

Dnaja3 Cas9-CKO Strategy

Designer:

Ruirui Zhang

Reviewer:

Huimin Su

Design Date:

2019-11-4

Project Overview

Project Name

Dnaja3

Project type

Cas9-CKO

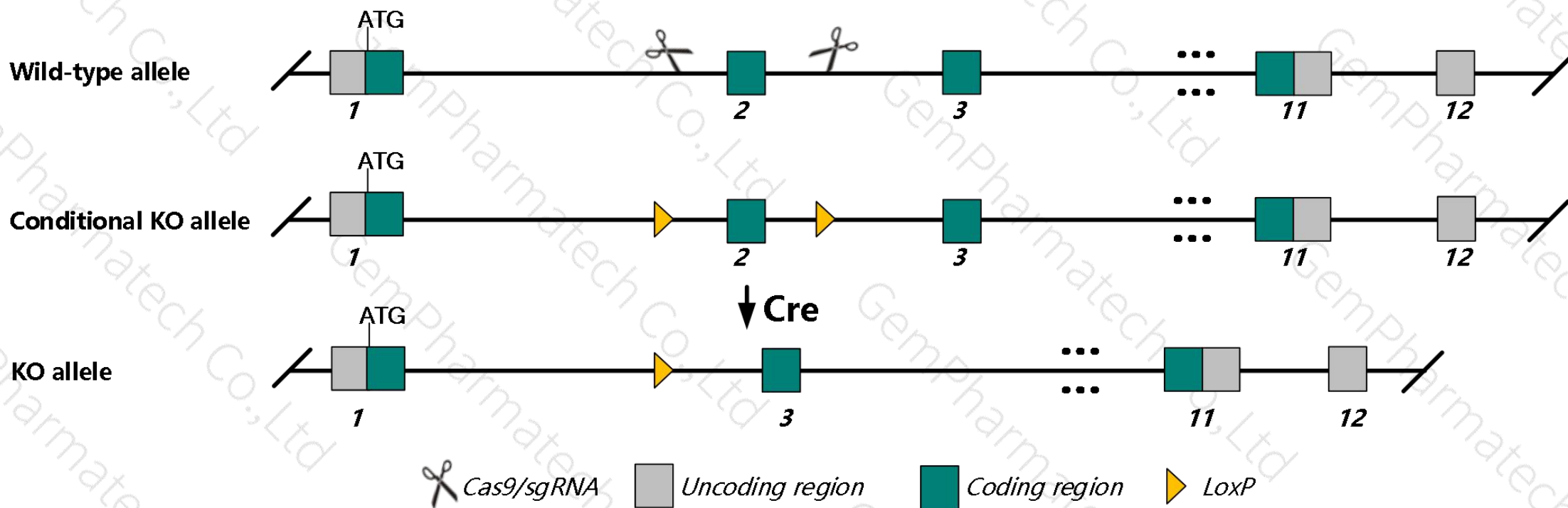
Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Dnaja3* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System



- The *Dnaja3* gene has 5 transcripts. According to the structure of *Dnaja3* gene, exon2 of *Dnaja3-201* (ENSMUST00000060067.11) transcript is recommended as the knockout region. The region contains 134bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Dnaja3* 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, early embryonic development of homozygous null embryos is disrupted. Blastocysts develop and implant but die afterwards.
- The *Dnaja3* gene is located on the Chr16. 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)

Dnaja3 DnaJ heat shock protein family (Hsp40) member A3 [*Mus musculus* (house mouse)]

