

# ***Tbk1* Cas9-KO Strategy**

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# Project Overview

**Project Name**

***Tbk1***

**Project type**

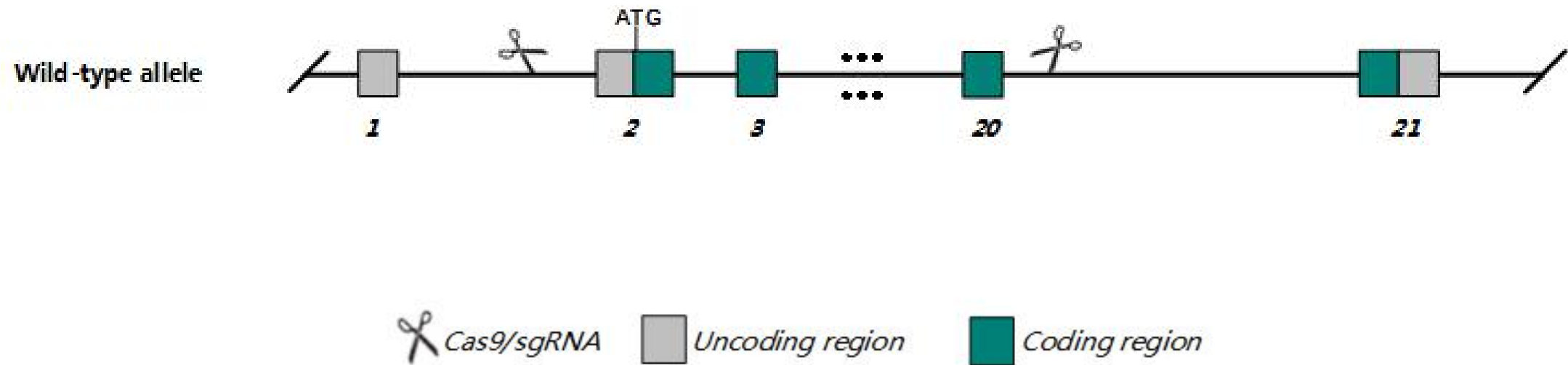
**Cas9-KO**

**Strain background**

**C57BL/6J**

# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Tbk1* gene. The schematic diagram is as follows:



- The *Tbk1* gene has 7 transcripts. According to the structure of *Tbk1* gene, exon2-exon20 of *Tbk1-201* (ENSMUST00000020316.3) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Tbk1* gene. The brief process is as follows: sgRNA was transcribed in vitro. Cas9 and sgRNA 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.

- According to the existing MGI data, Homozygous null mice display embryonic lethality. Mice homozygous for a different knock-out allele exhibit increased prenatal lethality, increased infiltration of mononuclear and/or granulomatous cells in multiple organs and tissues at 3 months of age, and increased lethality in response to LPS.
- The *Tbkl* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.



# Gene information (NCBI)

## Tbk1 TANK-binding kinase 1 [Mus musculus (house mouse)]

Gene ID: 56480, updated on 31-Jan-2019

### Summary



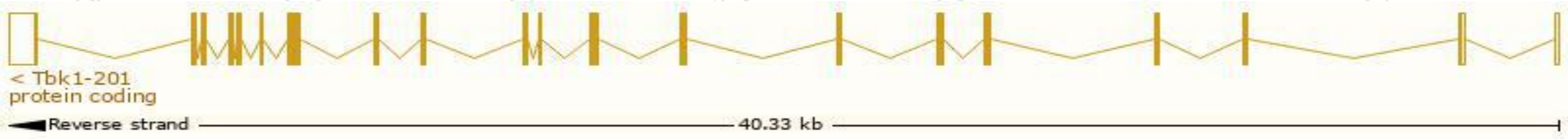
|                           |   |
|---------------------------|---|
| <b>Official Symbol</b>    | Tbk1 provided by <a href="#">MGI</a>  |
| <b>Official Full Name</b> | TANK-binding kinase 1 provided by <a href="#">MGI</a>   |
| <b>Primary source</b>     | <a href="#">MGI:MGI:1929658</a>   |
| <b>See related</b>        | <a href="#">Ensembl:ENSMUSG00000020115</a>  |
| <b>Gene type</b>          | protein coding  |
| <b>RefSeq status</b>      | VALIDATED   |
| <b>Organism</b>           | <a href="#">Mus musculus</a>  |
| <b>Lineage</b>            | Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus |
| <b>Also known as</b>      | 1200008B05Rik, AI462036, AW048562   |
| <b>Expression</b>         | Ubiquitous expression in bladder adult (RPKM 7.5), CNS E11.5 (RPKM 7.4) and 28 other tissues <a href="#">See more</a>   |
| <b>Orthologs</b>          | <a href="#">human</a> <a href="#">all</a>   |

