Usp18 Cas9-CKO Strategy

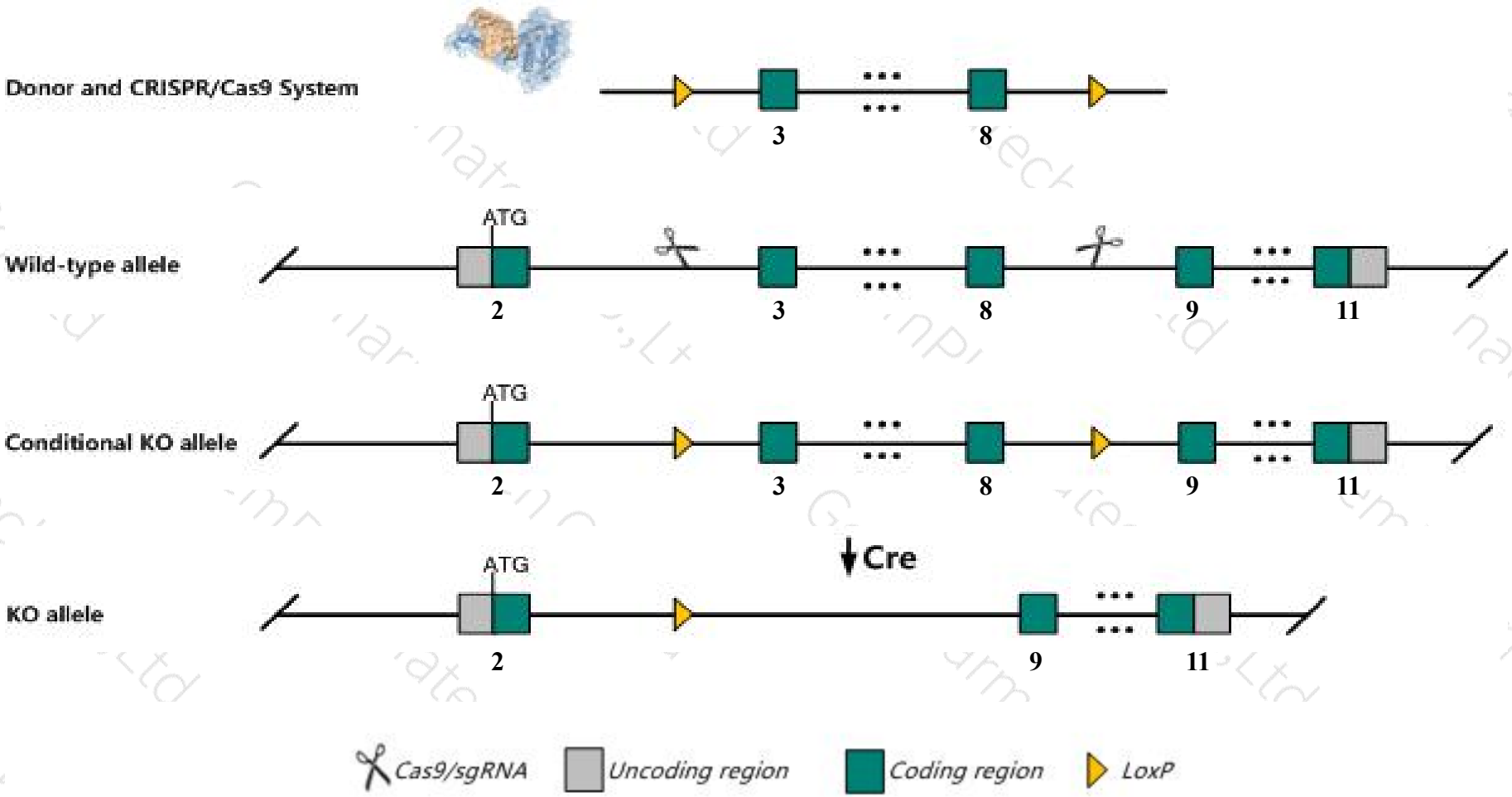
Designer: Xiaojing Li
Design Date: 2019-9-19
Reviewer: JiaYu

Project Overview

Project Name	***Usp18***
Project type	**Cas9-CKO**
Strain background	**C57BL/6J**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Usp18* gene. The schematic diagram is as follows:



Technical routes

- The *Usp18* gene has 4 transcripts. According to the structure of *Usp18* gene, exon3-exon8 of *Usp18-201* (ENSMUST00000032198.10) transcript is recommended as the knockout region.The region contains 731bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Usp18* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA and Donor 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.

