Trem2 Cas9-CKO Strategy

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Design Date: 2019-9-11
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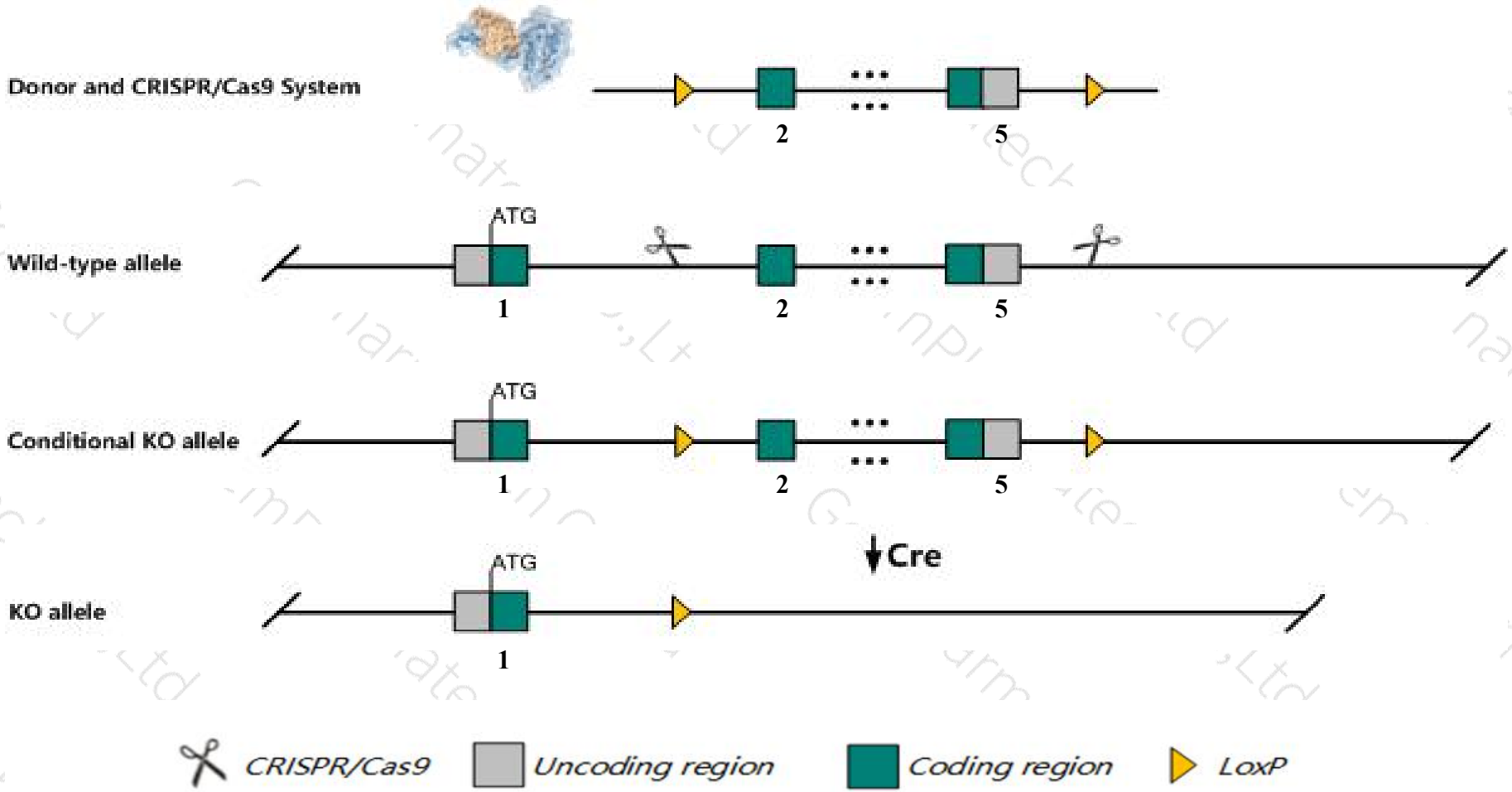
Project Overview

Project Name	***Trem2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Trem2* gene. The schematic diagram is as follows:



Technical routes

- The *Trem2* gene has 4 transcripts. According to the structure of *Trem2* gene, exon2-exon5 of *Trem2-202* (ENSMUST00000113237.3) transcript is recommended as the knockout region.The region contains 710bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Trem2* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 display enhanced cytokine production by macrophages in response to toll-like receptor agonists. Mice homozygous for a different knock-out allele show reduced microglial cell survival, proliferation and activation and cell cycle arrest at the G1/S checkpoint.