Gene ID: 83945, updated on 12-Aug-2019

Summary

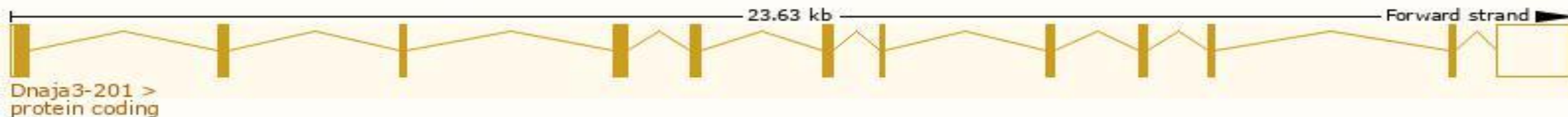
Official Symbol	Dnaja3 provided by MGI
Official Full Name	DnaJ heat shock protein family (Hsp40) member A3 provided by MGI
Primary source	MGI:MGI:1933786
See related	Ensembl:ENSMUSG00000004069
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Tid-1; Tid1l; C81173; 1200003J13Rik; 1810053A11Rik
Expression	Ubiquitous expression in heart adult (RPKM 29.3), kidney adult (RPKM 24.2) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

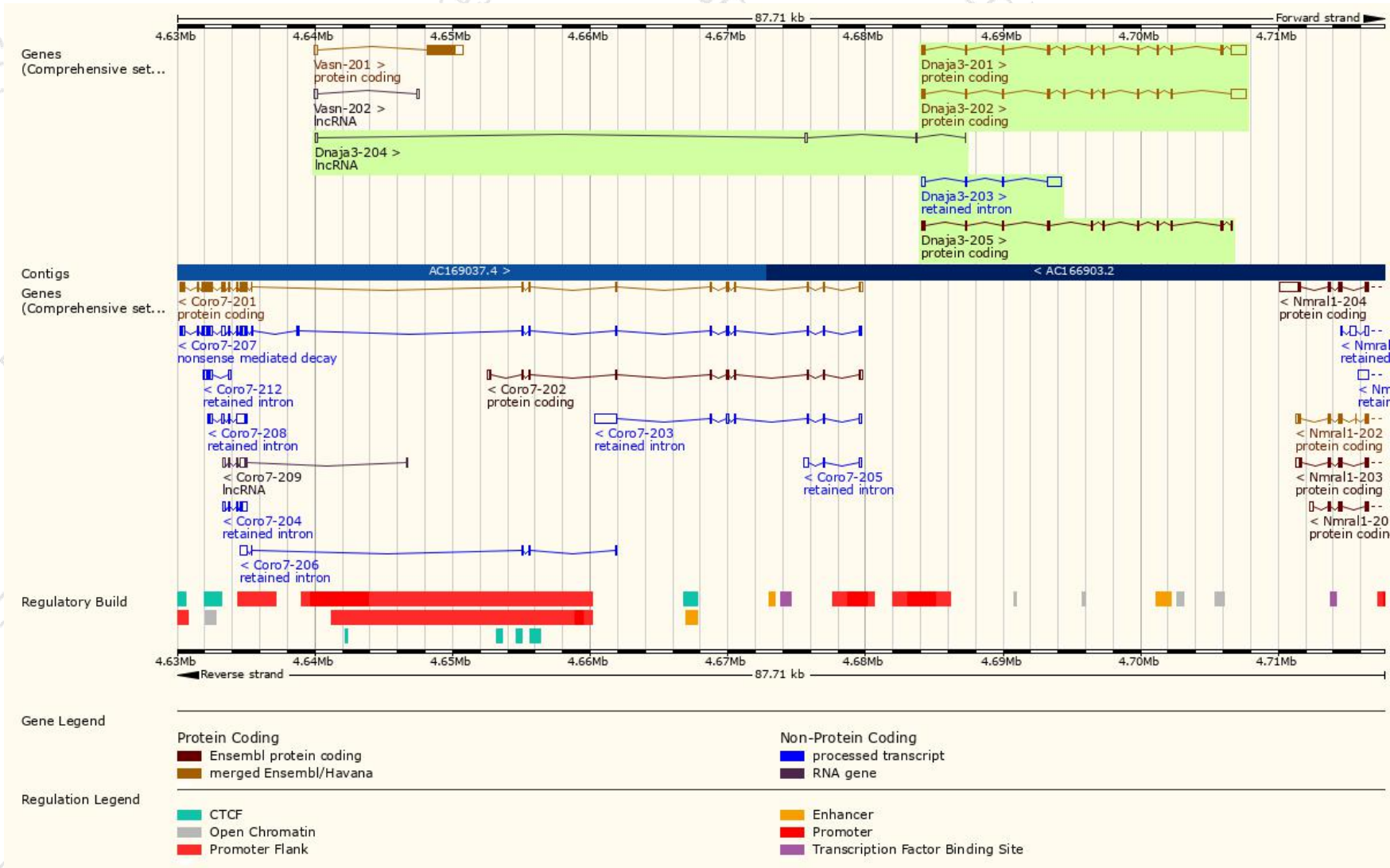
The gene has 5 transcripts, all transcripts are shown below:

Name ▲	Transcript ID ▲	bp ▲	Protein ▲	Biotype ▲	CCDS ▲	UniProt ▲	Flags ▲
Dnaja3-201	ENSMUST00000060067.11	2648	480aa	Protein coding	CCDS27922	Q99M87	TSL:1 Gencode basic APPRIS P3
Dnaja3-202	ENSMUST00000115854.3	2505	453aa	Protein coding	CCDS49748	Q3TJA9 Q99M87	TSL:1 Gencode basic APPRIS ALT1
Dnaja3-203	ENSMUST00000138495.1	1483	No protein	Retained intron	-	-	TSL:1
Dnaja3-204	ENSMUST00000144815.1	487	No protein	lncRNA	-	-	TSL:3
Dnaja3-205	ENSMUST00000229529.1	1433	429aa	Protein coding	-	Q99M87	Gencode basic

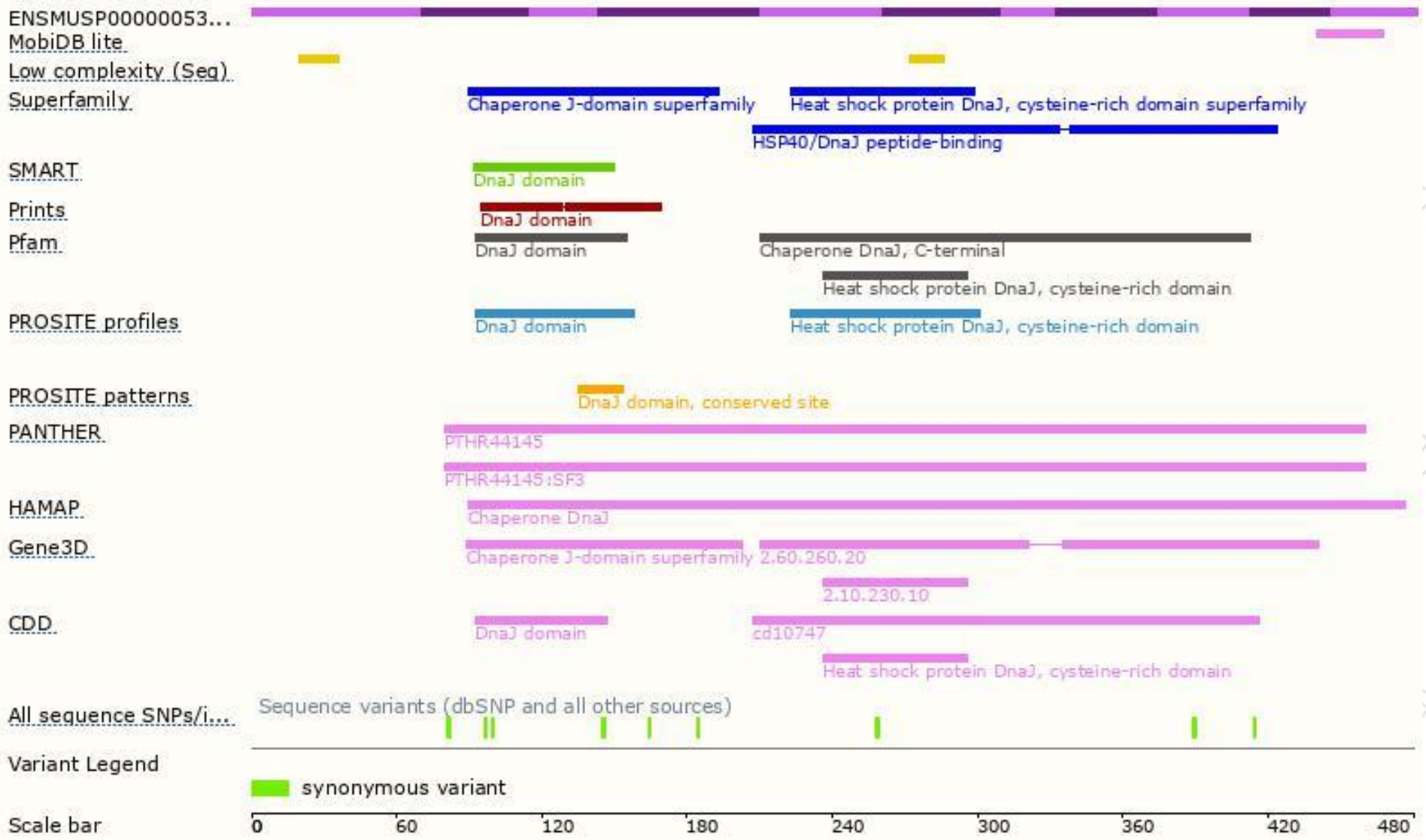
The strategy is based on the design of *Dnaja3-201* transcript, The transcription is shown below