- The flox mice was 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 null mutants die prematurely with cellular necrosis in the ependyma, breakdown of blood-brain barrier, hydrocephaly with enlarged ventricles, and severe neurological abnormalities. Mice homozygous for an ENU-induced allele exhibit increased susceptibility to Salmonella infection and LPS.

➢ The *Usp18* gene is located on the Chr6. 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）

集萃药康 GemPharmatech

Usp18 ubiquitin specific peptidase 18 [Mus musculus (house mouse)]

Gene ID: 24110, updated on 3-Feb-2019

Summary

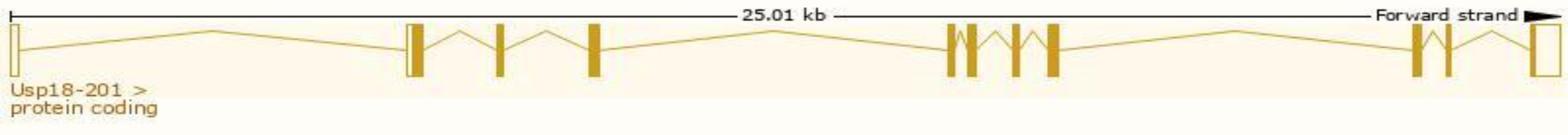
Official Symbol	Usp18 provided by MGI
Official Full Name	ubiquitin specific peptidase 18 provided by MGI
Primary source	MGI:MGI:1344364
See related	Ensembl:ENSMUSG00000030107
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1110058H21Rik, AW047653, UBP43, Ubp15
Expression	Broad expression in liver E18 (RPKM 13.2), thymus adult (RPKM 10.2) and 21 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

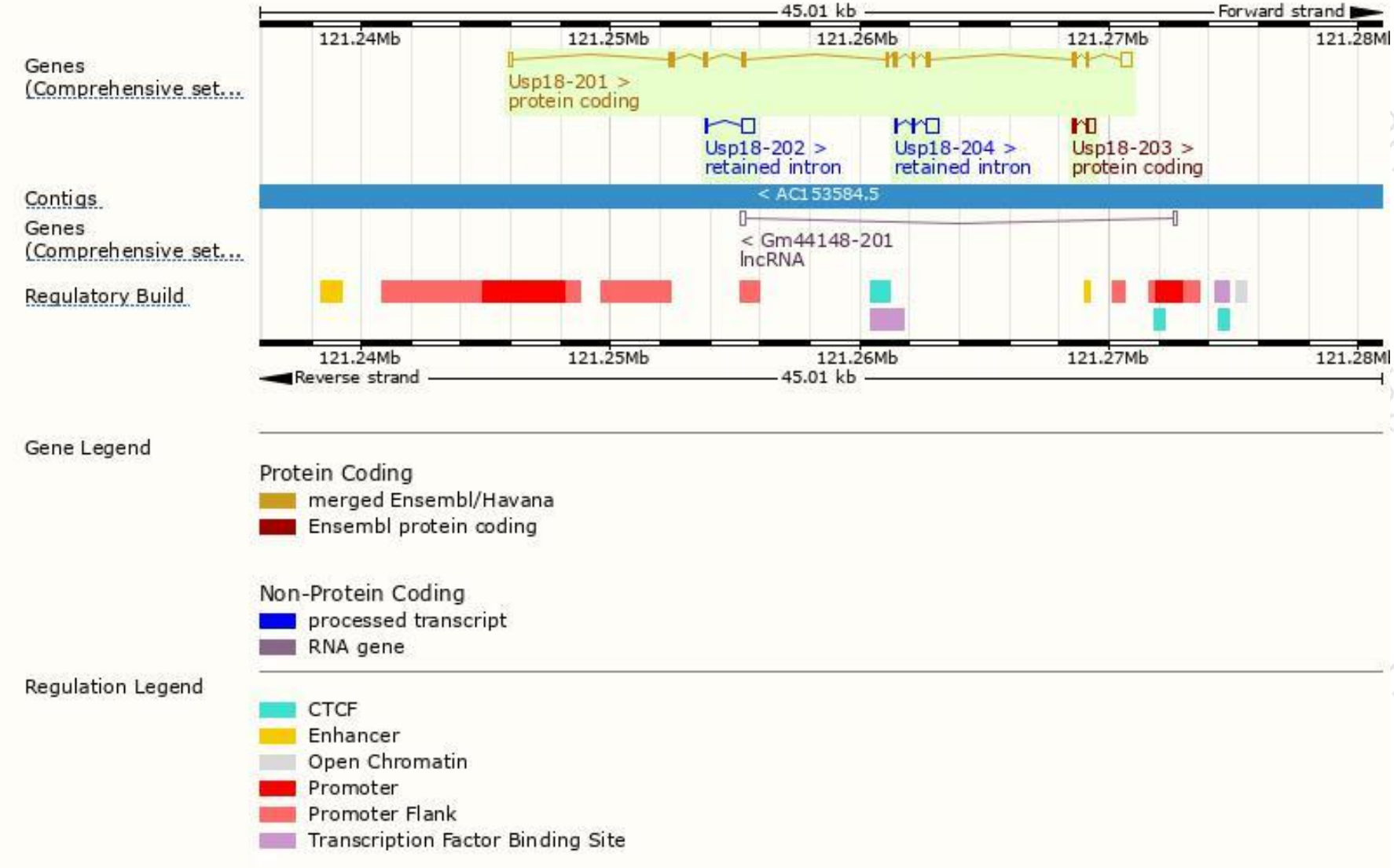
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Usp18-201	ENSMUST00000032198.10	1771	368aa	Protein coding	CCDS20489	Q9WTV6	TSL:1 GENCODE basic APPRIS P1
Usp18-203	ENSMUST00000204710.1	496	60aa	Protein coding	-	A0A0N4SVX5	CDS 5' incomplete TSL:3
Usp18-204	ENSMUST00000204926.1	717	No protein	Retained intron	-	-	TSL:3
Usp18-202	ENSMUST00000203250.1	556	No protein	Retained intron	-	-	TSL:3

