

# ***Tnnc1* Cas9-CKO Strategy**

Designer: Xueting Zhang

Reviewer: Yanhua Shen

Design Date: 2019-9-4

# Project Overview

**Project Name**

***Tnnc1***

**Project type**

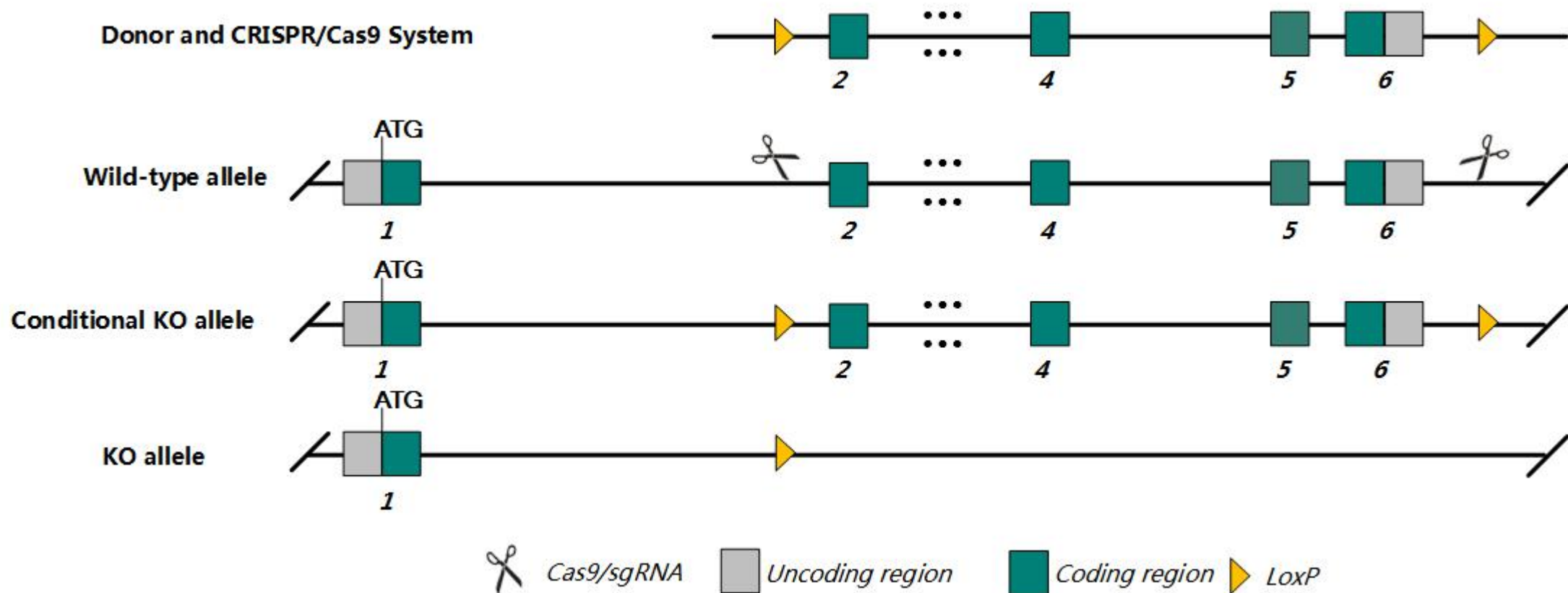
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Tnnc1* gene. The schematic diagram is as follows:



# Technical routes

- The *Tnnc1* gene has 3 transcripts. According to the structure of *Tnnc1* gene, exon2-6 of *Tnnc1*-202 (ENSMUST00000169169.7) transcript is recommended as the knockout region. The region contains most of coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Tnnc1* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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.

