

Hspa9 Cas9-CKO Strategy

Designer: Yupeng Yang

Reviewer: Wenjing Li

Design Date: 2018/10/16

Project Overview

Project Name

Hspa9

Project type

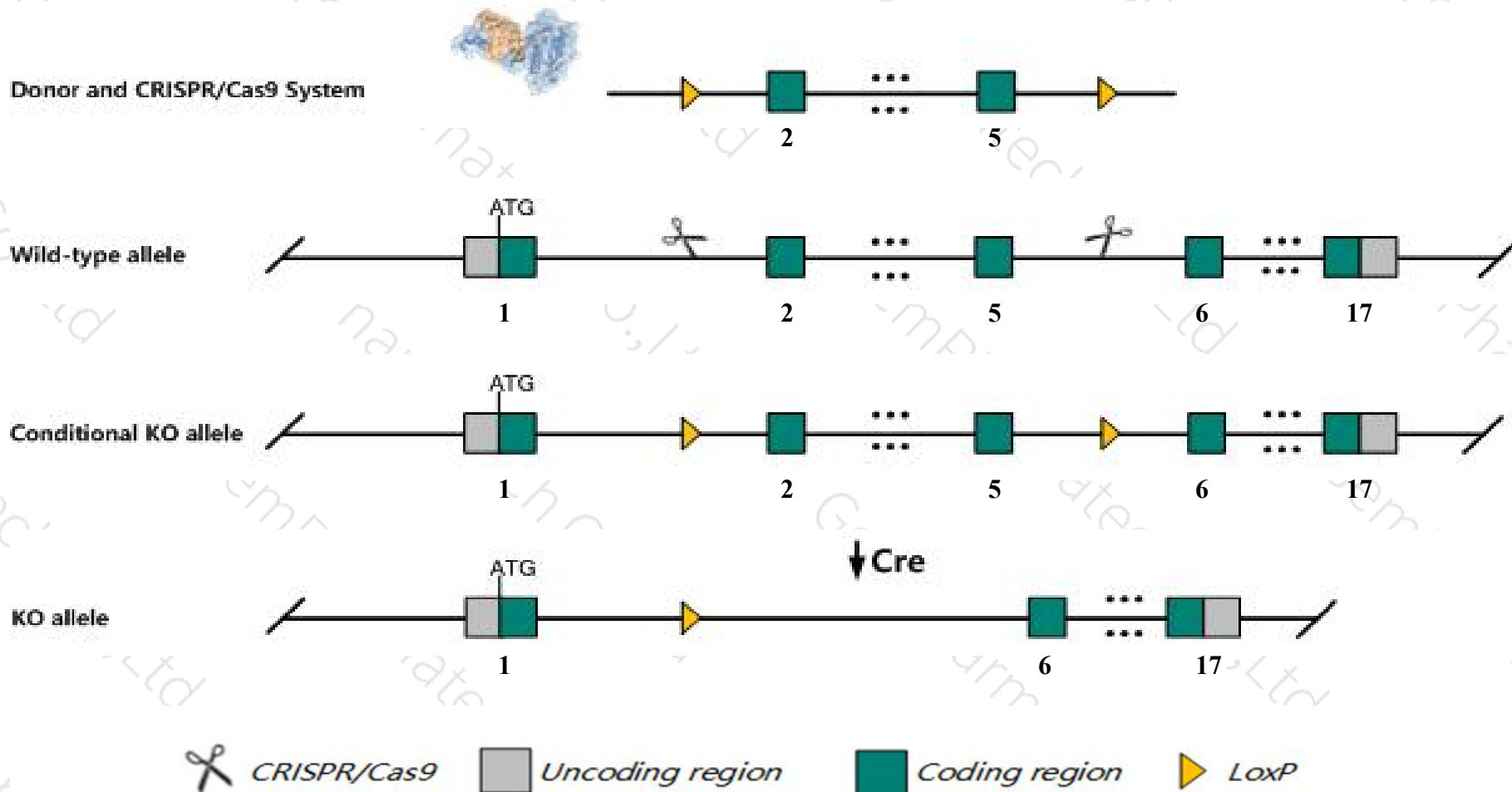
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hspa9* gene. The schematic diagram is as follows:



Technical routes

- The *Hspa9* gene has 5 transcripts. According to the structure of *Hspa9* gene, exon2-exon5 of *Hspa9*-201(ENSMUST00000025217.10) transcript is recommended as the knockout region. The region contains 454bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Hspa9* 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, mice homozygous for a knock-out allele exhibit complete embryonic lethality while heterozygotes display decreased pre-B cell number.
- The *Hspa9* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Hspa9 heat shock protein 9 [Mus musculus (house mouse)]

Gene ID: 15526, updated on 13-Mar-2020

Summary



Official Symbol Hspa9 provided by [MGI](#)

Official Full Name heat shock protein 9 provided by [MGI](#)

Primary source [MGI:MGI:96245](#)

See related [Ensembl:ENSMUSG00000024359](#)

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 74kDa, Csa, Grp75, Hsc74, Hsp74, Hsp74a, Hspa9a, Mortalin, Mot-2, Mot2, Mthsp70, Pbp74

Expression Ubiquitous expression in liver E18 (RPKM 85.2), placenta adult (RPKM 77.5) and 28 other tissues [See more](#)

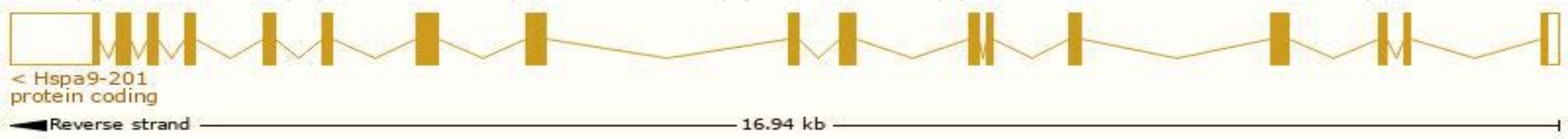
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

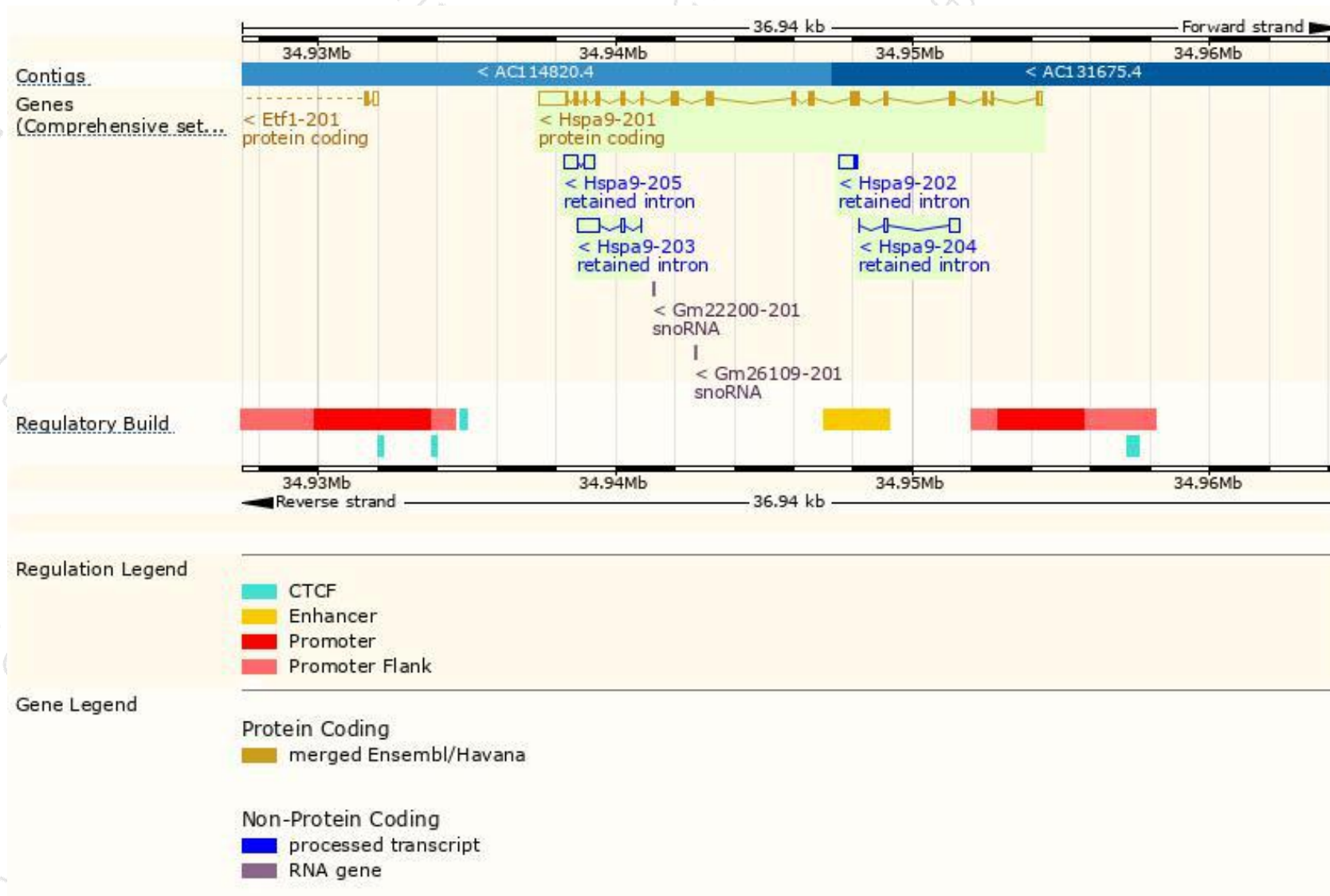
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hspa9-201	ENSMUST00000025217.10	3049	679aa	Protein coding	CCDS29138	P38647	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Hspa9-203	ENSMUST000000236908.1	883	No protein	Retained intron	-	-	
Hspa9-205	ENSMUST000000237399.1	836	No protein	Retained intron	-	-	
Hspa9-202	ENSMUST000000235244.1	504	No protein	Retained intron	-	-	
Hspa9-204	ENSMUST000000237289.1	483	No protein	Retained intron	-	-	

