

***Gfi1b* Cas9-CKO Strategy**

Designer: Yupeng Yang

Reviewer: Rui Xiong

Design Date: 2018-12-18

Project Overview

Project Name

Gfi1b

Project type

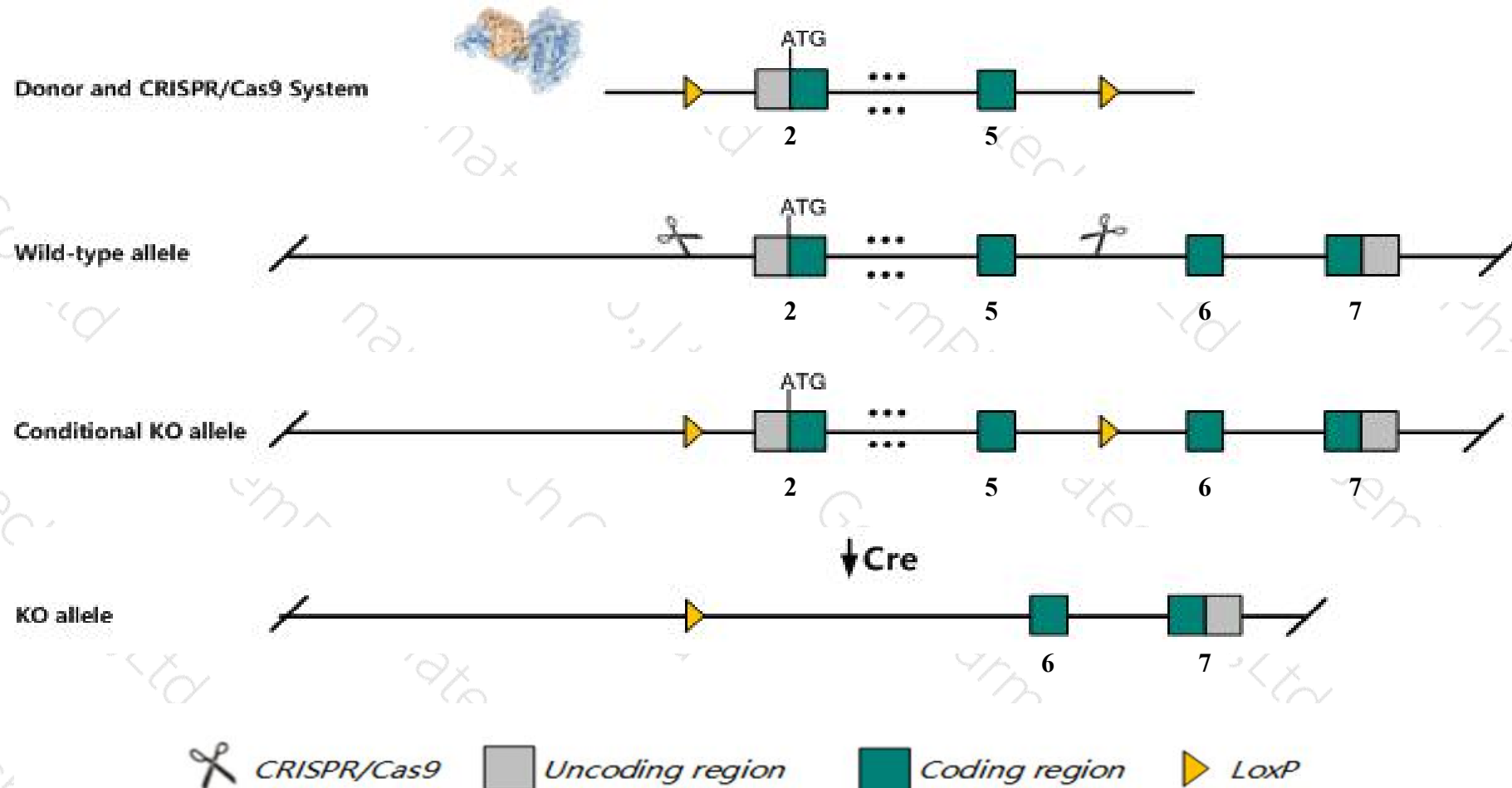
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gf11b* gene. The schematic diagram is as follows:



- The *Gfilb* gene has 4 transcripts. According to the structure of *Gfilb* gene, exon2-exon5 of *Gfilb*-204(ENSMUST00000164290.7) 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 *Gfilb* 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 disruption of this gene die as embryos by day E15. Mature adult red blood cells and megakaryocytes fail to develop.
- The *Gfilb* gene is located on the Chr2. 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)

Gfi1b growth factor independent 1B [Mus musculus (house mouse)]

Gene ID: 14582, updated on 13-Mar-2020

Summary



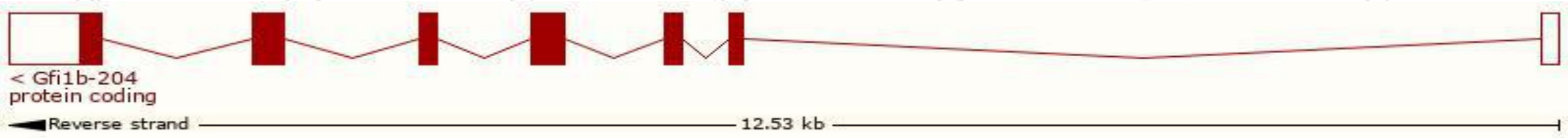
Official Symbol	Gfi1b provided by MGI
Official Full Name	growth factor independent 1B provided by MGI
Primary source	MGI:MGI:1276578
See related	Ensembl:ENSMUSG00000026815
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gfi-1B
Expression	Biased expression in liver E14.5 (RPKM 51.1), liver E14 (RPKM 38.6) and 2 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

