

***Ralb* Cas9-CKO Strategy**

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Reviewer:

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Project Overview

Project Name

Ralb

Project type

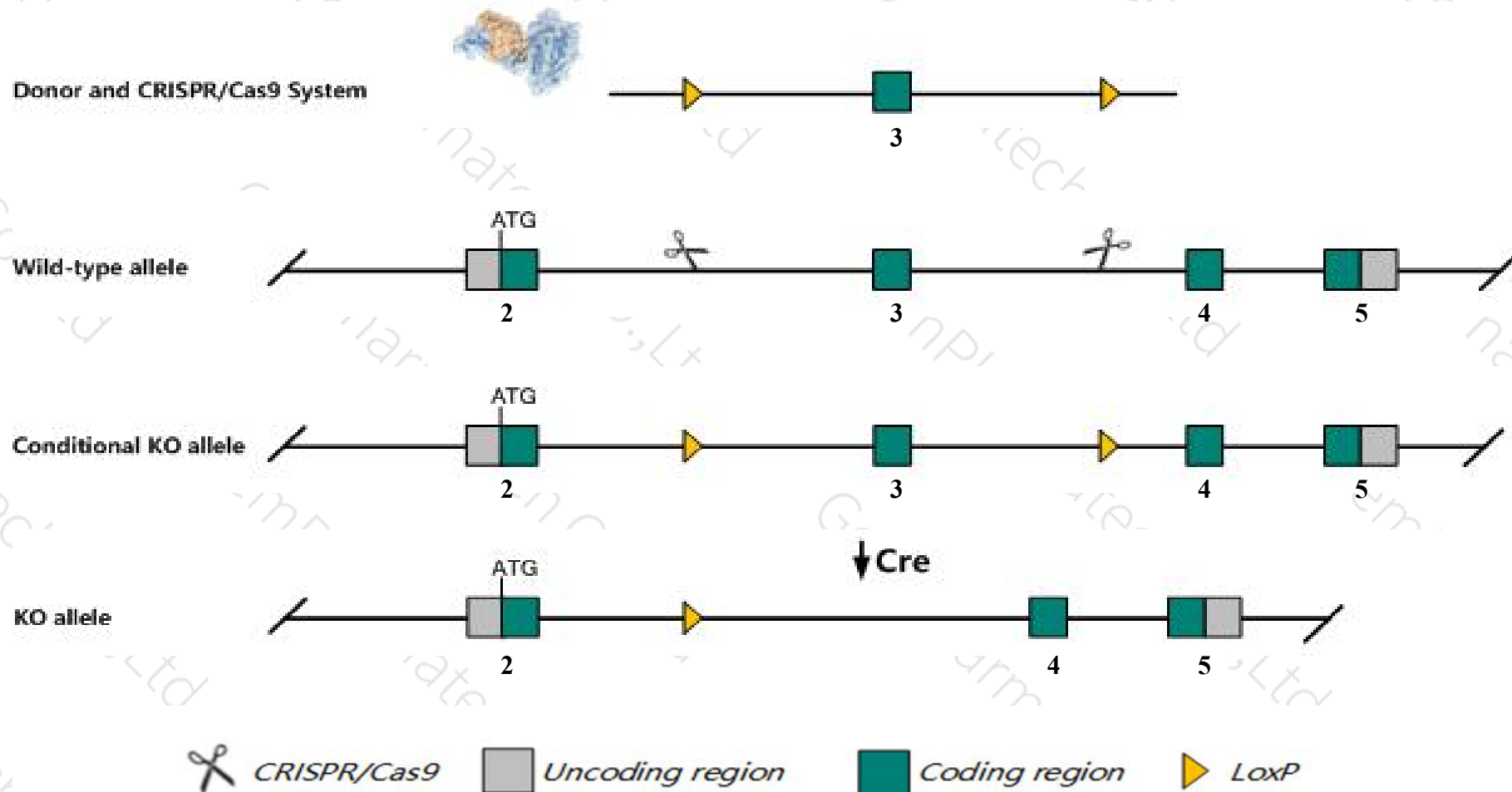
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



- The *Ralb* gene has 3 transcripts. According to the structure of *Ralb* gene, exon3 of *Ralb*-201 (ENSMUST00000004565.14) transcript is recommended as the knockout region. The region contains 209bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ralb* 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.

