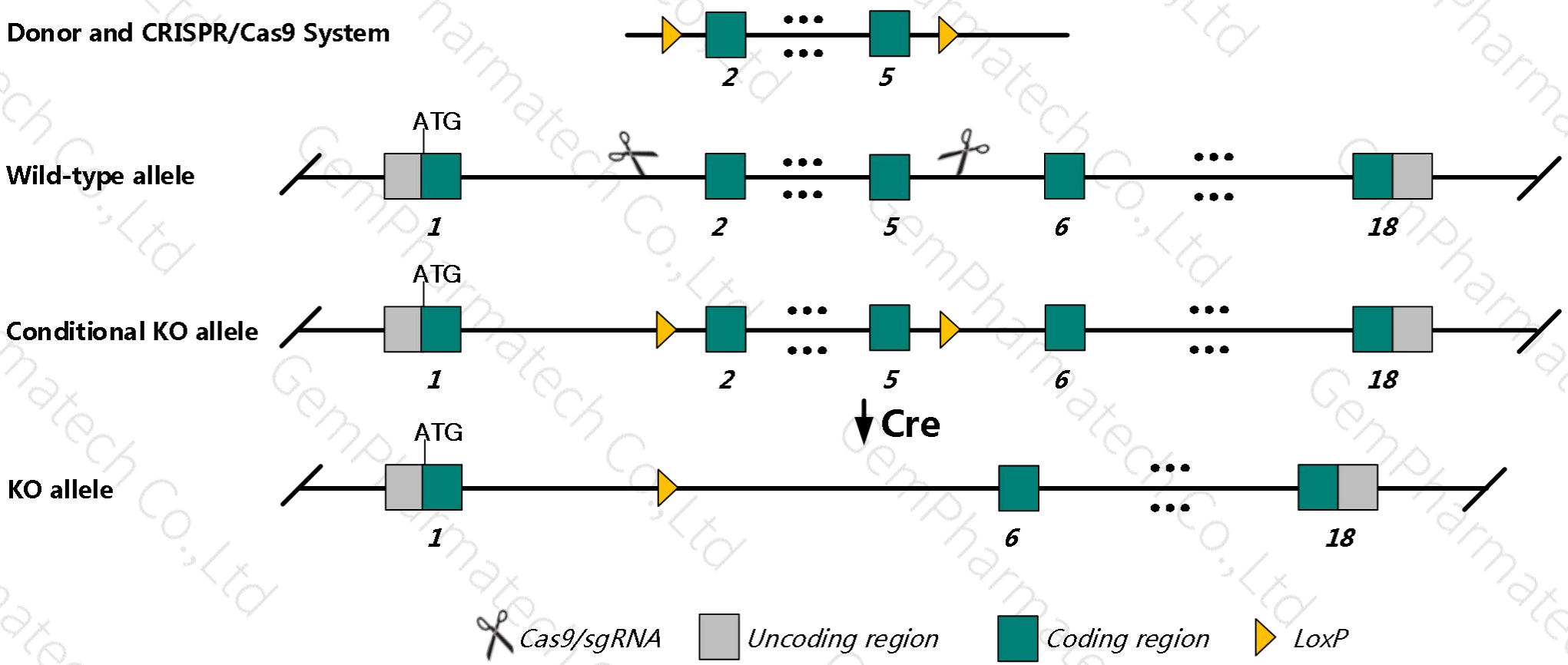
Project Overview

Project Name	***Atg16l1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

集萃药康 GemPharmatech

This model will use CRISPR/Cas9 technology to edit the *Atg16l1* gene. The schematic diagram is as follows:



Technical routes

- The *Atg16l1* gene has 11 transcript.According to the structure of *Atg16l1* gene, exon2-5 of *Atg16l1*-201 (ENSMUST00000027512.12) transcript is recommended as the knockout region.The region contains 526bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Atg16l1* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 or cell types.