- *Tnncl* is located in intron of *Nisch* gene, the partial sequence of intron of *Nisch* gene will be deleted together in this strategy.
- The floxed region is near to the N-terminal of *Nisch* gene and *Sema3g* gene, this strategy may influence the regulatory function of the N-terminal of these genes.
- The *Tnncl* gene is located on the Chr14. 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)

## Tnnc1 troponin C, cardiac/slow skeletal [ *Mus musculus* (house mouse) ]

Gene ID: 21924, updated on 12-Aug-2019

### Summary

Official Symbol	Tnnc1 provided by <a href="#">MGI</a>
Official Full Name	troponin C, cardiac/slow skeletal provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:98779</a>
See related	<a href="#">Ensembl:ENSMUSG00000091898</a>
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	TnC; cTnC; cTnl; tncc; AI874626
Expression	Biased expression in heart adult (RPKM 1462.3) and limb E14.5 (RPKM 100.7) <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

### Genomic context

Location: 14 B; 14 19.09 cM

See Tnnc1 in [Genome Data Viewer](#)

Exon count: 6

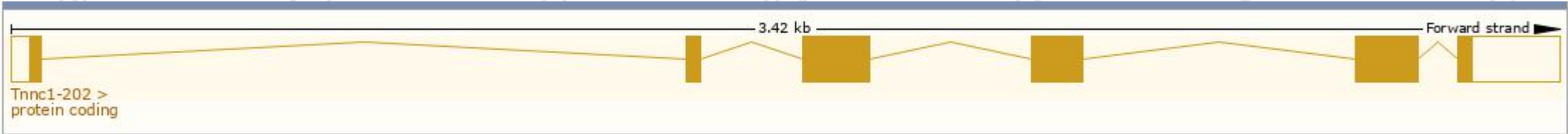
Annotation release	Status	Assembly	Chr	Location
<a href="#">108</a>	current	GRCm38.p6 ( <a href="#">GCF_000001635.26</a> )	14	NC_000080.6 (31208312..31211729)
Build 37.2	previous assembly	MGSCv37 ( <a href="#">GCF_000001635.18</a> )	14	NC_000080.5 (32021498..32024897)