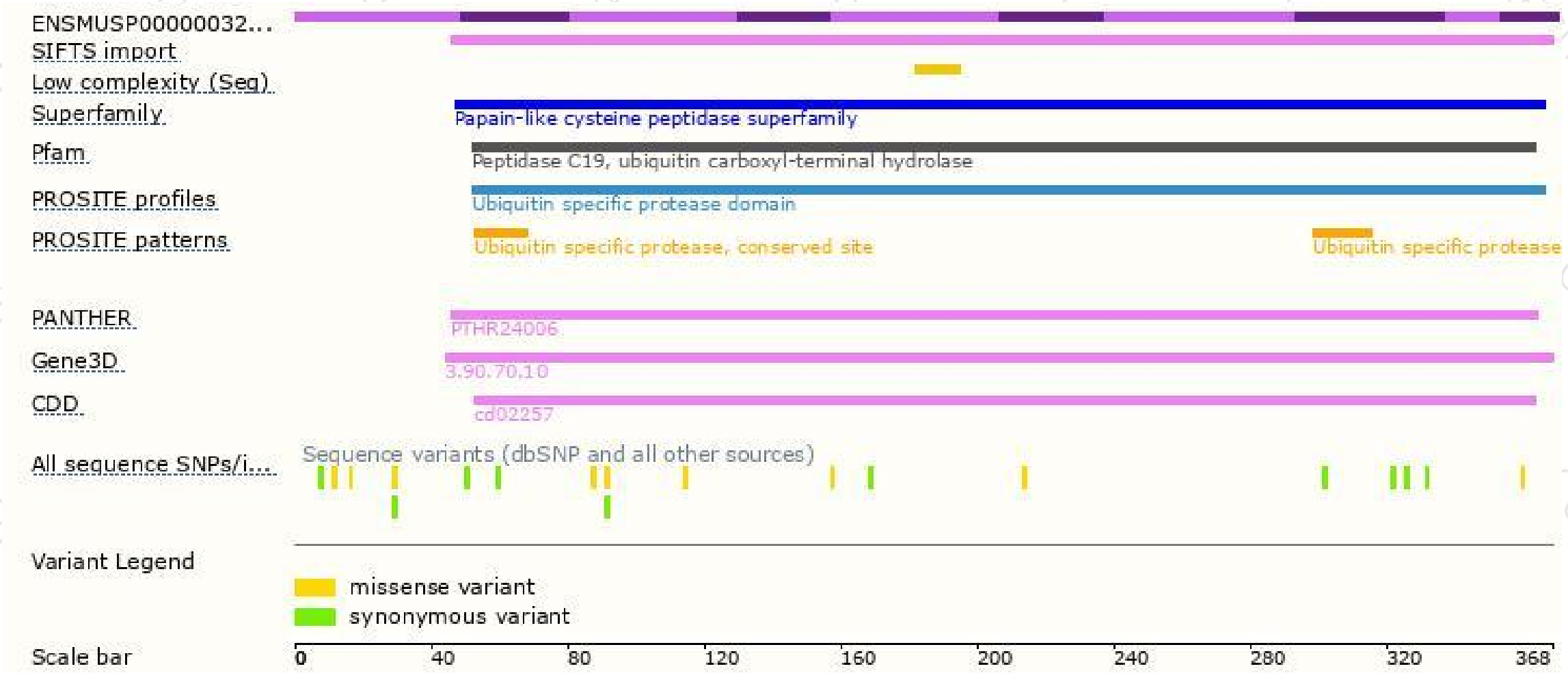
The strategy is based on the design of *Usp18-201* transcript,The transcription is shown below



