

Rassf8 Cas9-CKO Strategy

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Project Overview

Project Name

Rassf8

Project type

Cas9-CKO

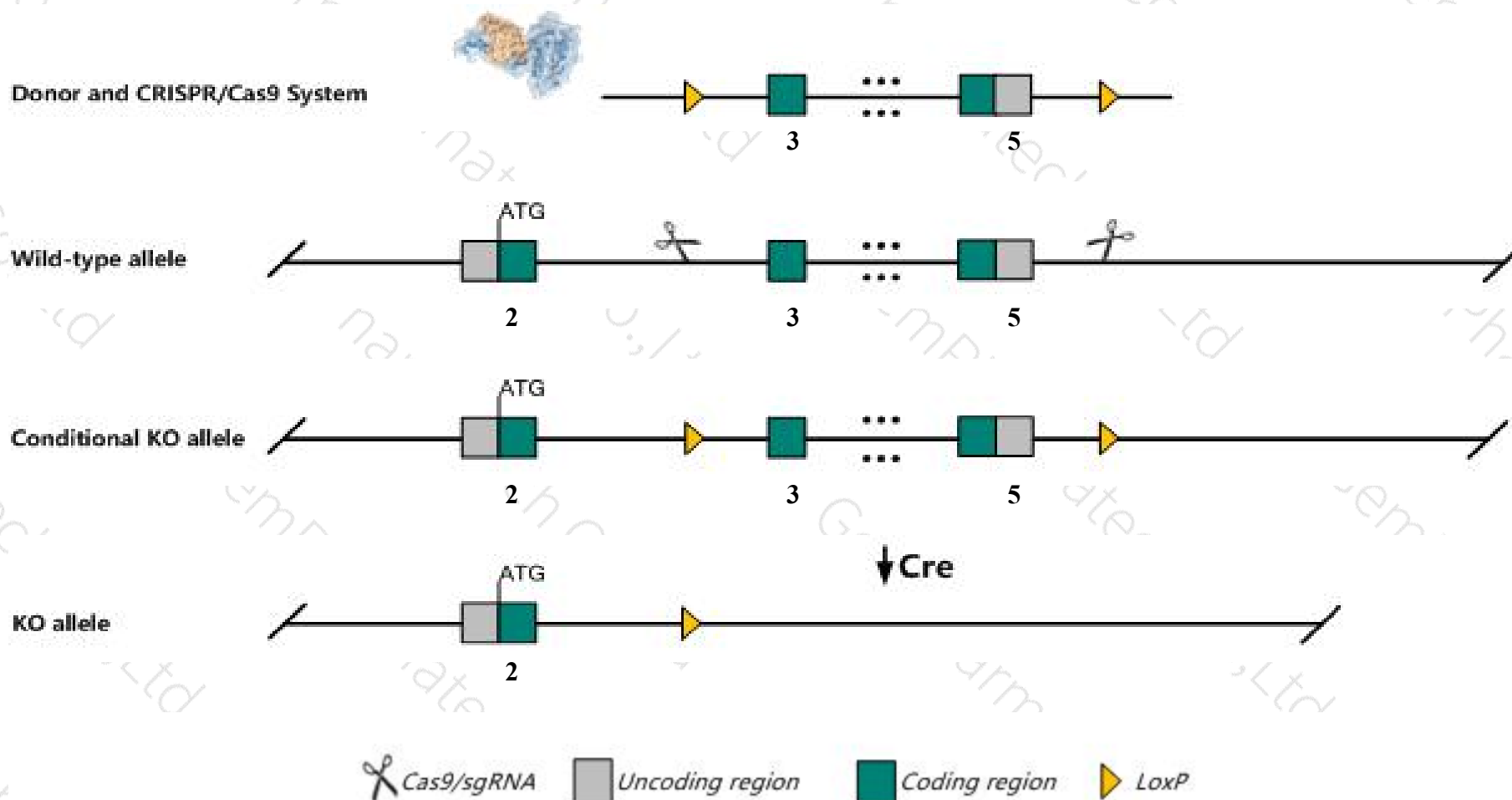
Strain background

C57BL/6JGpt

- The *Rassf8* gene has 4 transcripts. According to the structure of *Rassf8* gene, exon3-exon5 of *Rassf8*-203(ENSMUST00000111704.7) transcript is recommended as the knockout region. The region contains 1157bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rassf8* 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.

