

Tbp Cas9-CKO Strategy

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Reviewer:

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Design Date:

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Project Overview

Project Name

Tbp

Project type

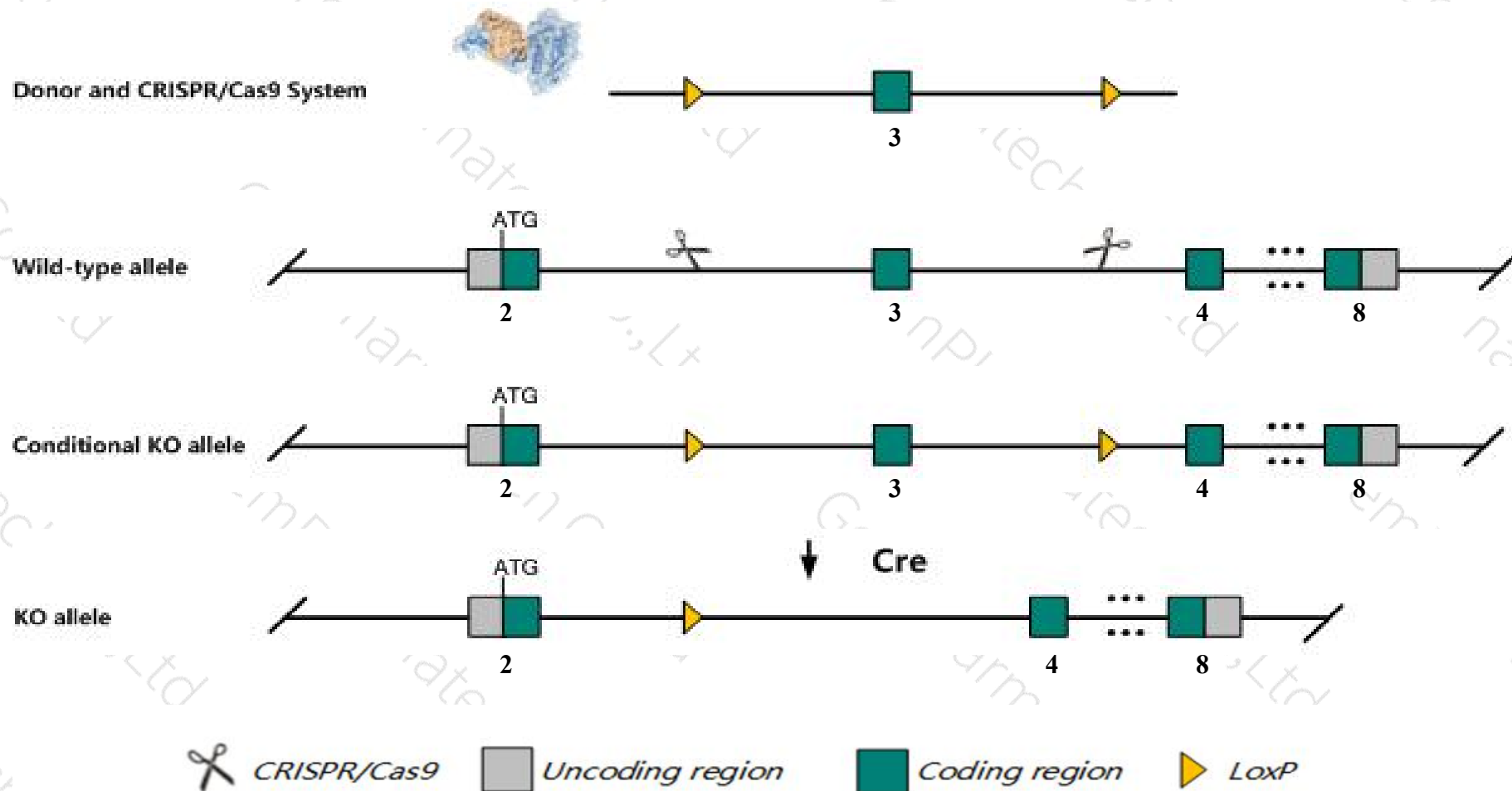
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Tbp* gene has 12 transcripts. According to the structure of *Tbp* gene, exon3 of *Tbp-211* (ENSMUST00000162505.7) transcript is recommended as the knockout region. The region contains 374bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Tbp* 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 one null allele exhibit embryonic lethality caused by hemorrhaging and clotting in the placenta and surviving embryos die prior to weaning. Mice homozygous for another null allele undergo growth arrest after hatching and apoptosis.
- The *Tbp* 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)