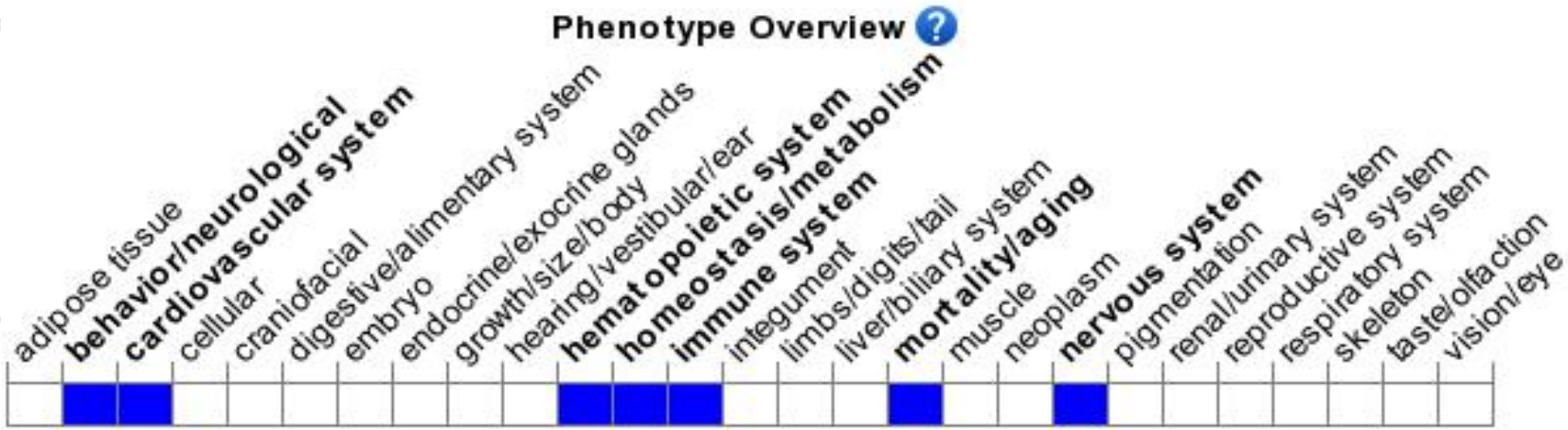
Genomic location distribution



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 null mutants die prematurely with cellular necrosis in the ependyma, breakdown of blood-brain barrier, hydrocephaly with enlarged ventricles, and severe neurological abnormalities. Mice homozygous for an ENU-induced allele exhibit increased susceptibility to Salmonella infection and LPS.

集萃药康
GemPharmatech

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

Tel: 025-5864 1534

集萃药康生物科技
GemPharmatech Co.,Ltd