# Transcript information（Ensembl）

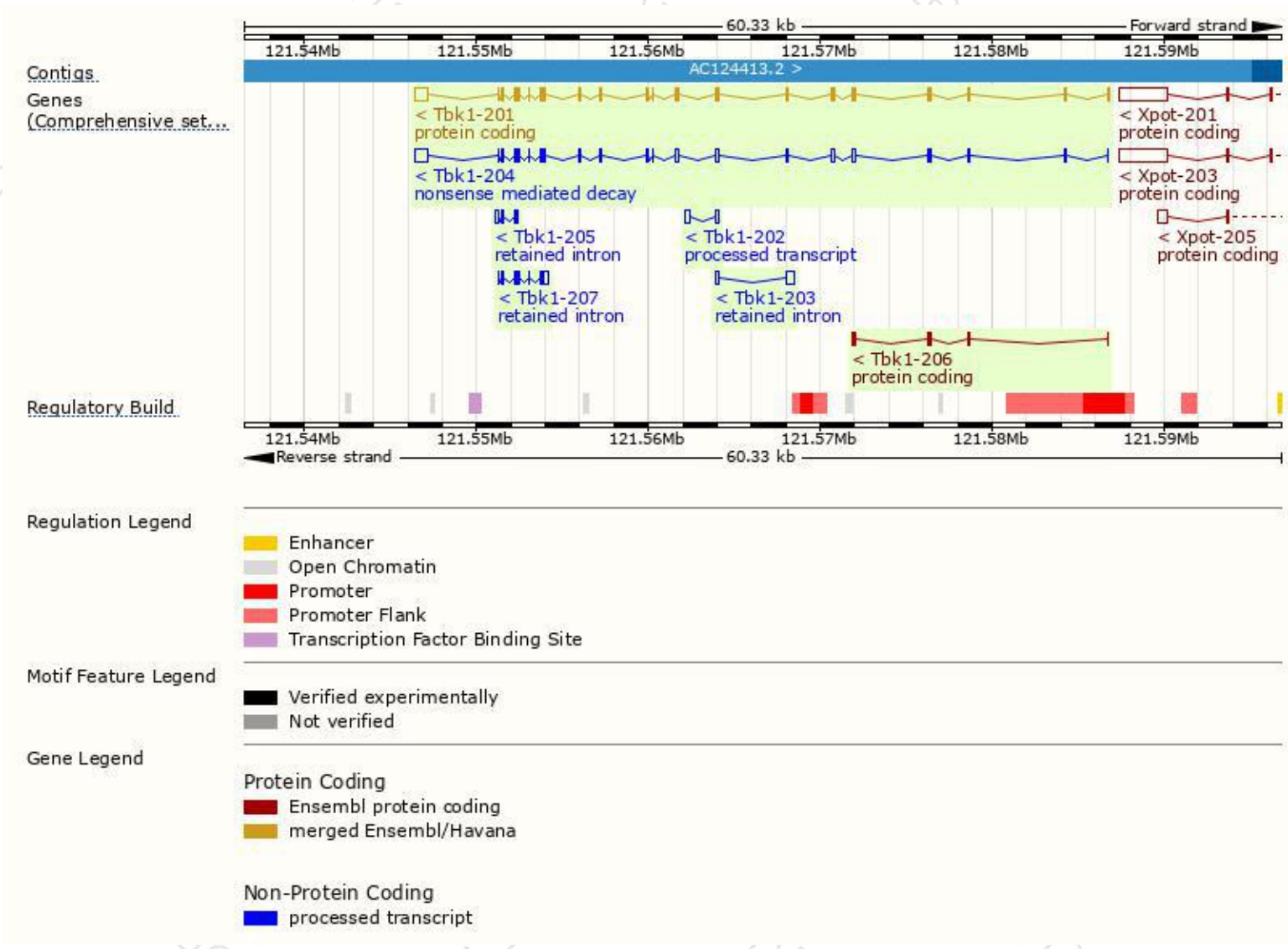
The gene has 7 transcripts,all transcripts are shown below:

