

Rap1a Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Rap1a

Project type

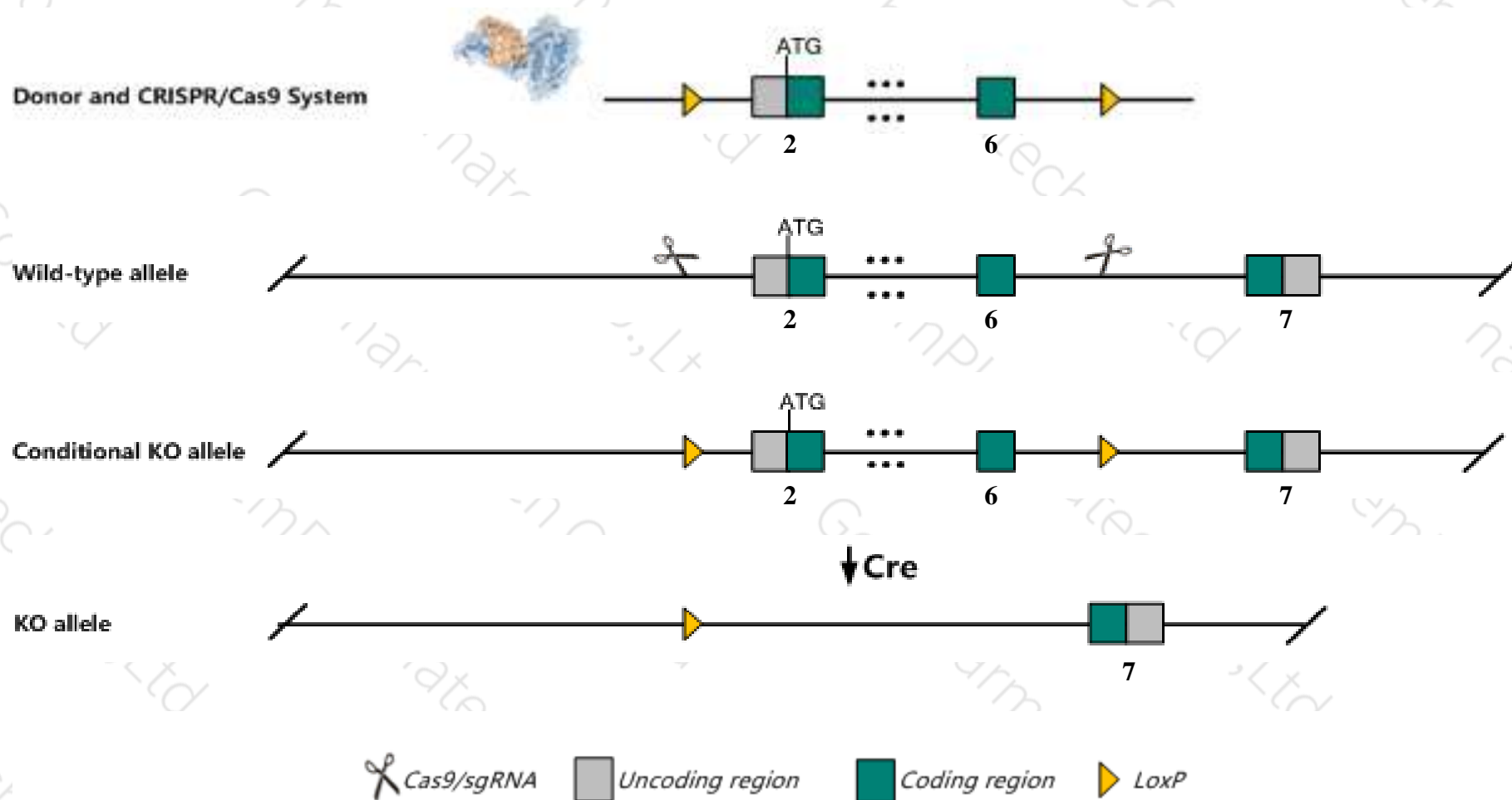
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rap1a* gene. The schematic diagram is as follows:



Technical routes

- The *Rap1a* gene has 4 transcripts. According to the structure of *Rap1a* gene, exon2-exon6 of *Rap1a-201* (ENSMUST00000090678.10) 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 *Rap1a* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was 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 a null allele exhibit impaired leukocyte migration and decreased angiogenesis.
- The *Rap1a* gene is located on the Chr3. 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)

Rap1a RAS-related protein 1a [*Mus musculus* (house mouse)]

Gene ID: 109905, updated on 25-Oct-2019

Summary

Official Symbol Rap1a provided by MGI

Official Full Name RAS-related protein 1a provided by MGI

Primary source [MGI:MGI:97852](#)

See related [Ensembl:ENSMUSG00000068798](#)

Gene type protein coding

RefSeq status VALIDATED

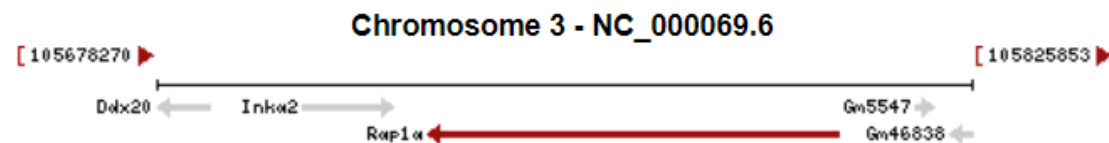
Organism [Mus musculus](#)

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as Rap1; G-22K; Krev-1; A1848598

Expression Broad expression in bladder adult (RPKM 24.1), liver E14 (RPKM 21.9) and 21 other tissues [See more](#)

Orthologs [human](#) [all](#)

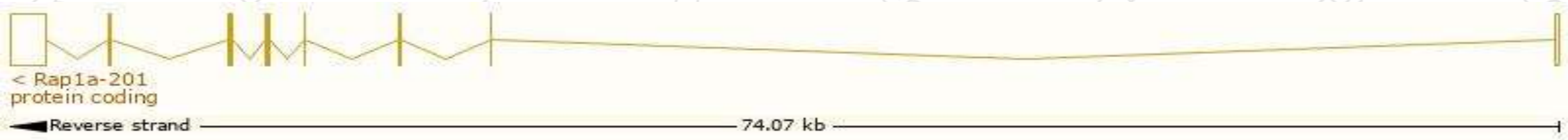


Transcript information (Ensembl)

