Marco Cas9-KO Strategy

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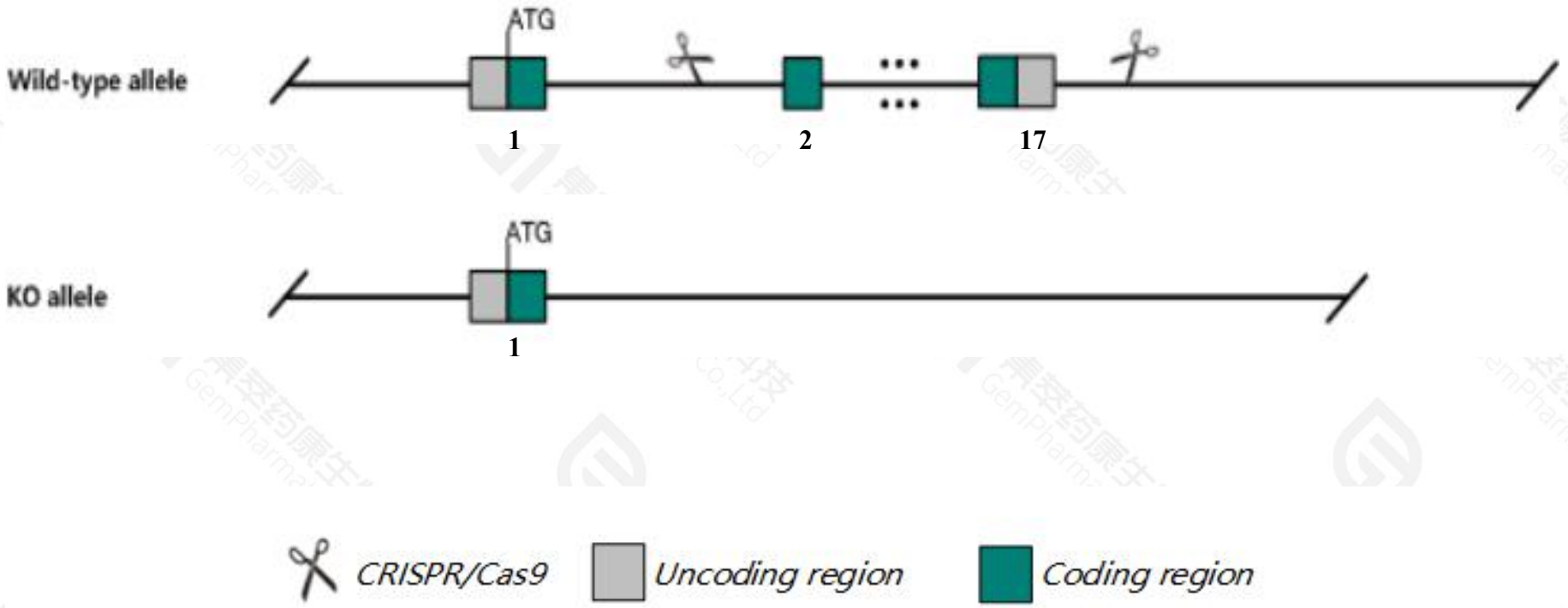
Project Overview

Project Name	***Marco***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

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



Technical routes

➢ The *Marco* gene has 2 transcripts. According to the structure of *Marco* gene, exon2-exon17 of *Marco-201*(ENSMUST00000027639.8) transcript is recommended as the knockout region.The region contains 1448bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Marco* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice

➢ According to the existing MGI data,mice homozygous for a null allele show altered spleen marginal zone architecture and impaired IgM responses to a pneumococcal polysaccharide vaccine. Mice homozygous for another null allele show increased susceptibility to bacterial pneumonia and enhanced inflammatory responses to inhaled particles.

➢ The *Marco* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

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Marco macrophage receptor with collagenous structure [Mus musculus (house mouse)]

Gene ID: 17167, updated on 15-Mar-2020

Summary

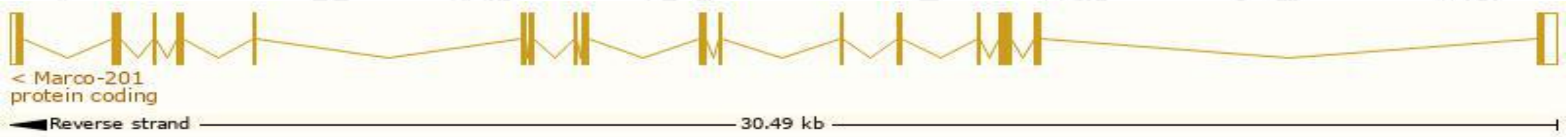
Official Symbol	Marco provided by MGI
Official Full Name	macrophage receptor with collagenous structure provided by MGI
Primary source	MGI:MGI:1309998
See related	Ensembl:ENSMUSG00000026390
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI323439, Ly112, Scara2
Expression	Biased expression in liver E18 (RPKM 27.1), liver E14 (RPKM 17.6) and 4 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

