

# *Itsn2* Cas9-KO Strategy

**Designer:**

**Huimin Su**

**Reviewer:**

**Ruirui Zhang**

**Design Date:**

**2020-2-26**

# Project Overview

---

**Project Name**

***Itsn2***

---

**Project type**

**Cas9-KO**

---

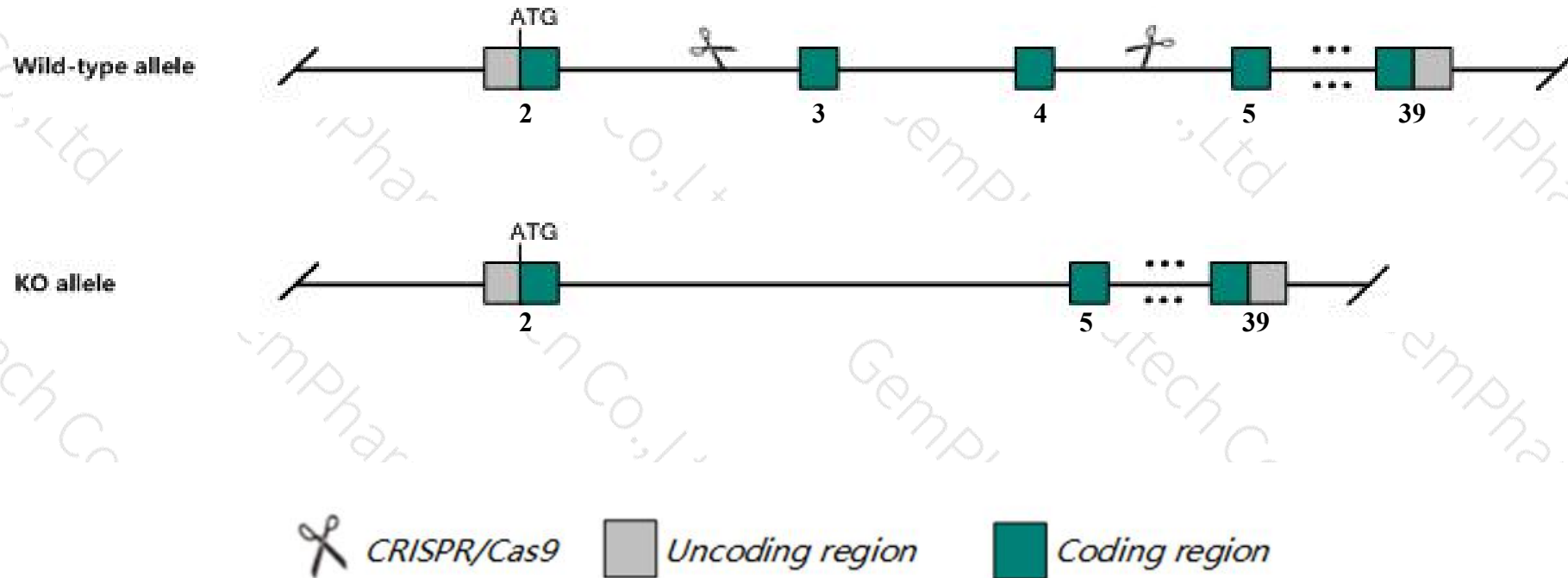
**Strain background**

**C57BL/6JGpt**

---

# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Itsn2* gene. The schematic diagram is as follows:



# Technical routes

- The *Itsn2* gene has 15 transcripts. According to the structure of *Itsn2* gene, exon3-exon4 of *Itsn2-201* (ENSMUST00000062580.7) transcript is recommended as the knockout region. The region contains 157bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Itsn2* gene. The brief process is as follows: CRISPR/Cas9 system were

- According to the existing MGI data, mice homozygous for a knock-out allele exhibit normal brain morphology and function and behavior. Mice lacking the long isoform exhibit delayed recovery from LPS-induced kidney injury.
- The knockout region contains part intron1-2 of *Gm48678-201* gene.
- The transcript *Itsn2-202* is incomplete, so the effect on it is unknown.
- The *Itsn2* gene is located on the Chr12. 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)