# Transcript information (Ensembl)

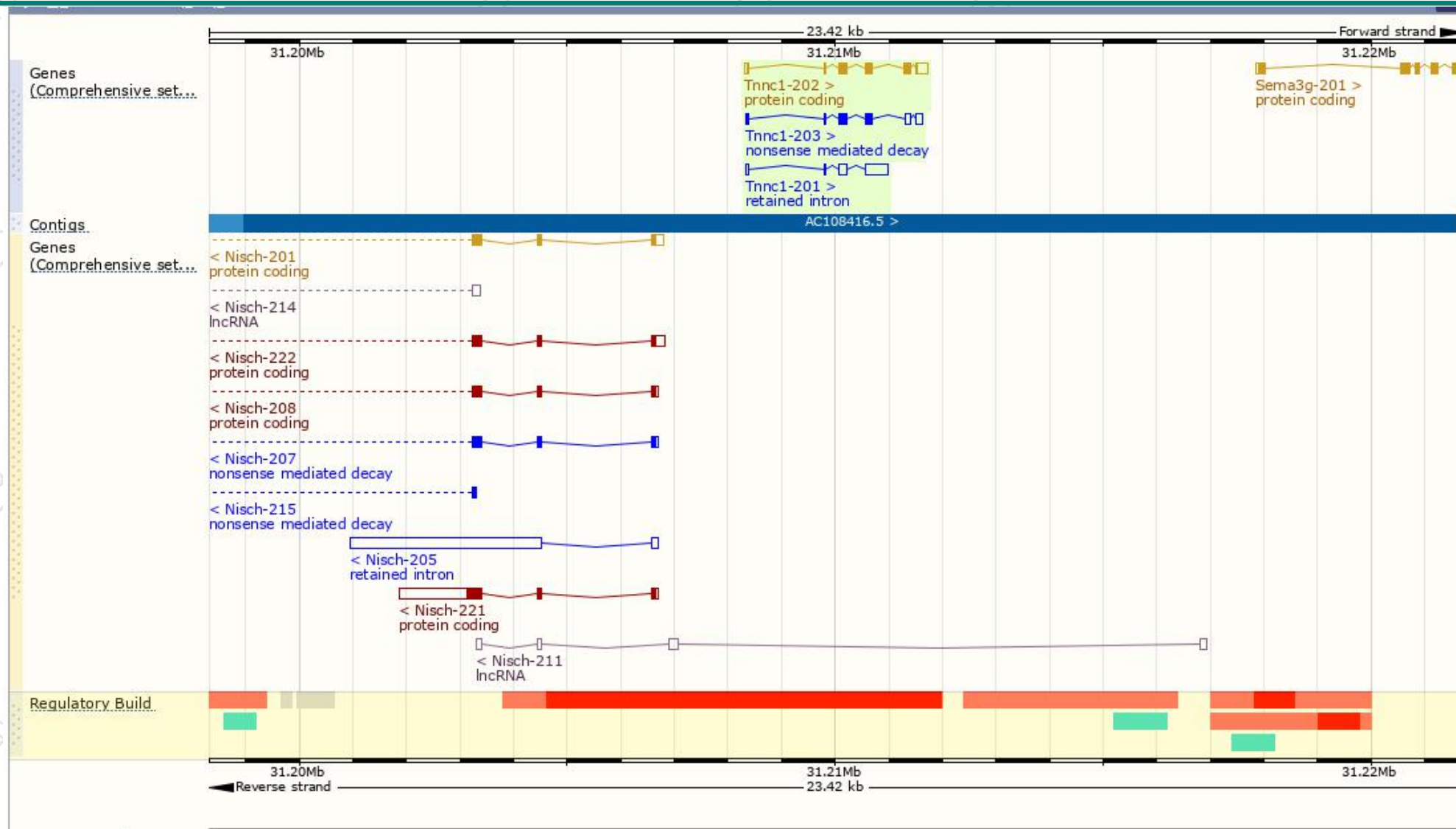
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tnnc1-201	<a href="#">ENSMUST00000161399.1</a>	660	No protein	Retained intron	-	-	TSL:2
Tnnc1-202	<a href="#">ENSMUST00000169169.7</a>	721	161aa	Protein coding	<a href="#">CCDS36854</a>	<a href="#">P19123</a>	TSL:1 GENCODE basic APPRIS P1
Tnnc1-203	<a href="#">ENSMUST00000170268.1</a>	584	106aa	Nonsense mediated decay	-	<a href="#">E9Q8P0</a>	TSL:5

The strategy is based on the design of *Tnnc1-202* transcript,The transcription is shown below