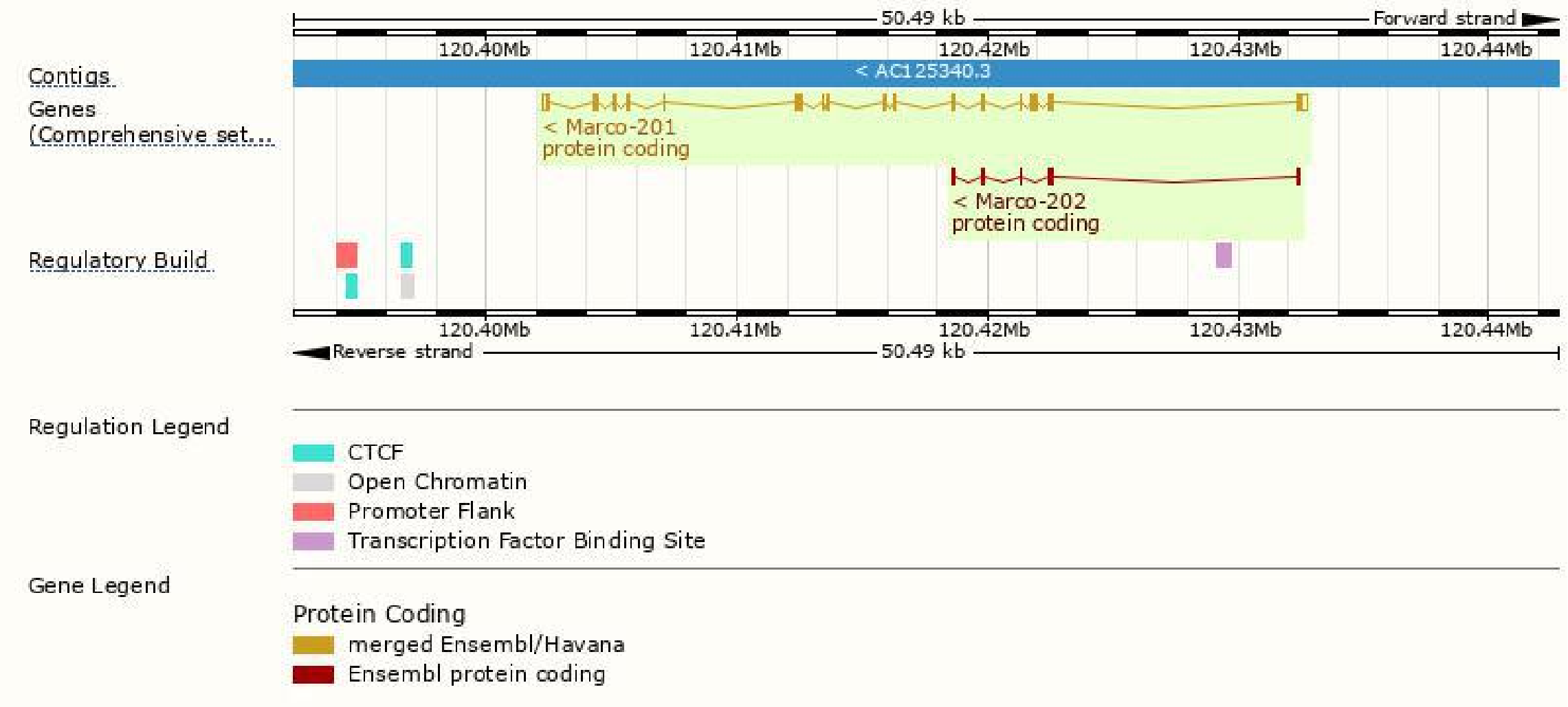
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Marco-201	ENSMUST00000027639.7	1925	518aa	Protein coding	CCDS15234	A2RT24 Q60754	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Marco-202	ENSMUST00000186432.2	370	123aa	Protein coding	-	A0A087WS94	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3