- The *Trem2* gene is located on the Chr17. 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）

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Trem2 triggering receptor expressed on myeloid cells 2 [Mus musculus (house mouse)]

Gene ID: 83433, updated on 9-Apr-2019

Summary

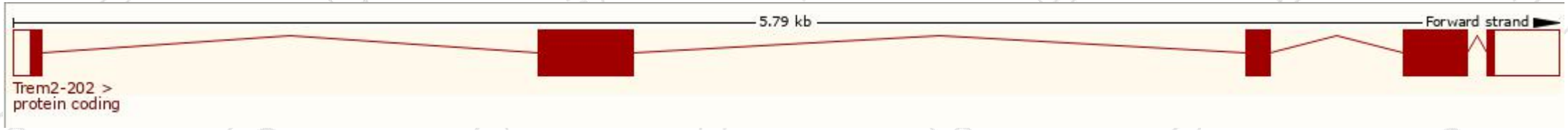
Official Symbol	Trem2 provided by MGI
Official Full Name	triggering receptor expressed on myeloid cells 2 provided by MGI
Primary source	MGI:MGI:1913150
See related	Ensembl:ENSMUSG00000023992
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	TREM-2, Trem2a, Trem2b, Trem2c
Summary	The protein encoded by this gene is part of the immunoglobulin and lectin-like superfamily and functions as part of the innate immune system. This gene forms part of a cluster of genes on mouse chromosome 17 thought to be involved in innate immunity. This protein associates with the adaptor protein Dap-12 and recruits several factors, such as kinases and phospholipase C-gamma, to form a receptor signaling complex that activates myeloid cells, including dendritic cells and microglia. In humans homozygous loss-of-function mutations in this gene cause Nasu-Hakola disease and mutations in this gene may be risk factors to the development of Alzheimer's disease. In mouse mutations of this gene serve as a pathophysiological model for polycystic lipomembranous osteodysplasia with sclerosing leukoencephalopathy (Nasu-Hakola disease) and for inflammatory bowel disease. Alternative splicing results in multiple transcript variants that encode different protein isoforms. [provided by RefSeq, Jan 2013]
Expression	Broad expression in subcutaneous fat pad adult (RPKM 19.6), ovary adult (RPKM 10.2) and 20 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