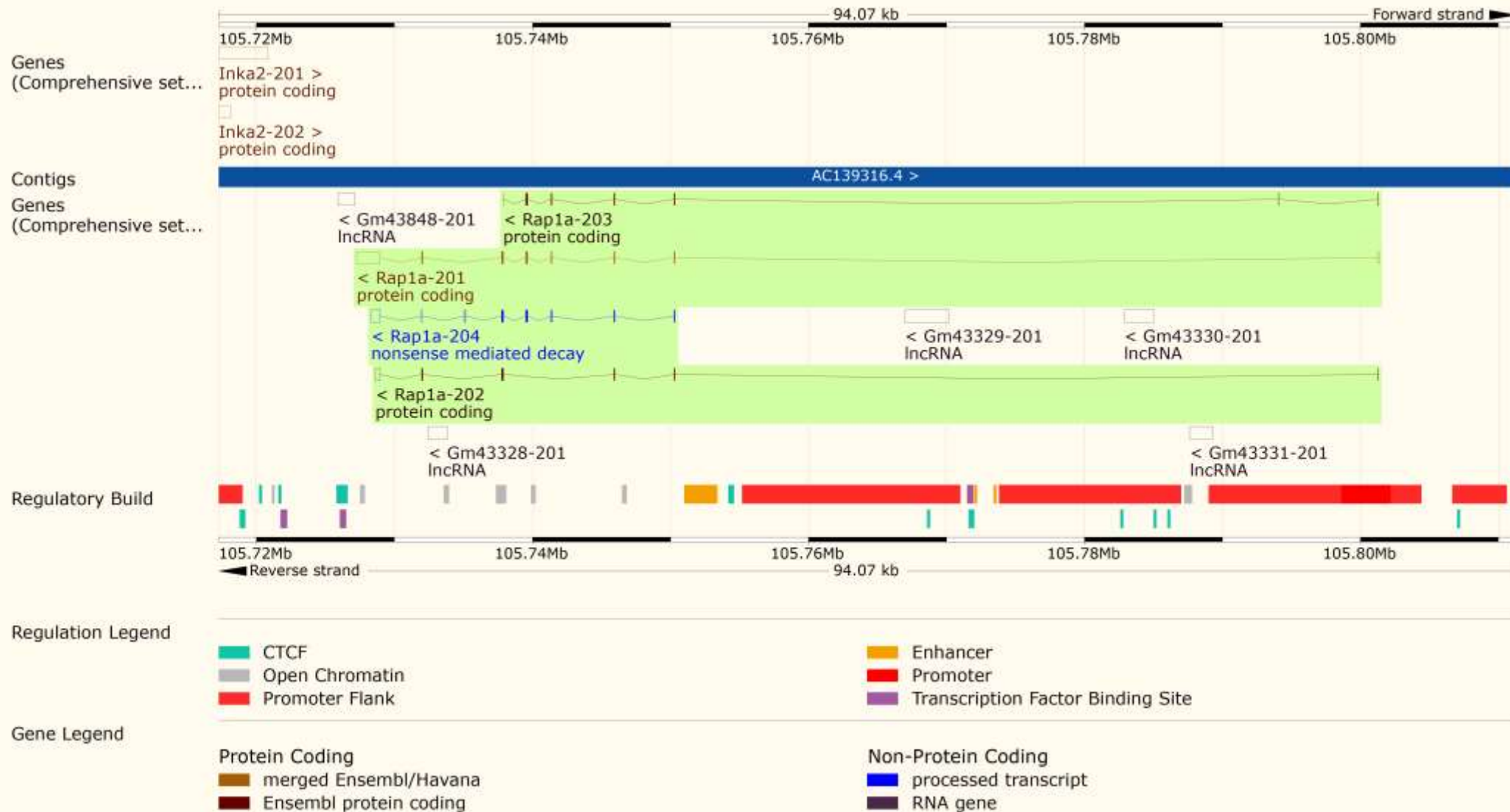
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Translation ID	Biotype	CCDS	UniProt	Flags
Rap1a-201	ENSMUST00000090678.10	2407	184aa	ENSMUSP00000088174.6	Protein coding	CCDS17711	P62835	TSL:1 GENCODE basic APPRIS P1
Rap1a-202	ENSMUST00000197094.4	782	118aa	ENSMUSP00000142419.1	Protein coding	-	A0A0G2JDL9	TSL:3 GENCODE basic
Rap1a-203	ENSMUST00000198004.1	511	109aa	ENSMUSP00000142733.1	Protein coding	-	A0A0G2JED9	CDS 3' incomplete TSL:5
Rap1a-204	ENSMUST00000199969.4	1377	159aa	ENSMUSP00000142634.1	Nonsense mediated decay	-	A0A0G2JE52	TSL:5