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rassf8* gene. The schematic diagram is as follows:



- The *Rassf8* gene is located on the Chr6. 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)

Rassf8 Ras association (RalGDS/AF-6) domain family (N-terminal) member 8 [Mus musculus (house mouse)]

Gene ID: 71323, updated on 13-Mar-2020

Summary



Official Symbol Rassf8 provided by [MGI](#)

Official Full Name Ras association (RalGDS/AF-6) domain family (N-terminal) member 8 provided by [MGI](#)

Primary source [MGI:MGI:1918573](#)

See related [Ensembl:ENSMUSG00000030259](#)

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 5133400D11Rik, AA815821, AW123240, Hoj1, mHoj-1

Expression Ubiquitous expression in placenta adult (RPKM 11.3), limb E14.5 (RPKM 7.6) and 26 other tissues [See more](#)

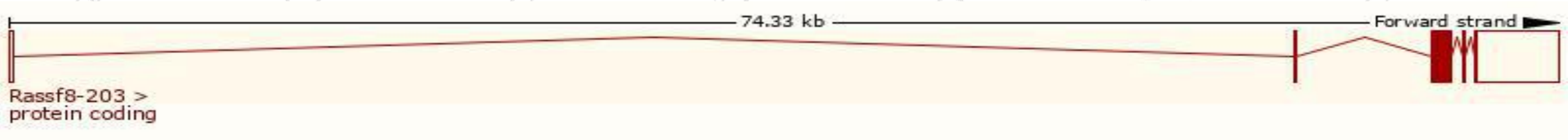
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