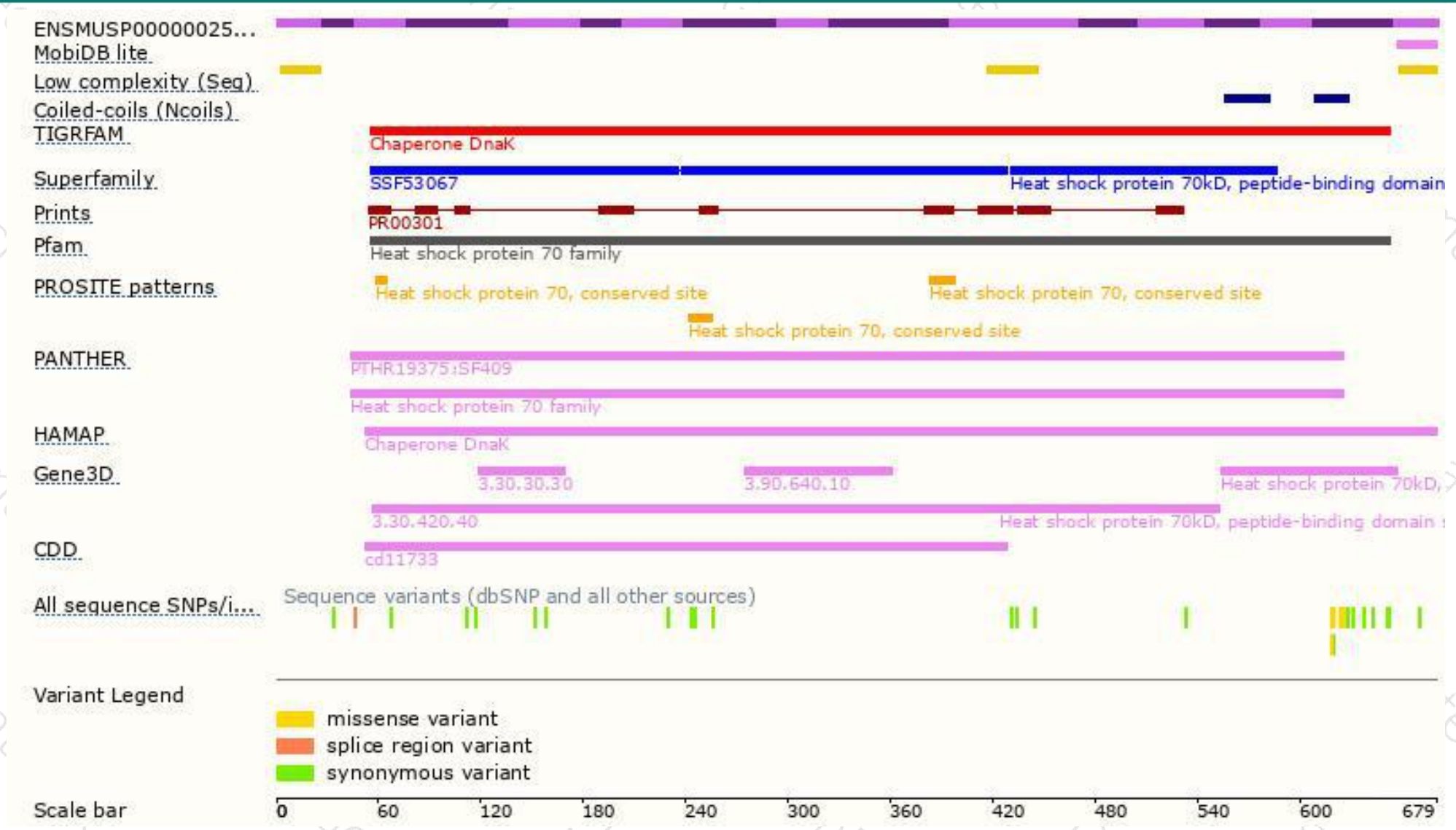
The strategy is based on the design of *Hspa9-201* transcript,the transcription is shown below:



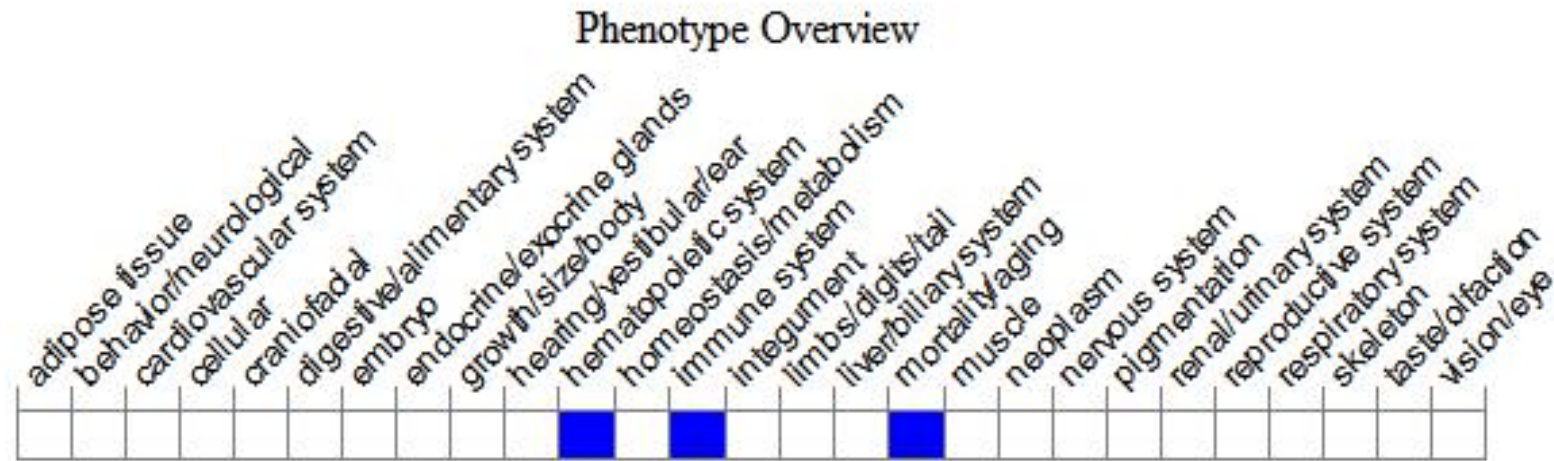
Genomic location distribution



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 complete embryonic lethality while heterozygotes display decreased pre-B cell number.

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

Tel: 400-9660890