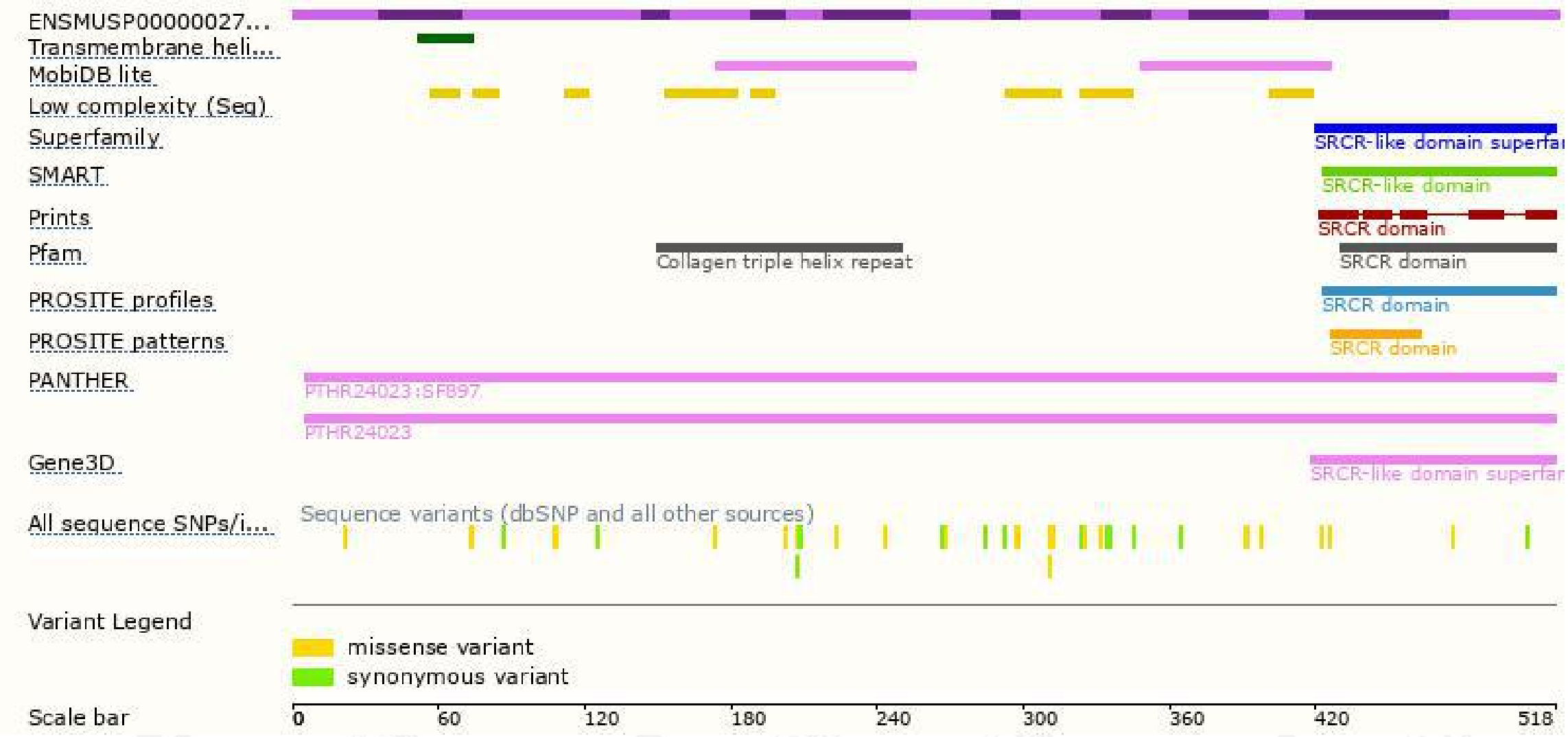
The strategy is based on the design of *Marco-201* transcript,the transcription is shown below:



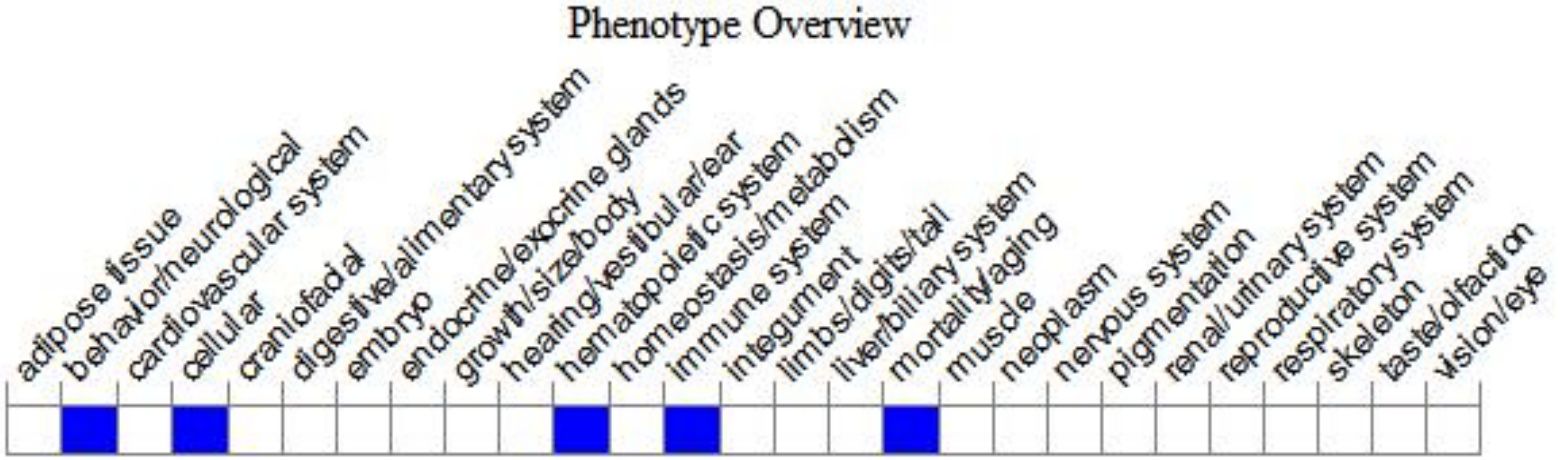
Genomic location distribution



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/).

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