## Itsn2 intersectin 2 [ *Mus musculus* (house mouse) ]

Gene ID: 20403, updated on 24-Oct-2019

### Summary

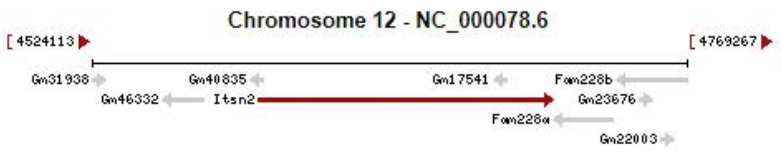
- Official Symbol [Itsn2](#) provided by [MGI](#)
- Official Full Name [intersectin 2](#) provided by [MGI](#)
- Primary source [MGI:MGI:1338049](#)
- See related [Ensembl:ENSMUSG00000020640](#)
- Gene type protein coding
- RefSeq status VALIDATED
- Organism [Mus musculus](#)
- Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
- Also known as [Ese2](#); [Sh3d1B](#); [Sh3p18](#); [AI327390](#); [mKIAA1256](#)
- Expression Ubiquitous expression in thymus adult (RPKM 7.2), spleen adult (RPKM 6.5) and 28 other tissues [See more](#)
- Orthologs [human](#) [all](#)

### Genomic context

Location: 12; 12 A1.1 [See Itsn2 in Genome Data Viewer](#)

Exon count: 42

| Annotation release  | Status            | Assembly                                       | Chr | Location                       |
|---------------------|-------------------|--|-----|--------------------------------|
| <a href="#">108</a> | current           | GRCm38.p6 ( <a href="#">GCF_000001635.26</a> ) | 12  | NC_000078.6 (4592643..4713952) |
| Build 37.2          | previous assembly | MGSCv37 ( <a href="#">GCF_000001635.18</a> )   | 12  | NC_000078.5 (4599814..4720758) |

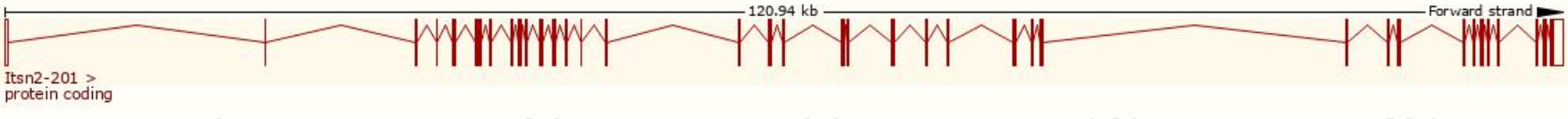


# Transcript information (Ensembl)

The gene has 15 transcripts,all transcripts are shown below:

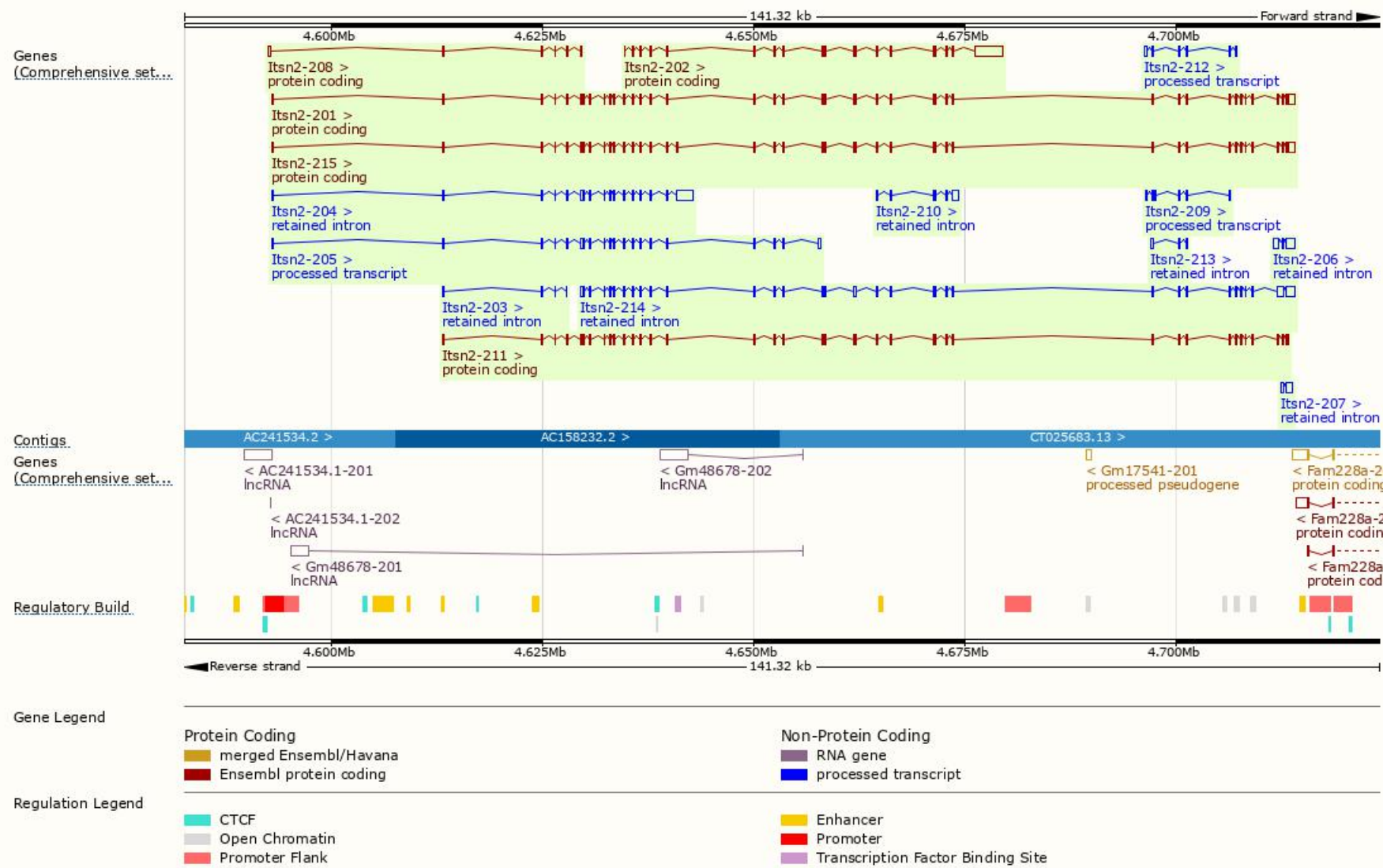
| Name      | Transcript ID                         | bp   | Protein                | Biotype              | CCDS                      | UniProt                    | Flags                           |
|-----------|---------------------------------------|------|------------------------|----------------------|---------------------------|----------------------------|---------------------------------|
| Itsn2-201 | <a href="#">ENSMUST00000062580.7</a>  | 5987 | <a href="#">1658aa</a> | Protein coding       | <a href="#">CCDS36398</a> | <a href="#">E9QNG1</a>     | TSL:5 GENCODE basic APPRIS P2   |
| Itsn2-211 | <a href="#">ENSMUST00000219007.1</a>  | 4977 | <a href="#">1658aa</a> | Protein coding       | <a href="#">CCDS36398</a> | <a href="#">E9QNG1</a>     | TSL:1 GENCODE basic APPRIS P2   |
| Itsn2-215 | <a href="#">ENSMUST000002220311.1</a> | 6080 | <a href="#">1685aa</a> | Protein coding       | -                         | <a href="#">B2RR82</a>     | TSL:1 GENCODE basic APPRIS ALT2 |
| Itsn2-202 | <a href="#">ENSMUST00000217672.1</a>  | 5431 | <a href="#">712aa</a>  | Protein coding       | -                         | <a href="#">A0A1W2P775</a> | CDS 5' incomplete TSL:1         |
| Itsn2-208 | <a href="#">ENSMUST00000218402.1</a>  | 851  | <a href="#">175aa</a>  | Protein coding       | -                         | <a href="#">A0A1W2P7G8</a> | CDS 3' incomplete TSL:2         |
| Itsn2-205 | <a href="#">ENSMUST00000218072.1</a>  | 2640 | No protein             | Processed transcript | -                         | -                          | TSL:1                           |