Tbp TATA box binding protein [Mus musculus (house mouse)]

Gene ID: 21374, updated on 31-Jan-2019

Summary



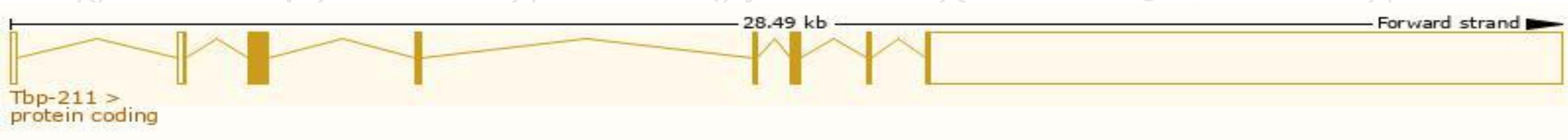
| | |
|---------------------------|---|
| Official Symbol | Tbp provided by MGI |
| Official Full Name | TATA box binding protein provided by MGI |
| Primary source | MGI:MGI:101838 |
| See related | Ensembl:ENSMUSG00000014767 |
| Gene type | protein coding |
| RefSeq status | VALIDATED |
| Organism | Mus musculus |
| Lineage | Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus |
| Also known as | GTF2D1, Gtf2d, SCA17, TFIID |
| Expression | Ubiquitous expression in testis adult (RPKM 40.6), CNS E14 (RPKM 17.0) and 27 other tissues See more |
| Orthologs | human all |

Transcript information (Ensembl)

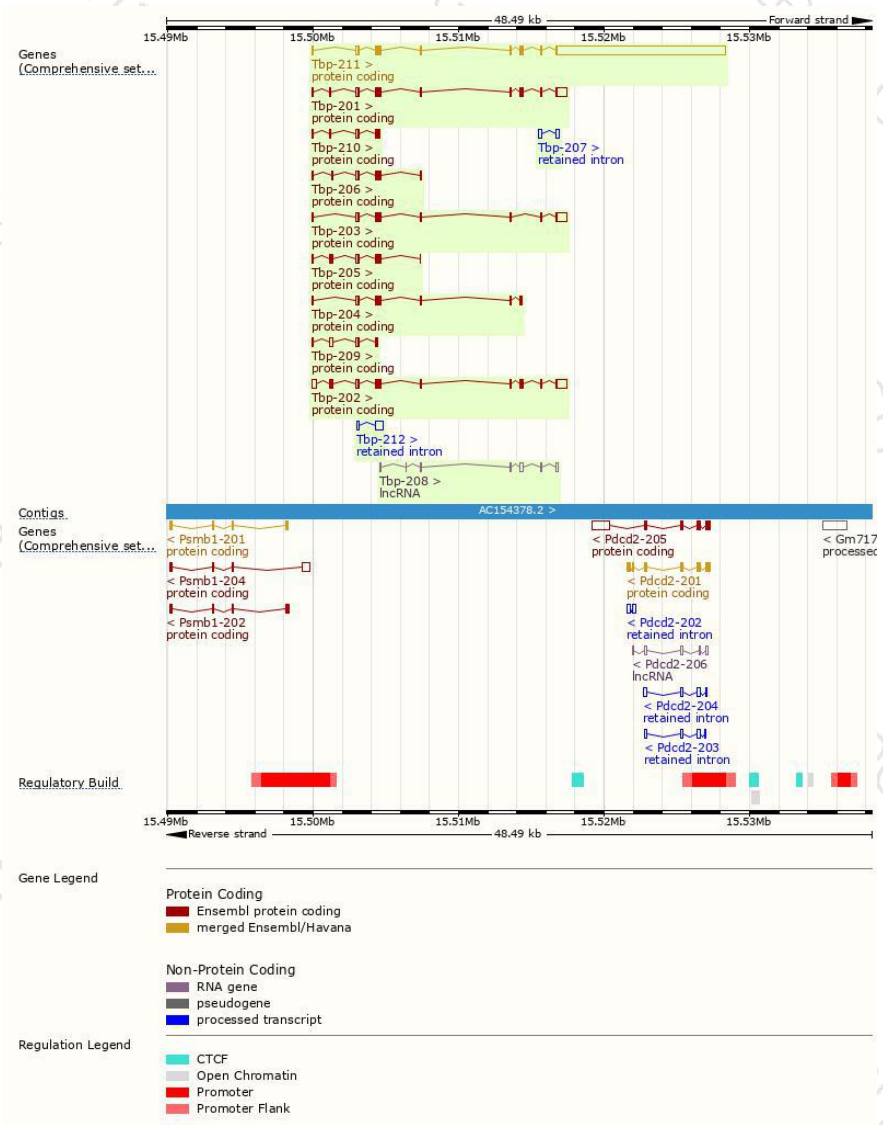
The gene has 12 transcripts,all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---------|---------------------------------------|-------|-----------------------|-----------------|---------------------------|-------------------------------|-------------------------------|
| Tbp-211 | ENSMUST00000162505.7 | 12794 | 316aa | Protein coding | CCDS37454 | P29037 Q6RI65 | TSL:1 GENCODE basic APPRIS P1 |
| Tbp-202 | ENSMUST00000117593.7 | 2067 | 316aa | Protein coding | CCDS37454 | P29037 Q6RI65 | TSL:1 GENCODE basic APPRIS P1 |
| Tbp-201 | ENSMUST00000014911.11 | 1891 | 316aa | Protein coding | CCDS37454 | P29037 Q6RI65 | TSL:1 GENCODE basic APPRIS P1 |
| Tbp-203 | ENSMUST00000118001.7 | 1652 | 260aa | Protein coding | - | Q91YM5 | TSL:1 GENCODE basic |
| Tbp-204 | ENSMUST00000119879.8 | 967 | 250aa | Protein coding | - | F8WJ67 | CDS 3' incomplete TSL:5 |
| Tbp-206 | ENSMUST00000147081.8 | 820 | 180aa | Protein coding | - | E9Q084 | CDS 3' incomplete TSL:3 |
| Tbp-205 | ENSMUST00000143924.7 | 763 | 148aa | Protein coding | - | E9Q9Z7 | CDS 3' incomplete TSL:5 |
| Tbp-210 | ENSMUST00000159197.7 | 685 | 128aa | Protein coding | - | E9Q6D9 | CDS 3' incomplete TSL:3 |
| Tbp-209 | ENSMUST00000155051.7 | 639 | 76aa | Protein coding | - | D3Z453 | CDS 3' incomplete TSL:2 |
| Tbp-212 | ENSMUST00000232661.1 | 691 | No protein | Retained intron | - | - | |
| Tbp-207 | ENSMUST00000149518.1 | 460 | No protein | Retained intron | - | - | TSL:2 |
| Tbp-208 | ENSMUST00000153480.1 | 712 | No protein | lncRNA | - | - | TSL:3 |

