

Hspa1a Cas9-CKO Strategy

Designer: JiaYu

Reviewer: Xiaojing Li

Design Date: 2021-9-24

Project Overview

Project Name

Hspa1a

Project type

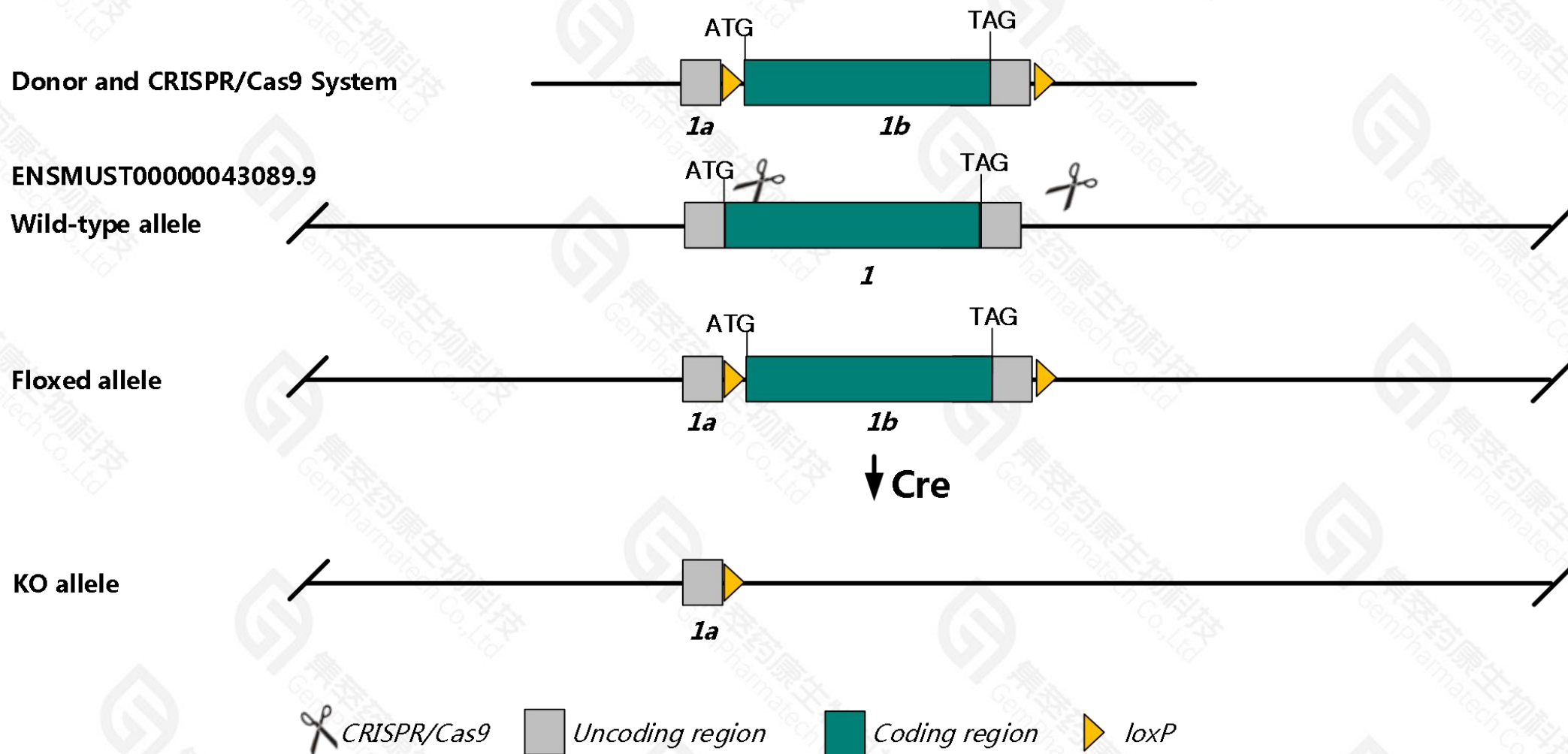
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hspa1a* gene. The schematic diagram is as follows:



Technical routes

- The *Hspala* gene has 1 transcript. According to the structure of *Hspala* gene, exon1 of *Hspala*-201(ENSMUST00000087328.4) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Hspala* 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, at the cellular level, mice homozygous for a knock-out allele exhibit impaired thermotolerance and increased sensitivity to heat stress-induced apoptosis.
- The flox region overlap with part of the *Gm20481* gene, which may delete this gene.
- The flox region is about 800bp away from the 5th end of the *Hspa11* gene, which may affect the regulation of this gene.
- The *Hspa1a* gene is located on the Chr17. 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)

Hspa1a heat shock protein 1A [Mus musculus (house mouse)]

Gene ID: 193740, updated on 12-Jan-2021

Summary



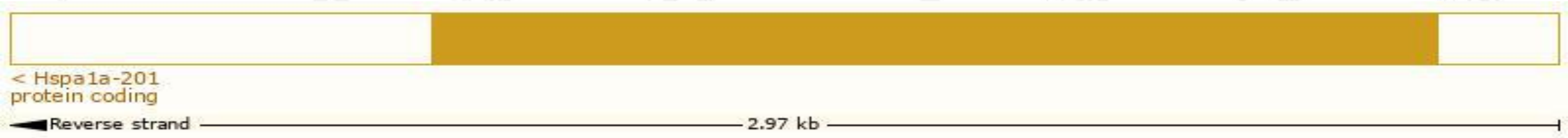
Official Symbol	Hspa1a provided by MGI
Official Full Name	heat shock protein 1A provided by MGI
Primary source	MGI:MGI:96244
See related	Ensembl:ENSMUSG00000091971
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Hsp, Hsp7, Hsp70, Hsp70-3, Hsp70.3, Hsp72, hsp68, hsp70A1
Orthologs	human all

Transcript information (Ensembl)

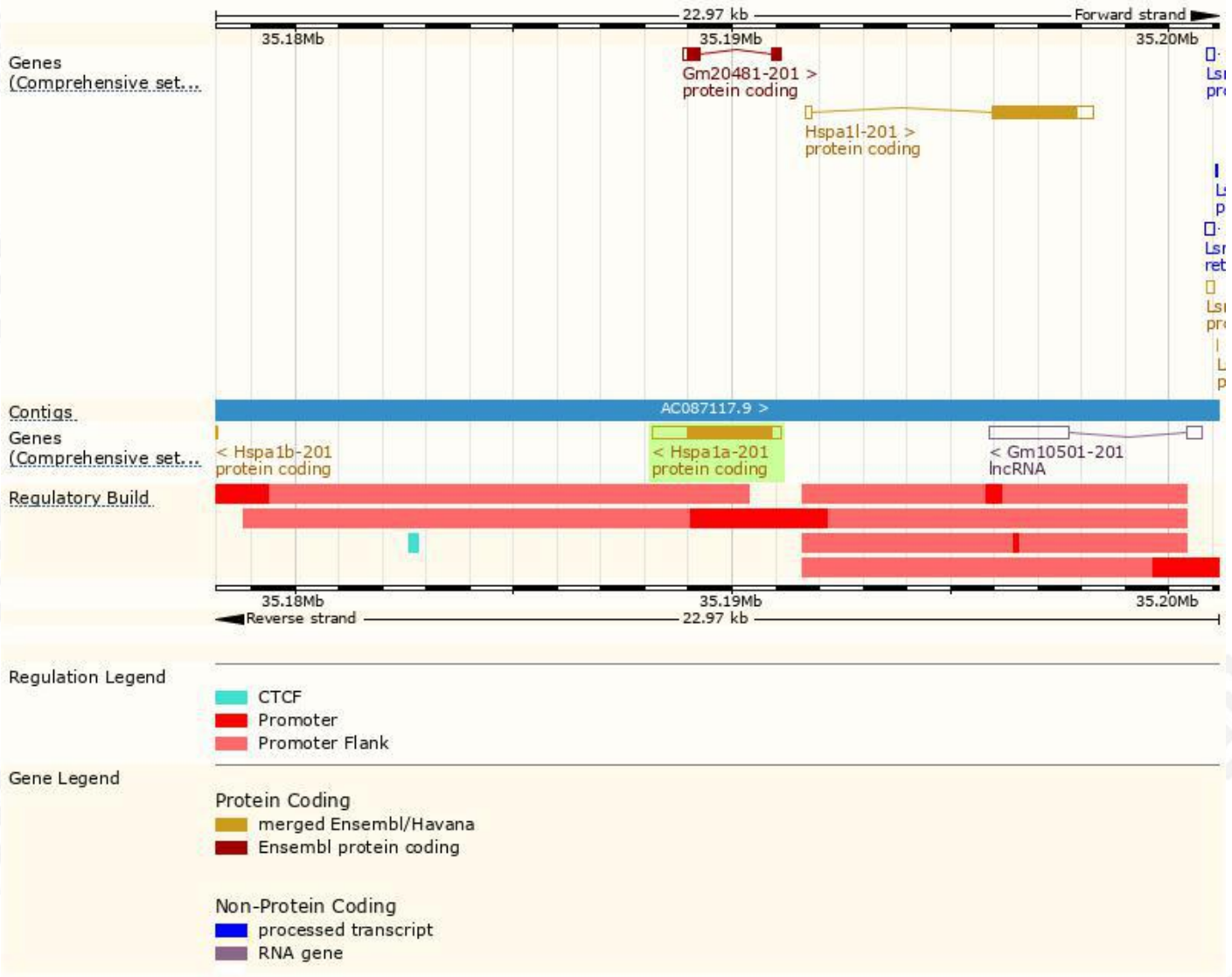
The gene has 1 transcript, and the transcript is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hspa1a-201	ENSMUST00000087328.4	2967	641aa	Protein coding	CCDS50080		TSL:NA , GENCODE basic , APPRIS P1 ,