| Name     | Transcript ID                        | bp   | Protein               | Biotype                 | CCDS                      | UniProt                       | Flags                         |
|----------|--------------------------------------|------|-----------------------|-------------------------|---------------------------|-------------------------------|-------------------------------|
| Tbk1-201 | <a href="#">ENSMUST00000020316.3</a> | 3024 | <a href="#">729aa</a> | Protein coding          | <a href="#">CCDS24212</a> | <a href="#">A1L361 Q9WUN2</a> | TSL:1 GENCODE basic APPRIS P1 |
| Tbk1-206 | <a href="#">ENSMUST00000219493.1</a> | 442  | <a href="#">108aa</a> | Protein coding          | -                         | <a href="#">A0A1W2P835</a>    | CDS 3' incomplete TSL:3       |
| Tbk1-204 | <a href="#">ENSMUST00000219400.1</a> | 3005 | <a href="#">121aa</a> | Nonsense mediated decay | -                         | <a href="#">A0A1W2P6W7</a>    | TSL:2                         |
| Tbk1-202 | <a href="#">ENSMUST00000218460.1</a> | 406  | No protein            | Processed transcript    | -                         | -                             | TSL:3                         |
| Tbk1-207 | <a href="#">ENSMUST00000220386.1</a> | 747  | No protein            | Retained intron         | -                         | -                             | TSL:3                         |
| Tbk1-203 | <a href="#">ENSMUST00000219244.1</a> | 634  | No protein            | Retained intron         | -                         | -                             | TSL:3                         |
| Tbk1-205 | <a href="#">ENSMUST00000219439.1</a> | 485  | No protein            | Retained intron         | -                         | -                             | TSL:3                         |

The strategy is based on the design of *Tbk1-201* transcript,The transcription is shown below