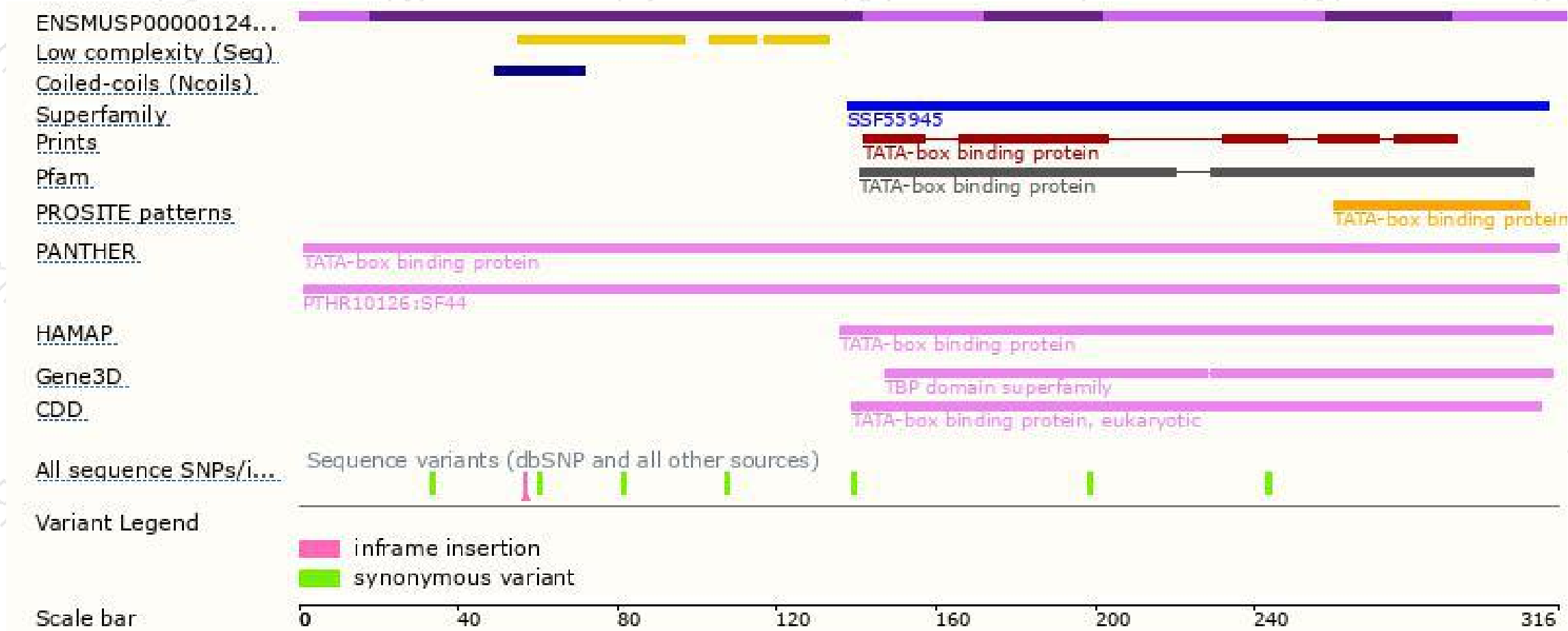
The strategy is based on the design of *Tbp-211* transcript,The transcription is shown below