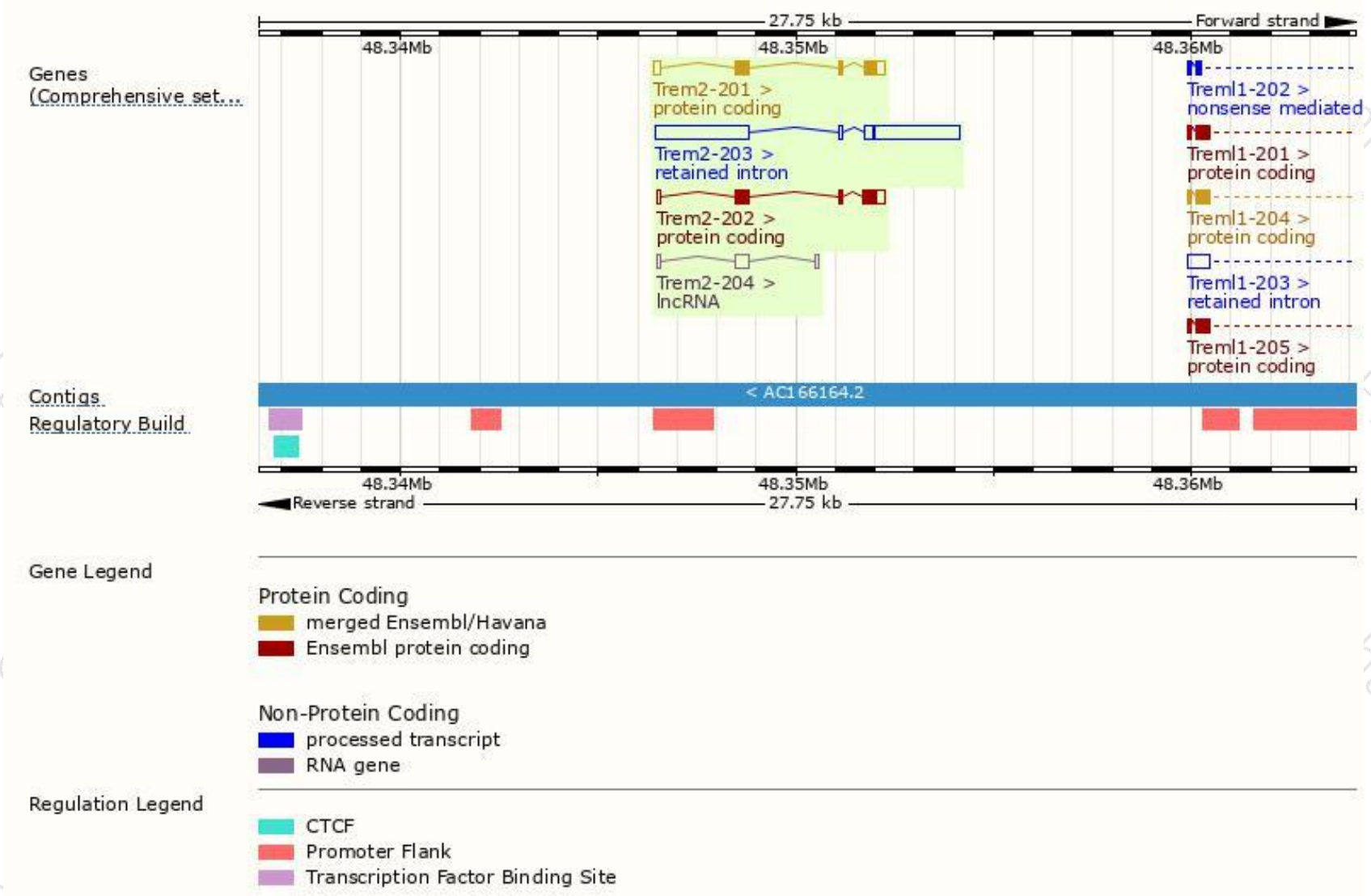
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Trem2-201	ENSMUST00000024791.14	1088	227aa	Protein coding	CCDS28865	Q99NH8	TSL:1 GENCODE basic APPRIS P3
Trem2-202	ENSMUST00000113237.3	1056	249aa	Protein coding	CCDS70825	Q99NH8	TSL:1 GENCODE basic APPRIS ALT2
Trem2-204	ENSMUST00000148545.1	526	No protein	Processed transcript	-	-	TSL:3
Trem2-203	ENSMUST00000132340.1	4784	No protein	Retained intron	-	-	TSL:2

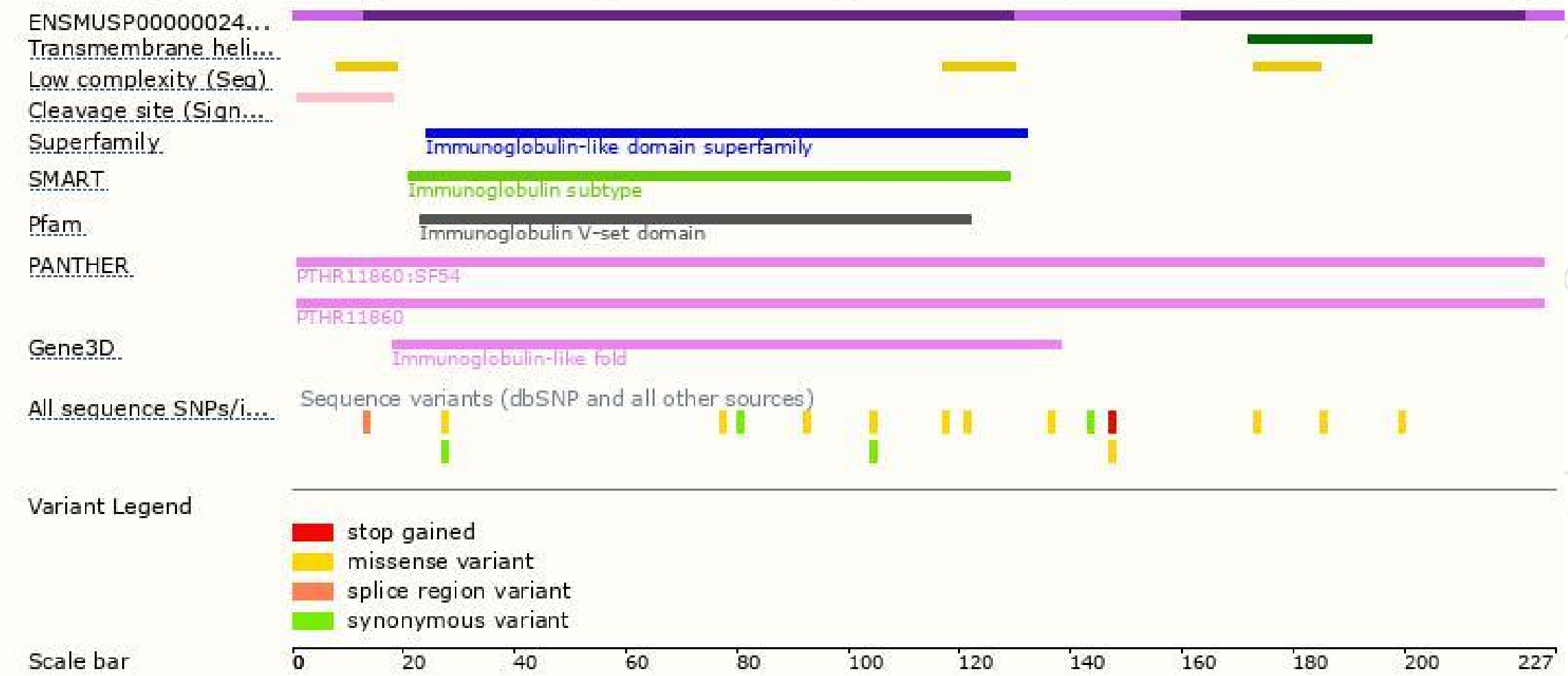
The strategy is based on the design of *Trem2-202* transcript,The transcription is shown below