Notice

- Null homozygotes have a cellular defect in autophagy that results in lethality during the neonatal starvation period. Mice homozygous for hypomorphic alleles have Paneth cells with aberrant, disorganized granules similar to those found in patients with Crohn's disease.
- The *Atg16l1* gene is located on the Chr1. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

集萃药康 GemPharmatech

Atg16l1 autophagy related 16-like 1 (S. cerevisiae) [*Mus musculus* (house mouse)]

Gene ID: 77040, updated on 8-Dec-2018

Summary

Official Symbol	Atg16l1 provided by MGI
Official Full Name	autophagy related 16-like 1 (S. cerevisiae) provided by MGI
Primary source	MGI:MGI:1924290
See related	Ensembl:ENSMUSG00000026289
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	WDR30; Apg16l; Atg16l; 1500009K01Rik
Expression	Ubiquitous expression in CNS E18 (RPKM 18.9), CNS E14 (RPKM 16.2) and 28 other tissues See more
Orthologs	human all

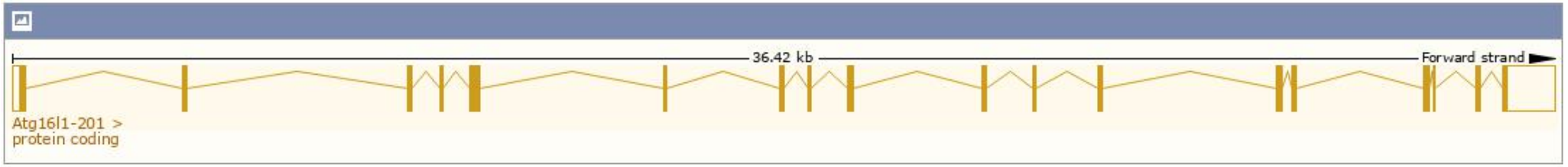
Transcript information（Ensembl）

The gene has 11 transcripts, and all transcripts are shown below:

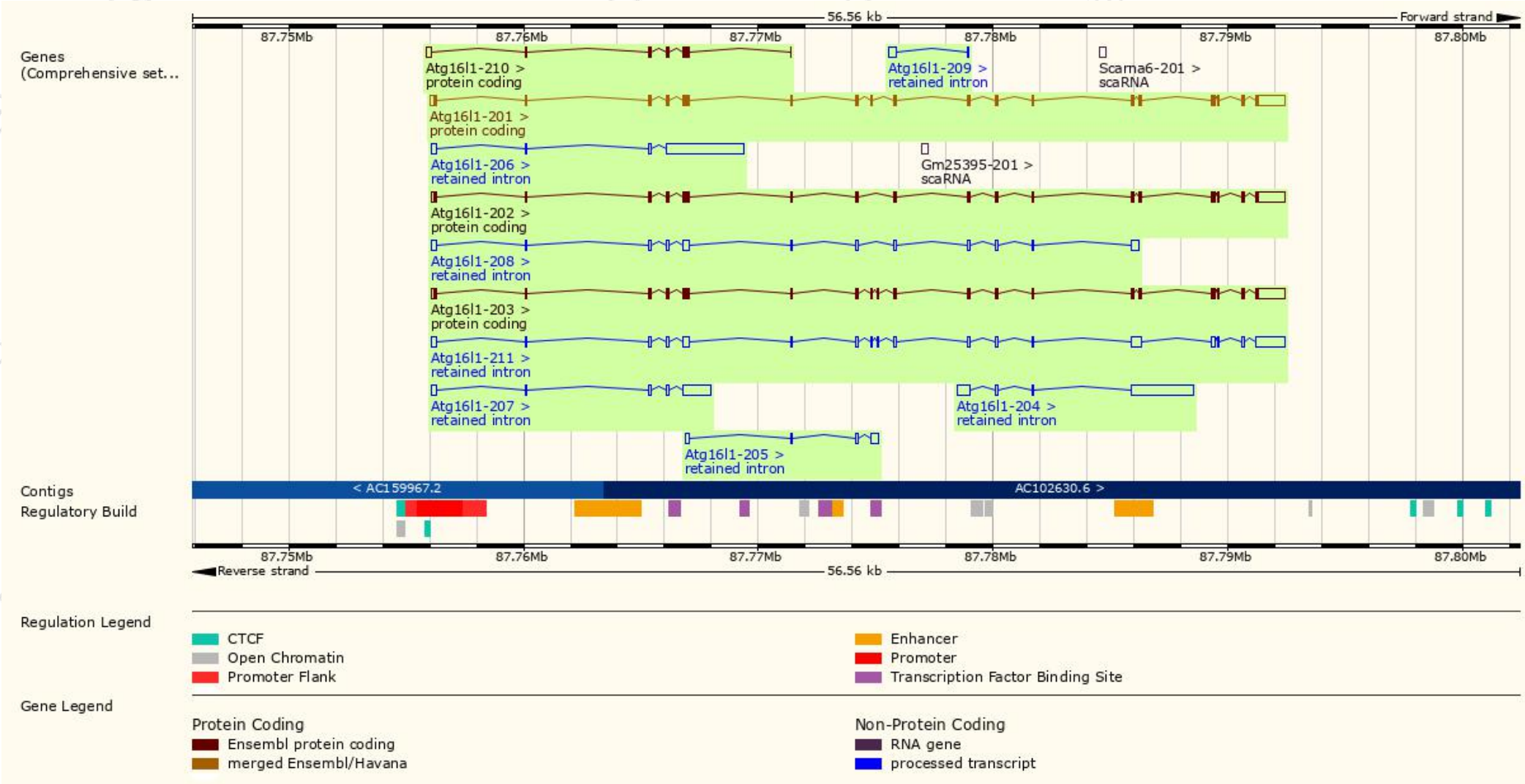
Show/hide columns (1 hidden) Filter

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags
Atg16l1-201	ENSMUST00000027512.12	3130	607aa	Protein coding	CCDS15136	Q8C0J2	NM_029846 NP_084122	TSL:1 GENCODE basic APPRIS P3
Atg16l1-203	ENSMUST00000113190.2	3102	623aa	Protein coding	CCDS56637	G9M4M6 Q8C0J2	NM_001205391 NP_001192320	TSL:1 GENCODE basic APPRIS ALT1
Atg16l1-202	ENSMUST00000113186.7	3004	588aa	Protein coding	CCDS56638	Q3TDQ5 Q8C0J2	NM_001205392 NP_001192321	TSL:1 GENCODE basic APPRIS ALT1
Atg16l1-210	ENSMUST00000144047.7	749	154aa	Protein coding	-	D3YZW7	-	CDS 3' incomplete TSL:5
Atg16l1-206	ENSMUST00000133072.7	3751	No protein	Retained intron	-	-	-	TSL:2
Atg16l1-204	ENSMUST00000127884.1	3400	No protein	Retained intron	-	-	-	TSL:2
Atg16l1-211	ENSMUST00000151037.7	3344	No protein	Retained intron	-	-	-	TSL:2
Atg16l1-207	ENSMUST00000134603.1	1674	No protein	Retained intron	-	-	-	TSL:2
Atg16l1-208	ENSMUST00000137638.7	1594	No protein	Retained intron	-	-	-	TSL:5
Atg16l1-205	ENSMUST00000129431.1	640	No protein	Retained intron	-	-	-	TSL:3
Atg16l1-209	ENSMUST00000139628.1	332	No protein	Retained intron	-	-	-	TSL:3