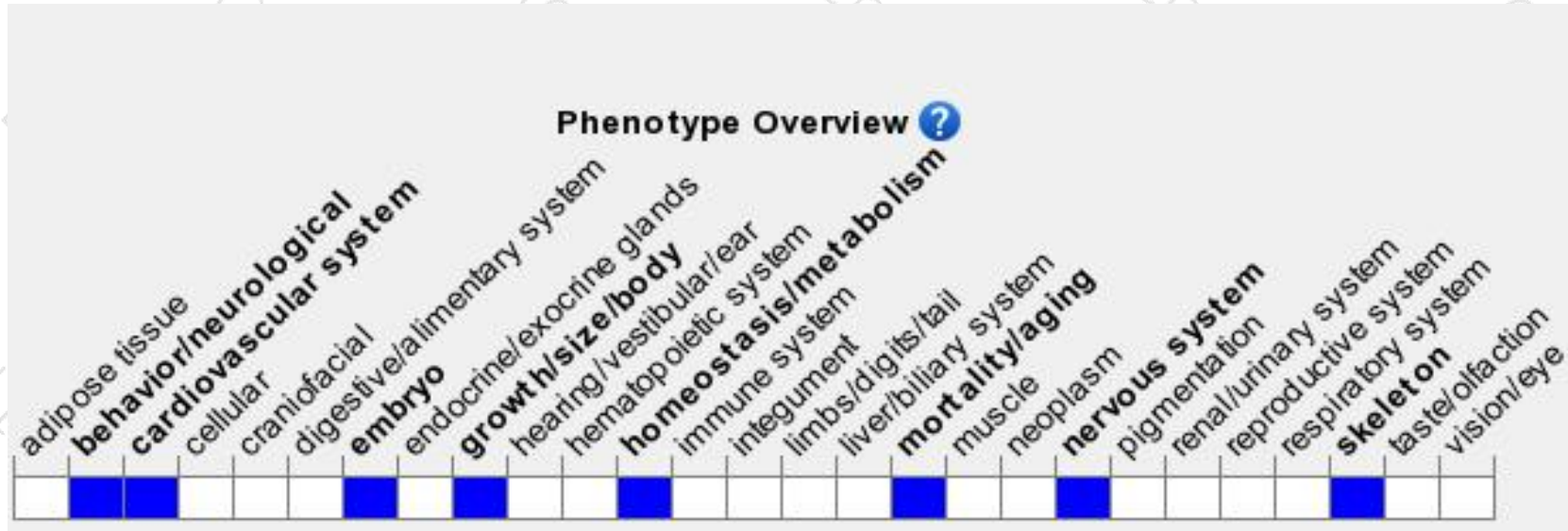
Genomic location distribution



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 one null allele exhibit embryonic lethality caused by hemorrhaging and clotting in the placenta and surviving embryos die prior to weaning. Mice homozygous for another null allele undergo growth arrest after hatching and apoptosis.

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

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