Notice

- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit no obvious abnormalities.
- Transcript 202 CDS 5' incomplete the influences is unknown.
- The *Ralb* gene is located on the Chr1. 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)

Ralb v-ral simian leukemia viral oncogene B [Mus musculus (house mouse)]

Gene ID: 64143, updated on 31-Jan-2019

Summary



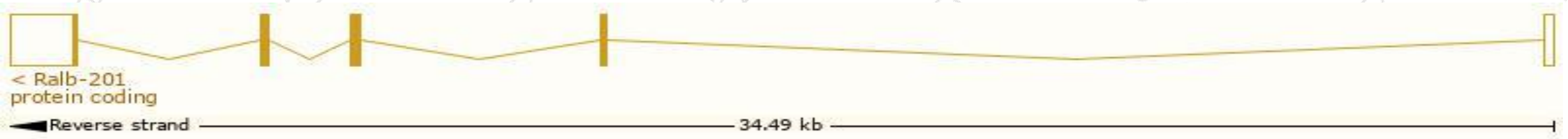
Official Symbol	Ralb provided by MGI
Official Full Name	v-ral simian leukemia viral oncogene B provided by MGI
Primary source	MGI:MGI:1927244
See related	Ensembl:ENSMUSG000000004451
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	5730472O18Rik
Expression	Ubiquitous expression in placenta adult (RPKM 48.8), large intestine adult (RPKM 23.0) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

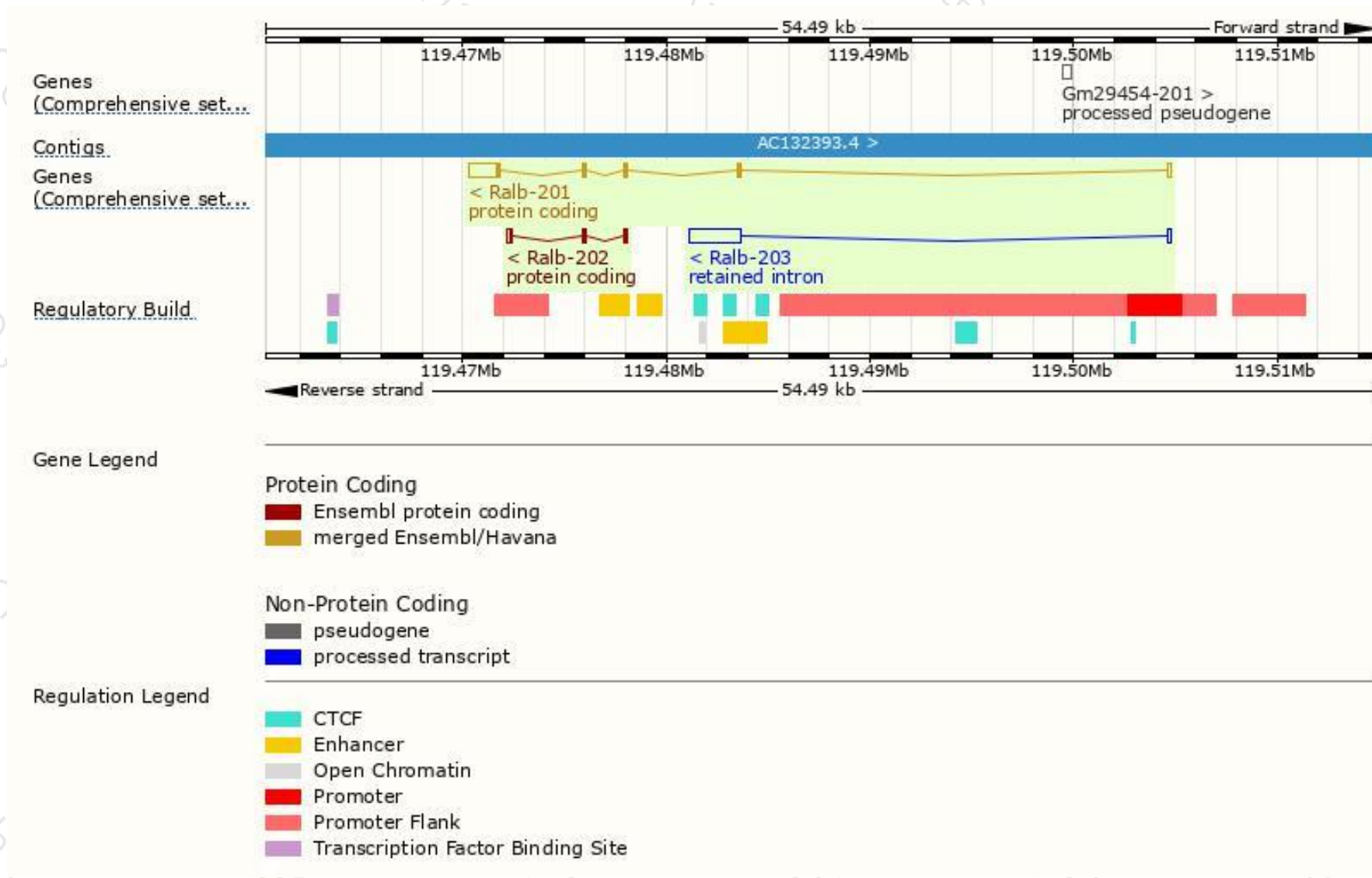
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ralb-201	ENSMUST00000004565.14	2289	206aa	Protein coding	CCDS15225	Q9JIW9	TSL:1 GENCODE basic APPRIS P1
Ralb-202	ENSMUST00000142945.2	623	149aa	Protein coding	-	F6QC68	CDS 5' incomplete TSL:3
Ralb-203	ENSMUST00000156213.1	2742	No protein	Retained intron	-	-	TSL:1