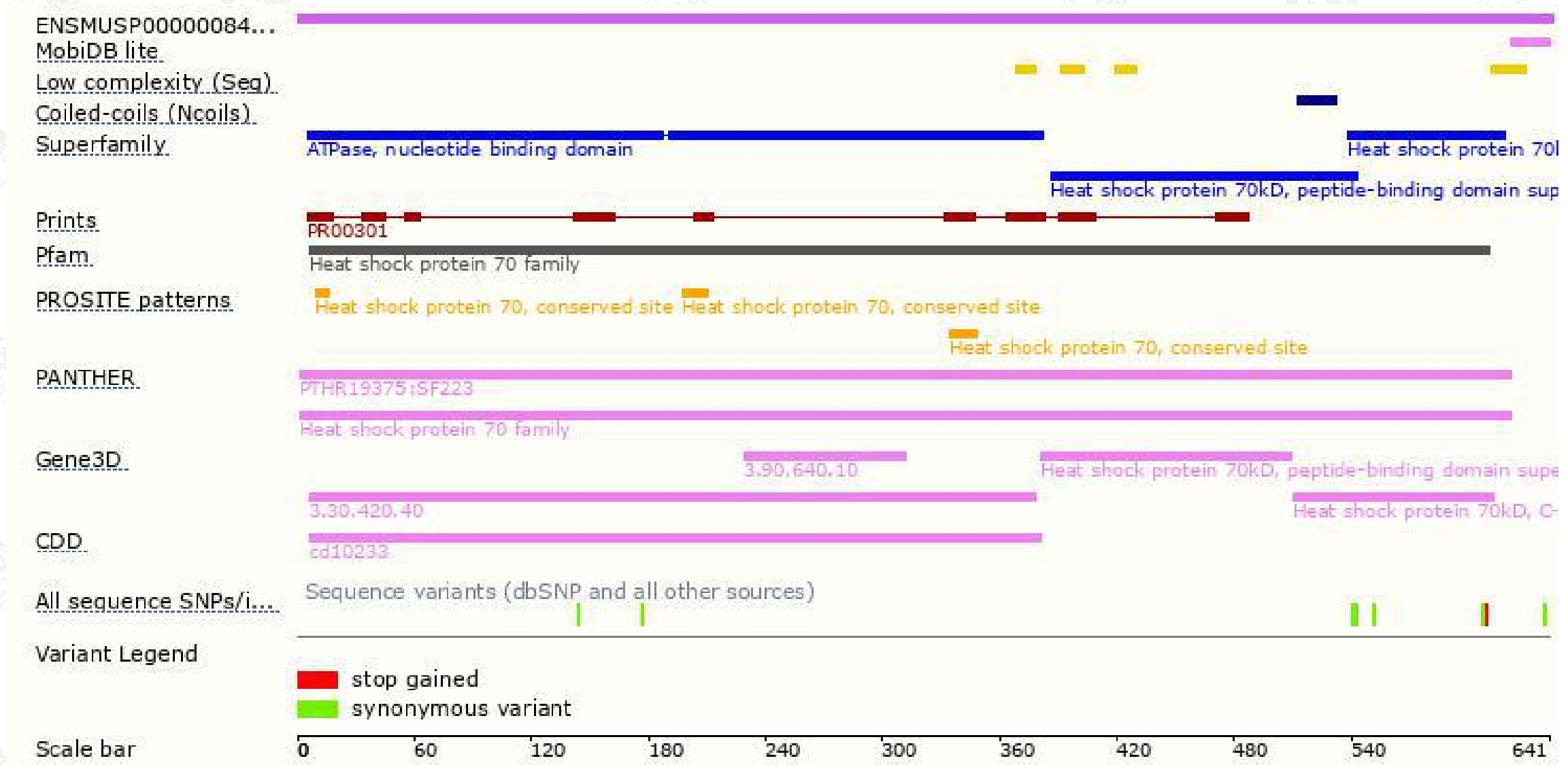
The strategy is based on the design of *Hspa1a-201* transcript, the transcription is shown below:



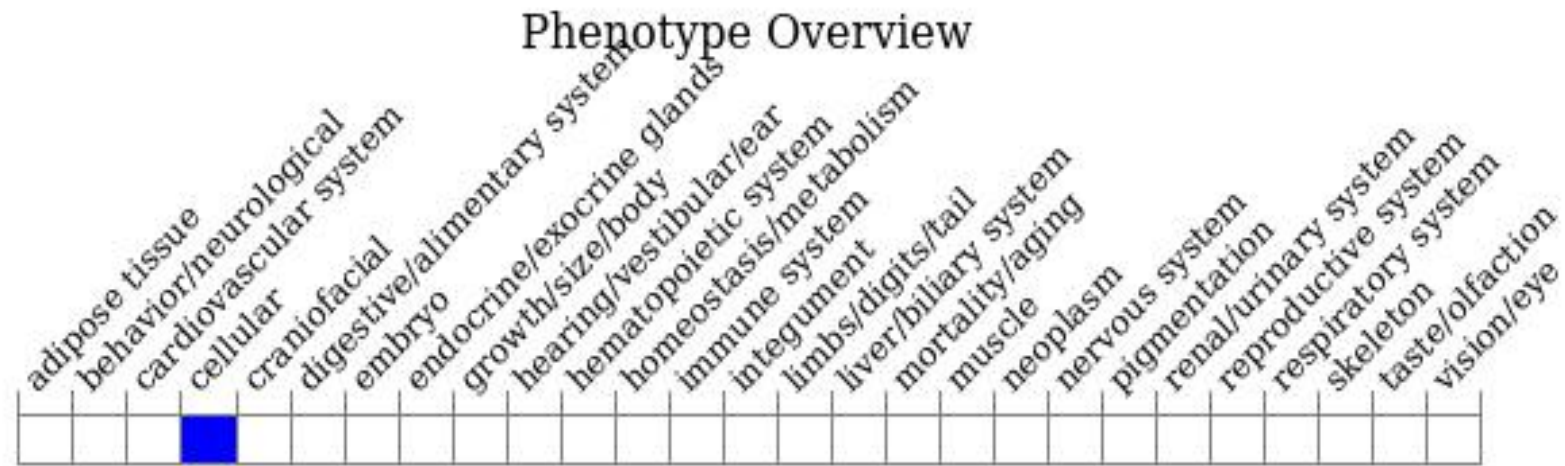
Genomic location distribution



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, at the cellular level, mice homozygous for a knock-out allele exhibit impaired thermotolerance and increased sensitivity to heat stress-induced apoptosis.

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