| Itsn2-212 | <a href="#">ENSMUST00000219182.1</a>  | 832  | No protein             | Processed transcript | -                         | -                          | TSL:3                           |
| Itsn2-209 | <a href="#">ENSMUST00000218923.1</a>  | 751  | No protein             | Processed transcript | -                         | -                          | TSL:3                           |
| Itsn2-214 | <a href="#">ENSMUST00000219832.1</a>  | 5804 | No protein             | Retained intron      | -                         | -                          | TSL:1                           |
| Itsn2-204 | <a href="#">ENSMUST00000217981.1</a>  | 3968 | No protein             | Retained intron      | -                         | -                          | TSL:1                           |
| Itsn2-206 | <a href="#">ENSMUST00000218084.1</a>  | 1606 | No protein             | Retained intron      | -                         | -                          | TSL:1                           |
| Itsn2-210 | <a href="#">ENSMUST00000218985.1</a>  | 1272 | No protein             | Retained intron      | -                         | -                          | TSL:2                           |
| Itsn2-207 | <a href="#">ENSMUST00000218211.1</a>  | 942  | No protein             | Retained intron      | -                         | -                          | TSL:1                           |
| Itsn2-213 | <a href="#">ENSMUST00000219308.1</a>  | 616  | No protein             | Retained intron      | -                         | -                          | TSL:3                           |
| Itsn2-203 | <a href="#">ENSMUST00000217942.1</a>  | 319  | No protein             | Retained intron      | -                         | -                          | TSL:3                           |