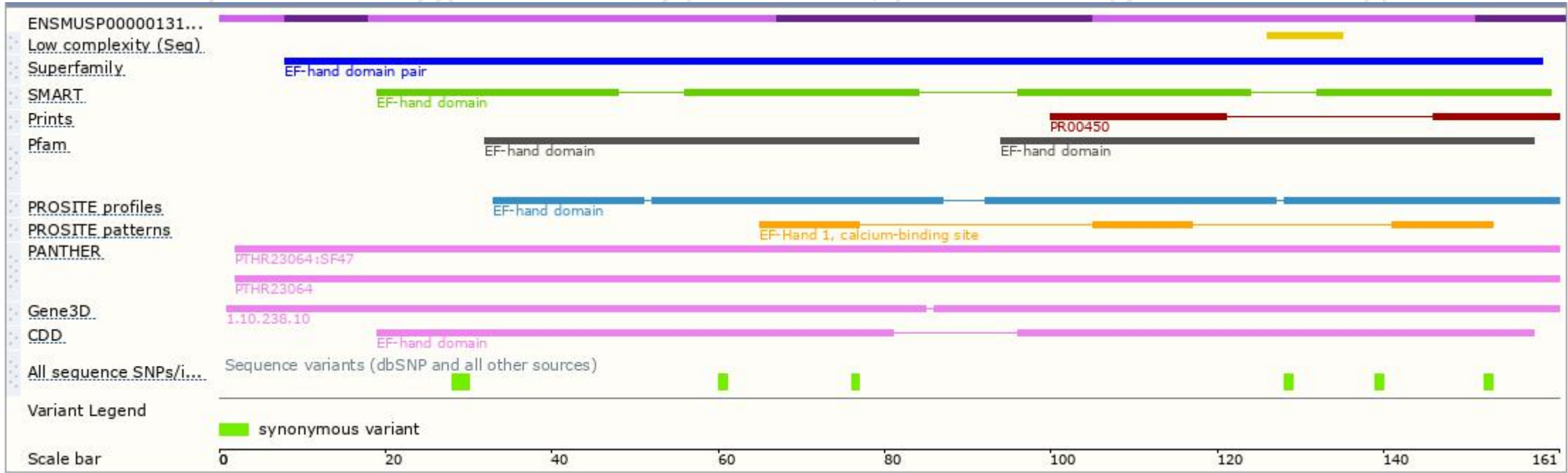


# Genomic location distribution

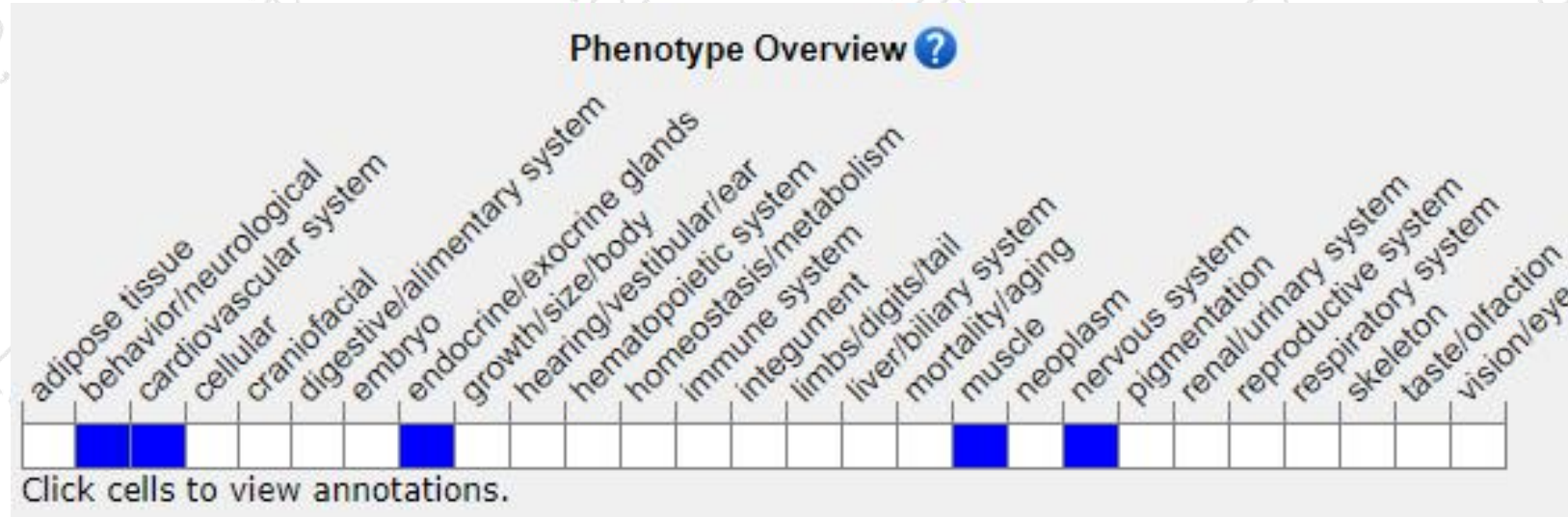




# Protein domain



# Mouse phenotype description(MGI)



*Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).*

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

Tel: 400-9660890