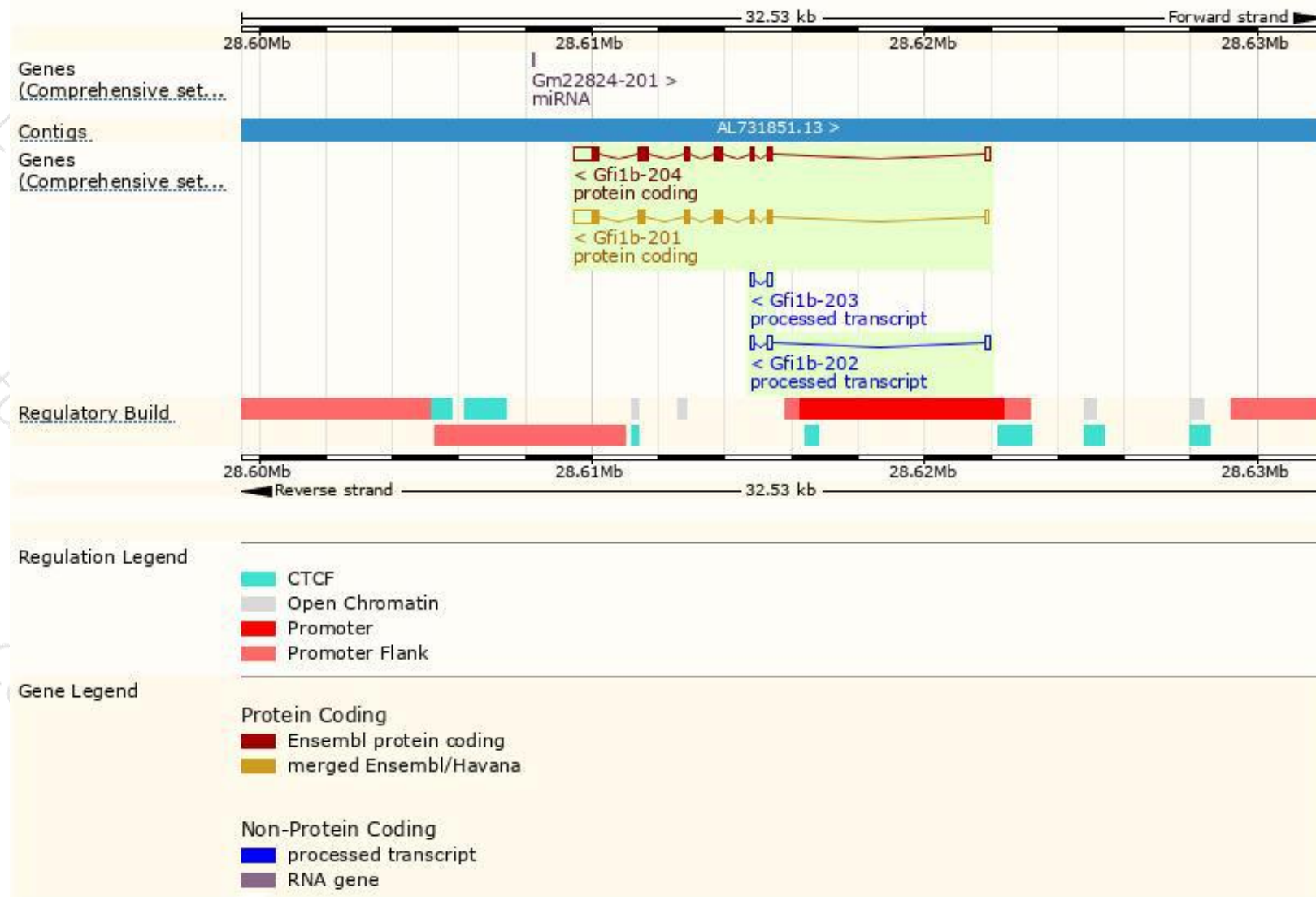
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gfi1b-204	ENSMUST00000164290.7	1822	363aa	Protein coding	CCDS50549	B7ZNH2	TSL:1 GENCODE basic APPRIS ALT2
Gfi1b-201	ENSMUST00000028156.7	1681	330aa	Protein coding	CCDS15843	O70237	TSL:1 GENCODE basic APPRIS P3
Gfi1b-202	ENSMUST00000145690.1	357	No protein	Processed transcript	-	-	TSL:2
Gfi1b-203	ENSMUST00000155686.1	238	No protein	Processed transcript	-	-	TSL:3

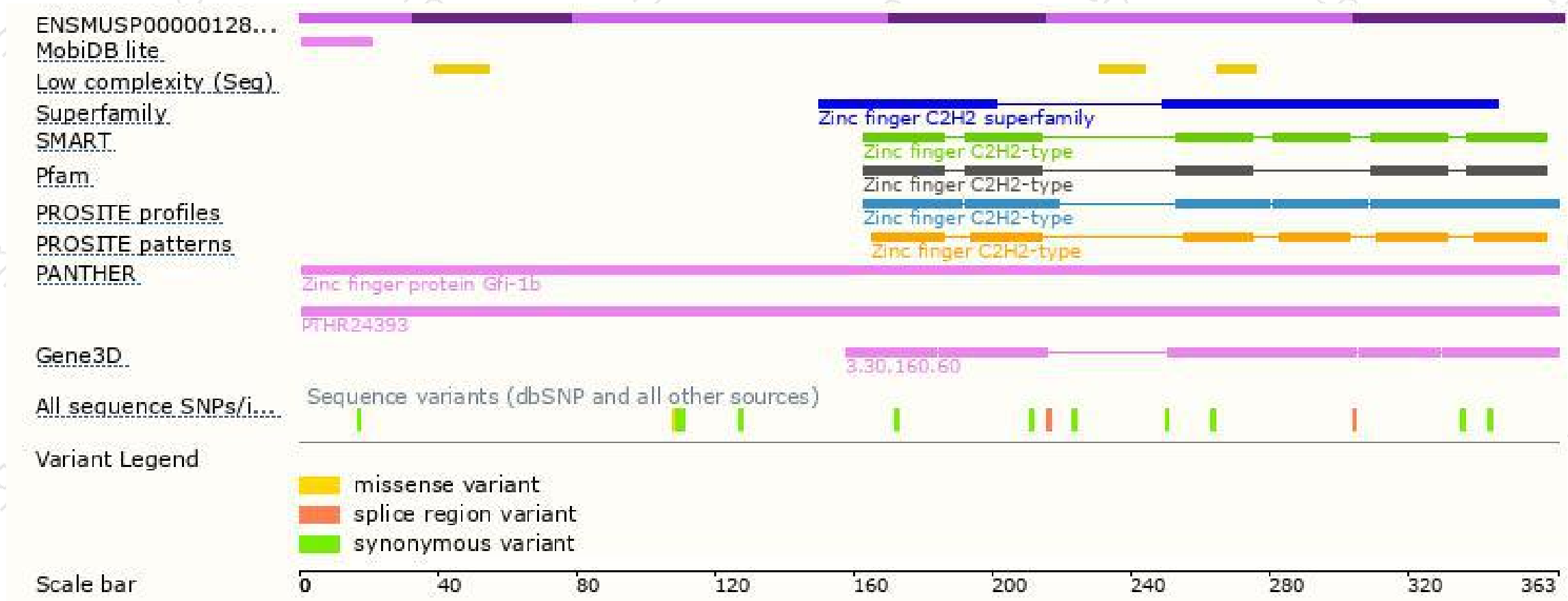
The strategy is based on the design of *Gfi1b-204* transcript,the transcription is shown below:



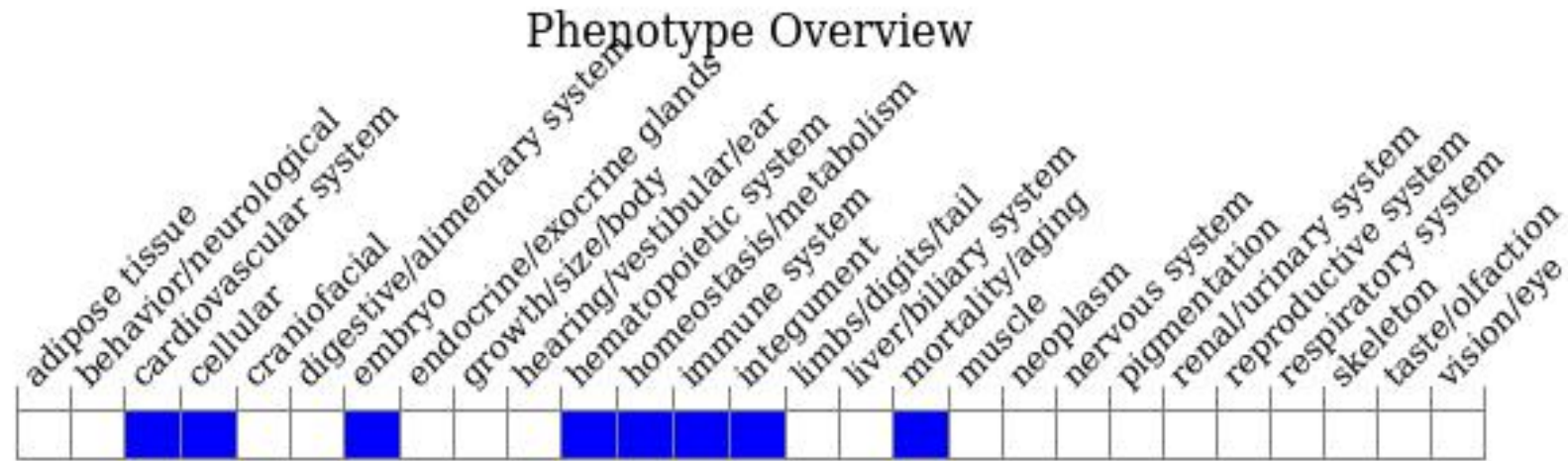
Genomic location distribution



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 disruption of this gene die as embryos by day E15. Mature adult red blood cells and megakaryocytes fail to develop.

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

Tel: 400-9660890