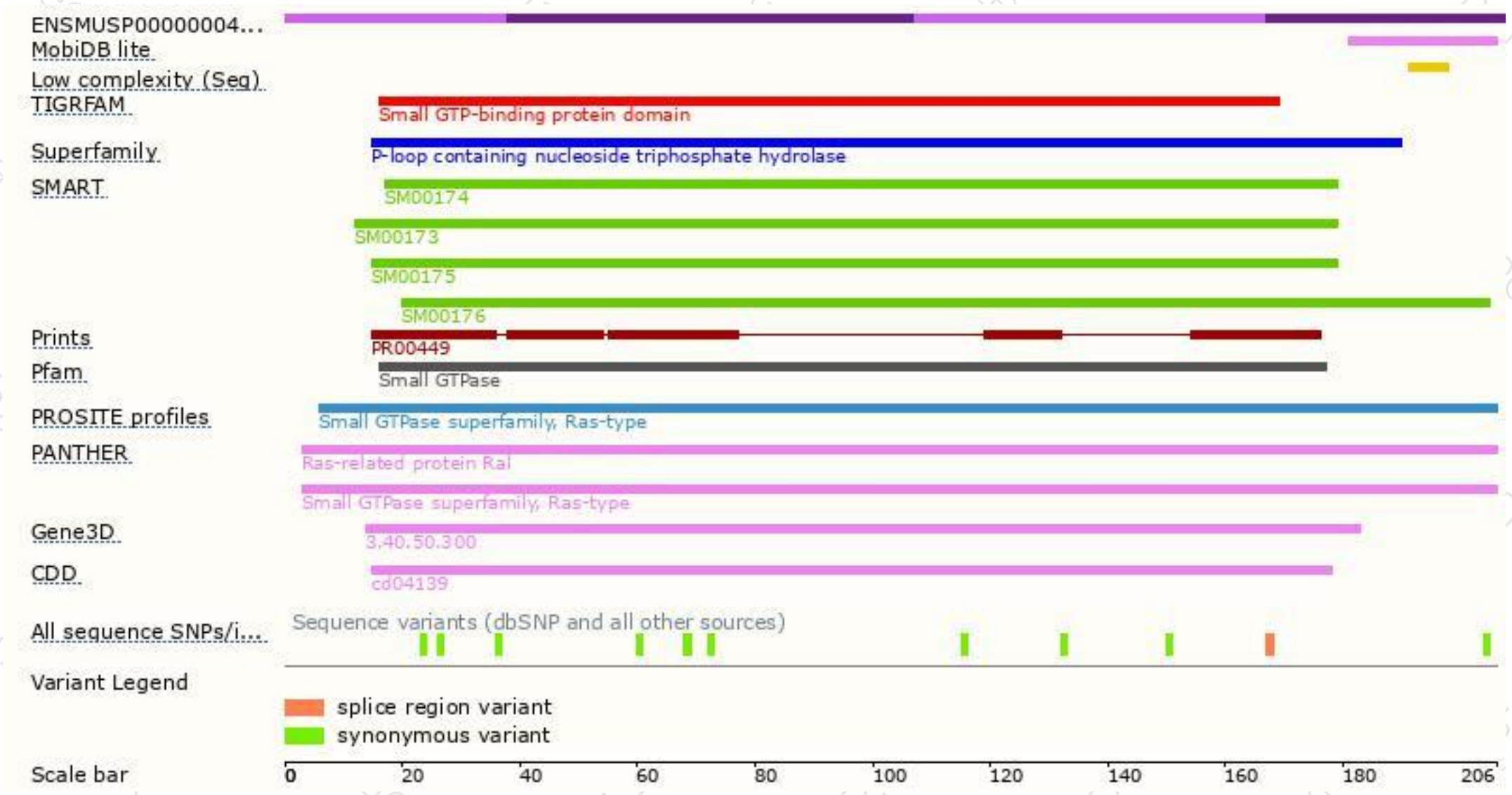
The strategy is based on the design of *Ralb-201* transcript,The transcription is shown below