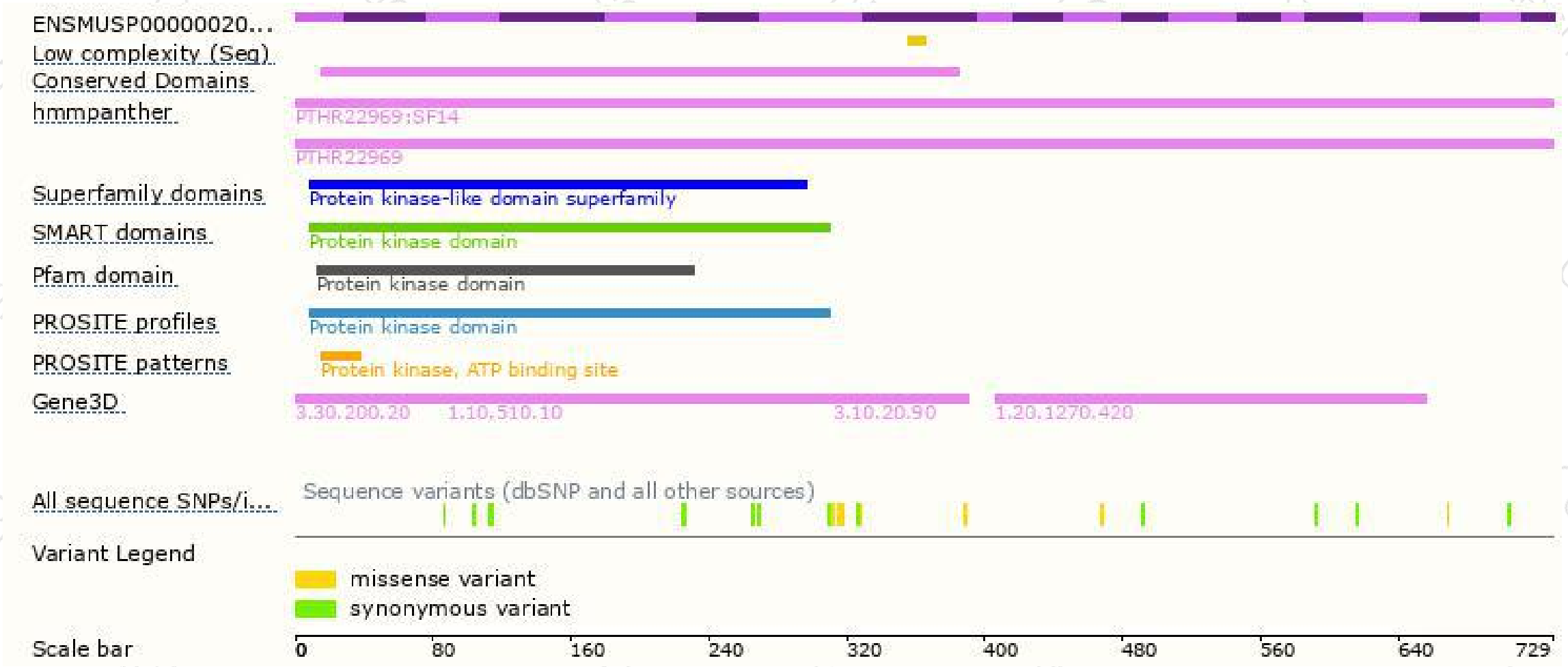


# Genomic location distribution

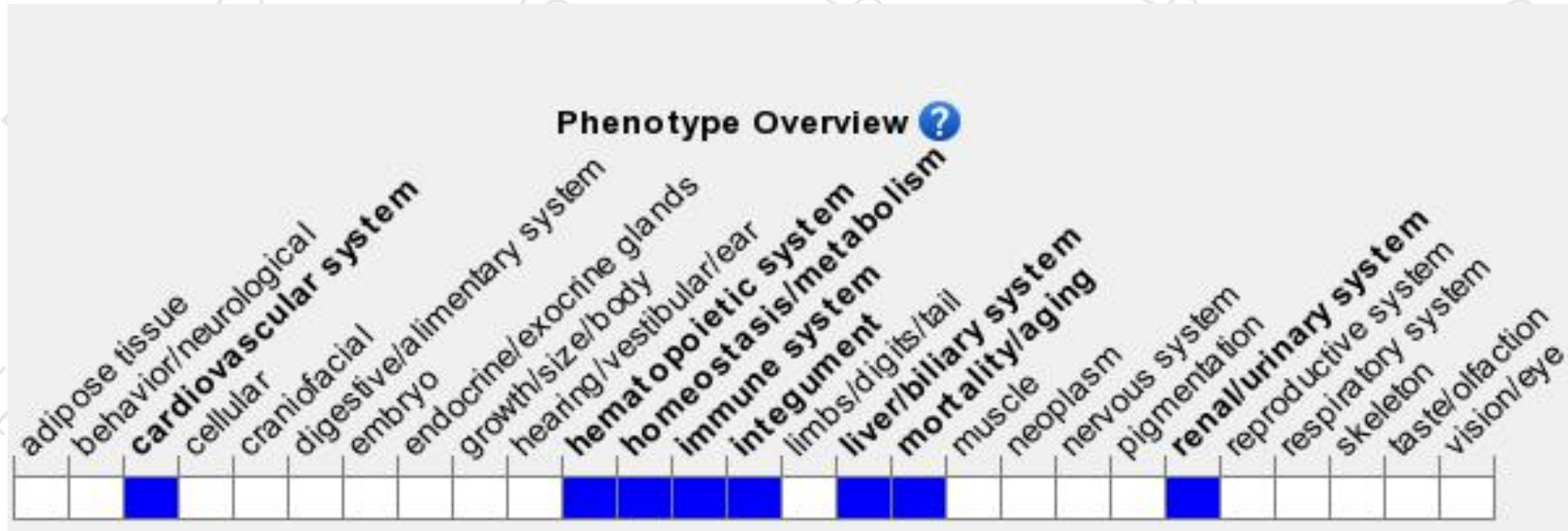




# Protein domain



# 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 mice display embryonic lethality. Mice homozygous for a different knock-out allele exhibit increased prenatal lethality, increased infiltration of mononuclear and/or granulomatous cells in multiple organs and tissues at 3 months of age, and increased lethality in response to LPS.

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

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