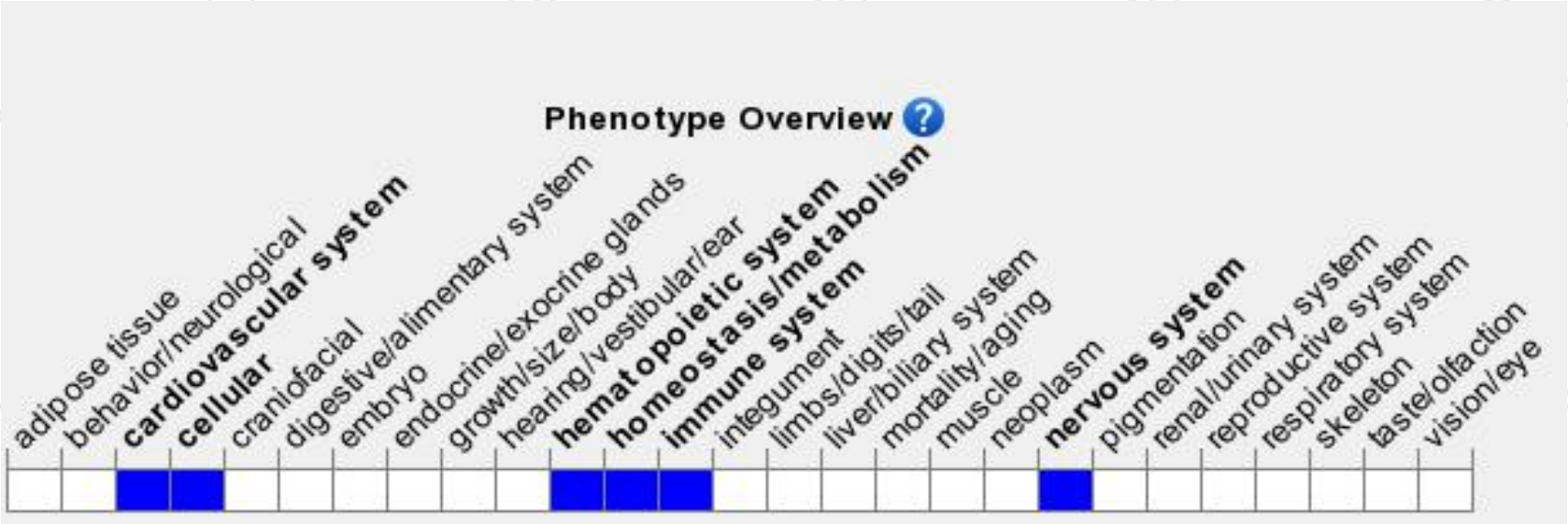
Genomic location distribution



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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