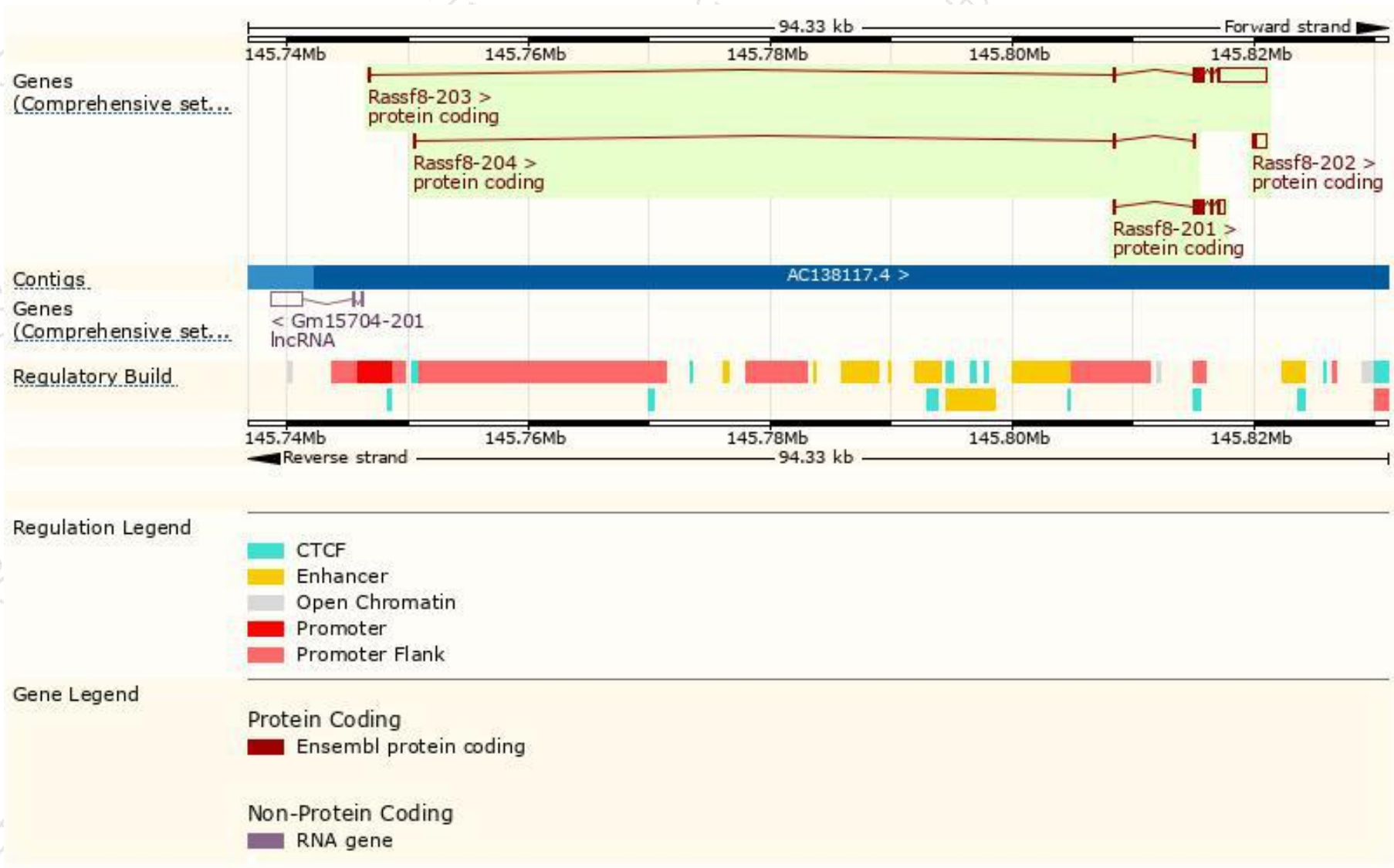
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rassf8-203	ENSMUST00000111704.7	5446	419aa	Protein coding	CCDS20696	Q8CJ96	TSL:5 GENCODE basic APPRIS P1
Rassf8-201	ENSMUST00000032388.4	1733	419aa	Protein coding	CCDS20696	Q8CJ96	TSL:1 GENCODE basic APPRIS P1
Rassf8-202	ENSMUST00000058538.6	1131	102aa	Protein coding	-	F7BB71	TSL:NA GENCODE basic
Rassf8-204	ENSMUST00000140966.1	427	79aa	Protein coding	-	E9QA00	CDS 3' incomplete TSL:2

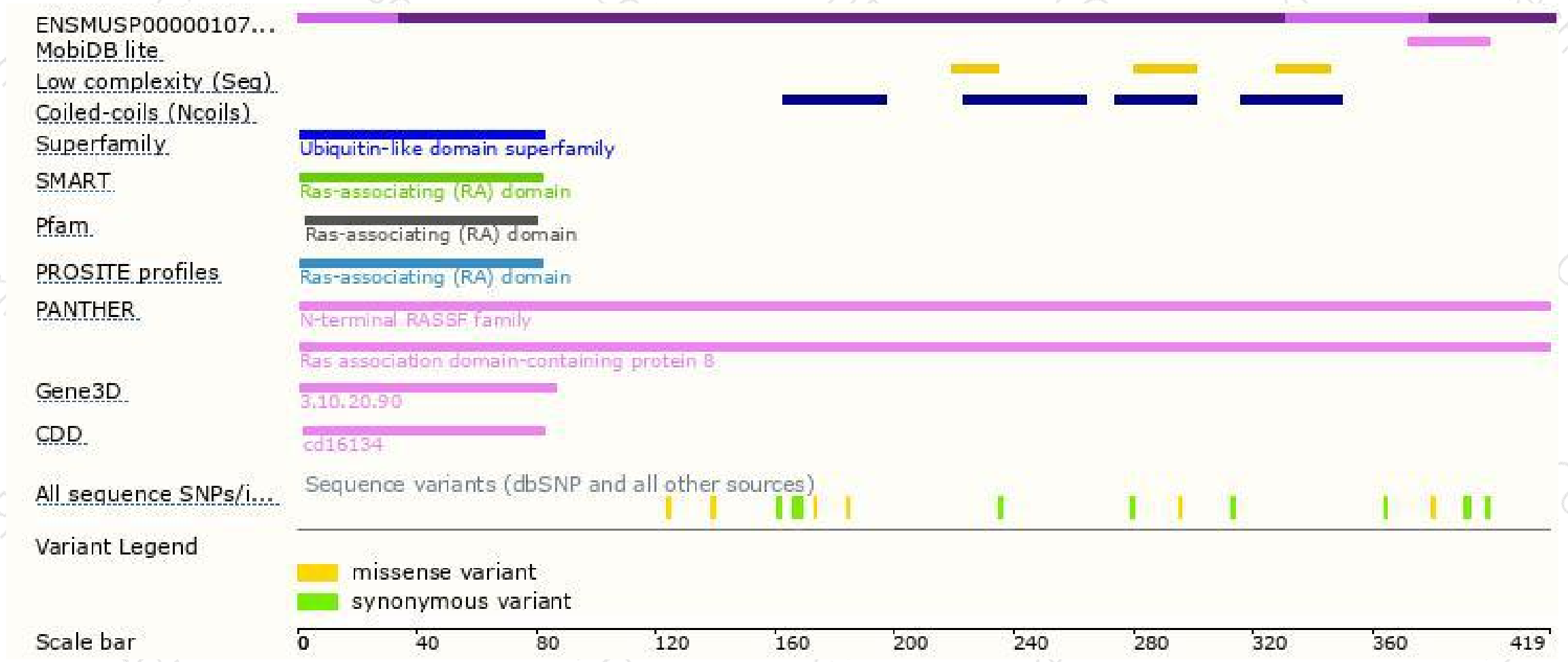
The strategy is based on the design of *Rassf8-203* transcript,the transcription is shown below:



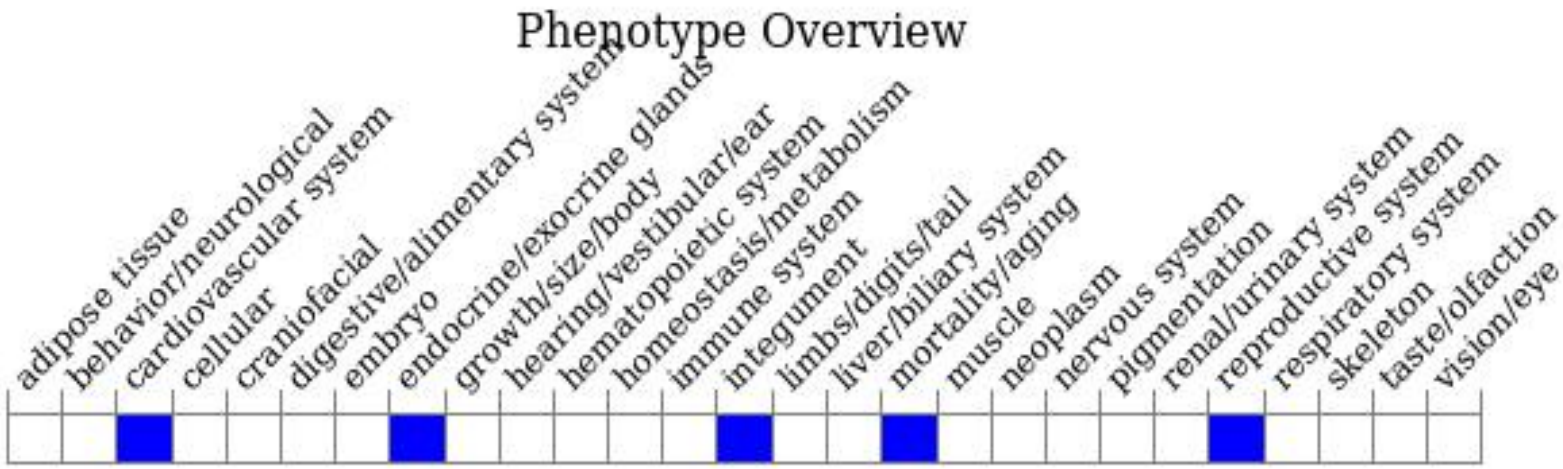
Genomic location distribution



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.

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