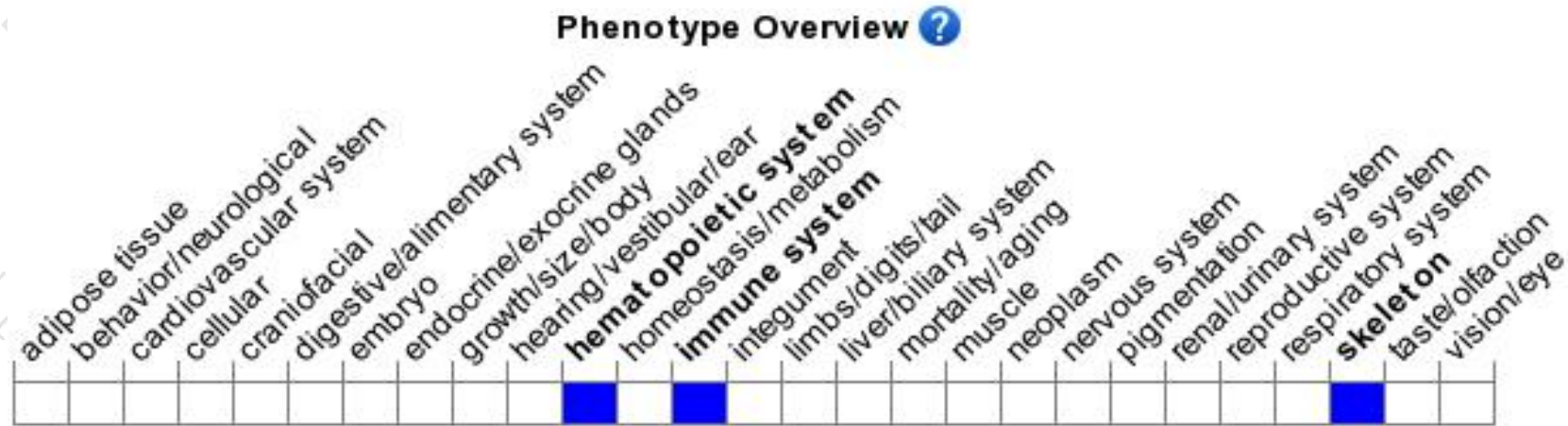
Genomic location distribution



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 no obvious abnormalities.

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

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