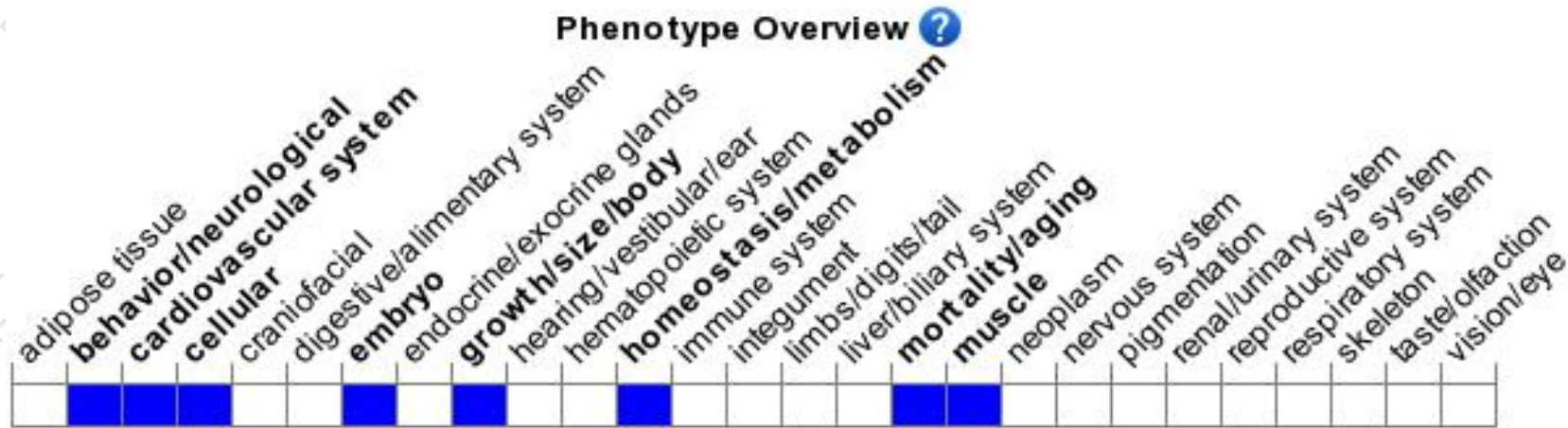
Genomic location distribution



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, Early embryonic development of homozygous null embryos is disrupted. Blastocysts develop and implant but die afterwards.

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

Tel: 400-9660890