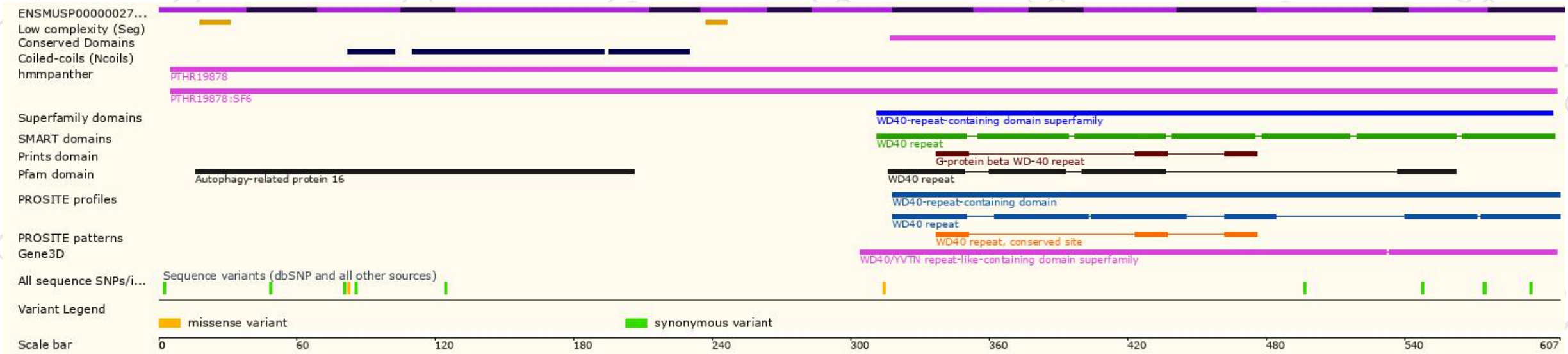
The strategy is based on the design of *Atg16l1*-201 transcript,The transcription is shown below



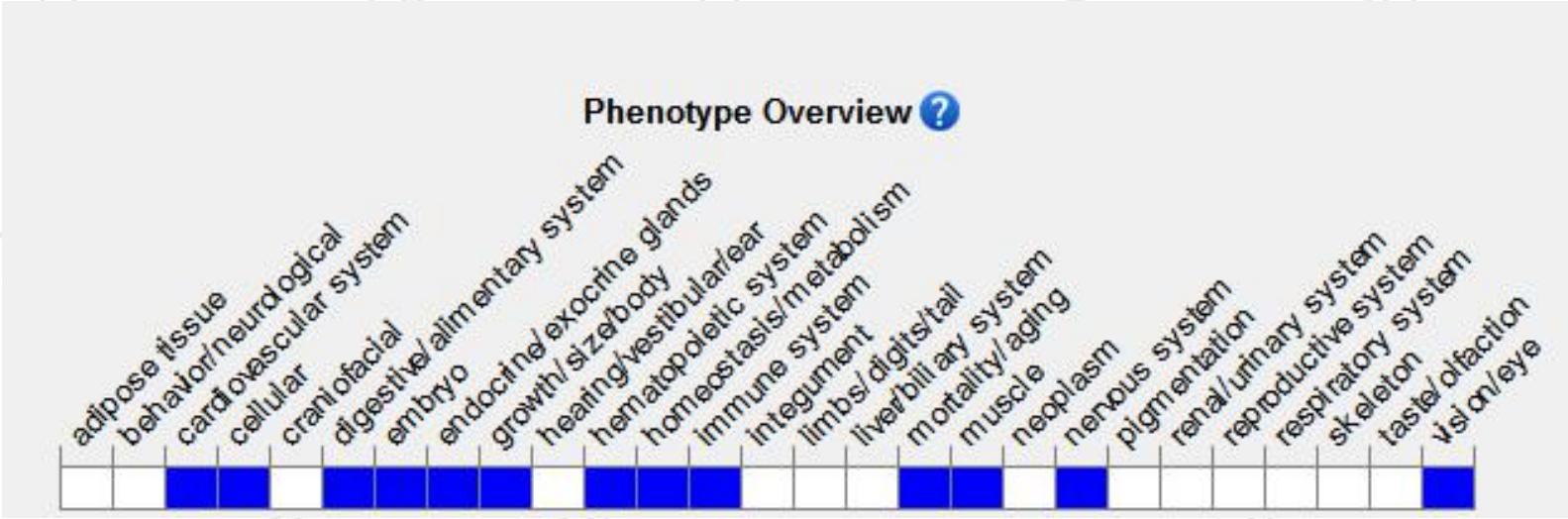
Genomic location distribution



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/) .

Null homozygotes have a cellular defect in autophagy that results in lethality during the neonatal starvation period. Mice homozygous for hypomorphic alleles have Paneth cells with aberrant, disorganized granules similar to those found in patients with Crohn's disease.

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

集萃药康生物科技
GemPharmatech Co.,Ltd