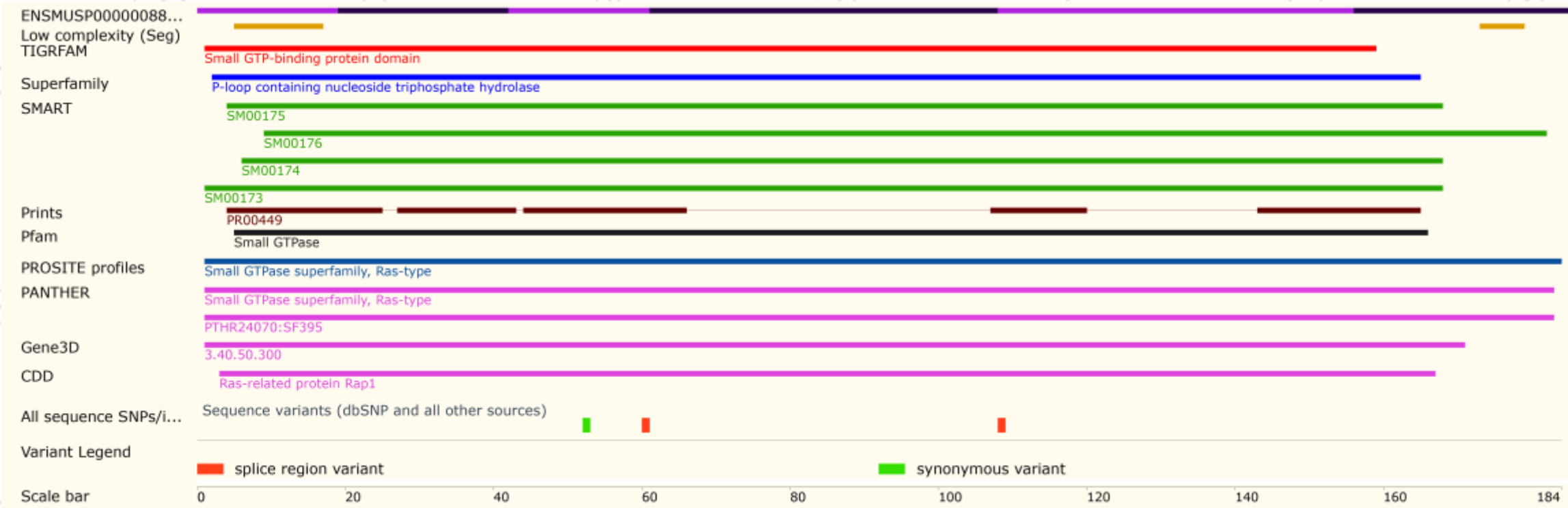
The strategy is based on the design of *Rap1a-201* transcript,The transcription is shown below



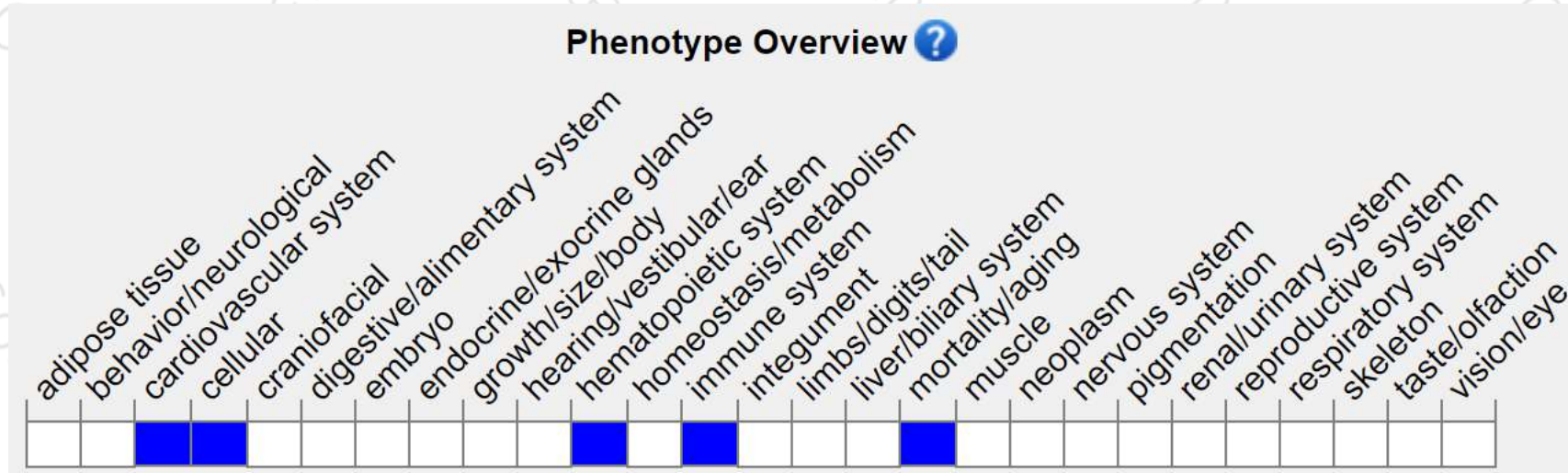
Genomic location distribution



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 null allele exhibit impaired leukocyte migration and decreased angiogenesis.

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

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