The strategy is based on the design of *Itsn2-201* transcript, the transcription is shown below:



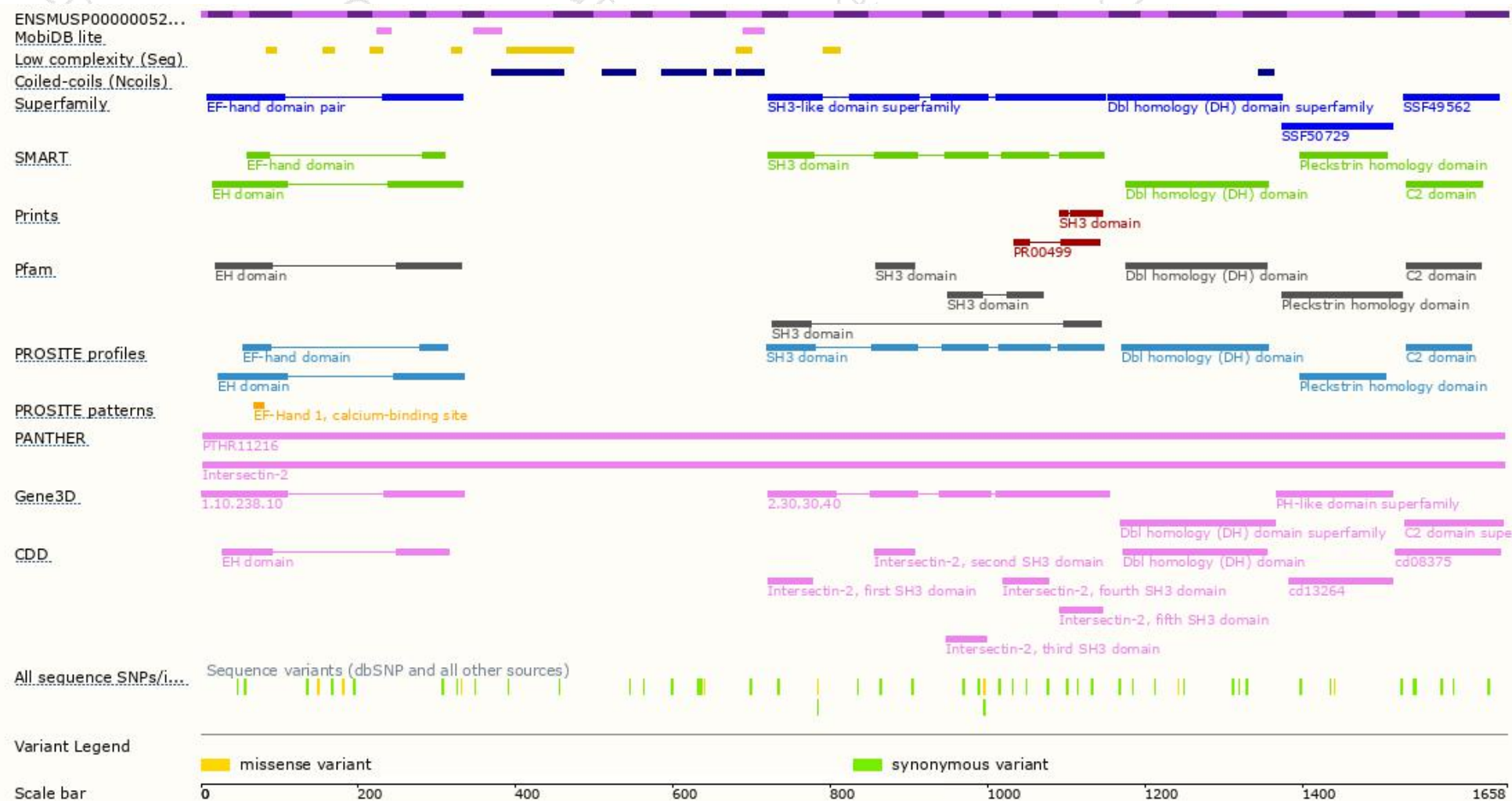


# Genomic location distribution

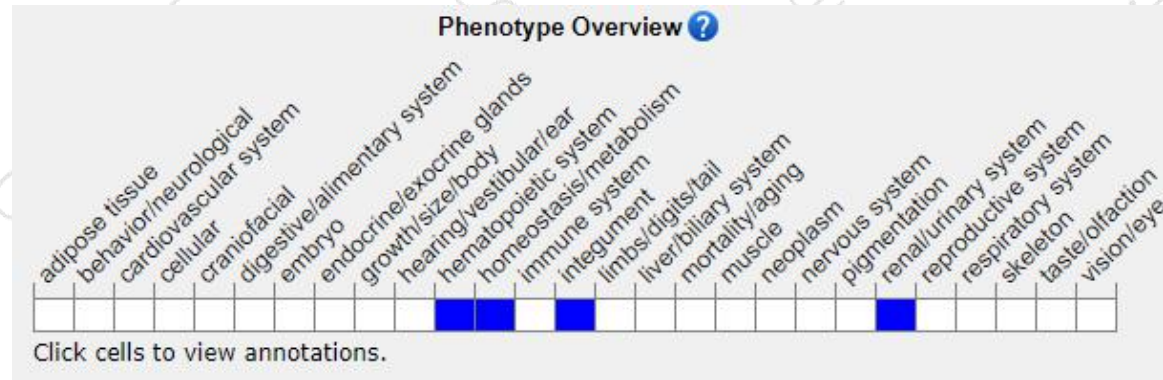




# Protein domain



# 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 normal brain morphology and function and behavior. Mice lacking the long isoform exhibit delayed recovery from LPS-induced kidney injury.

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

Tel: 400-9660890

