Myh9 Cas9-CKO Strategy

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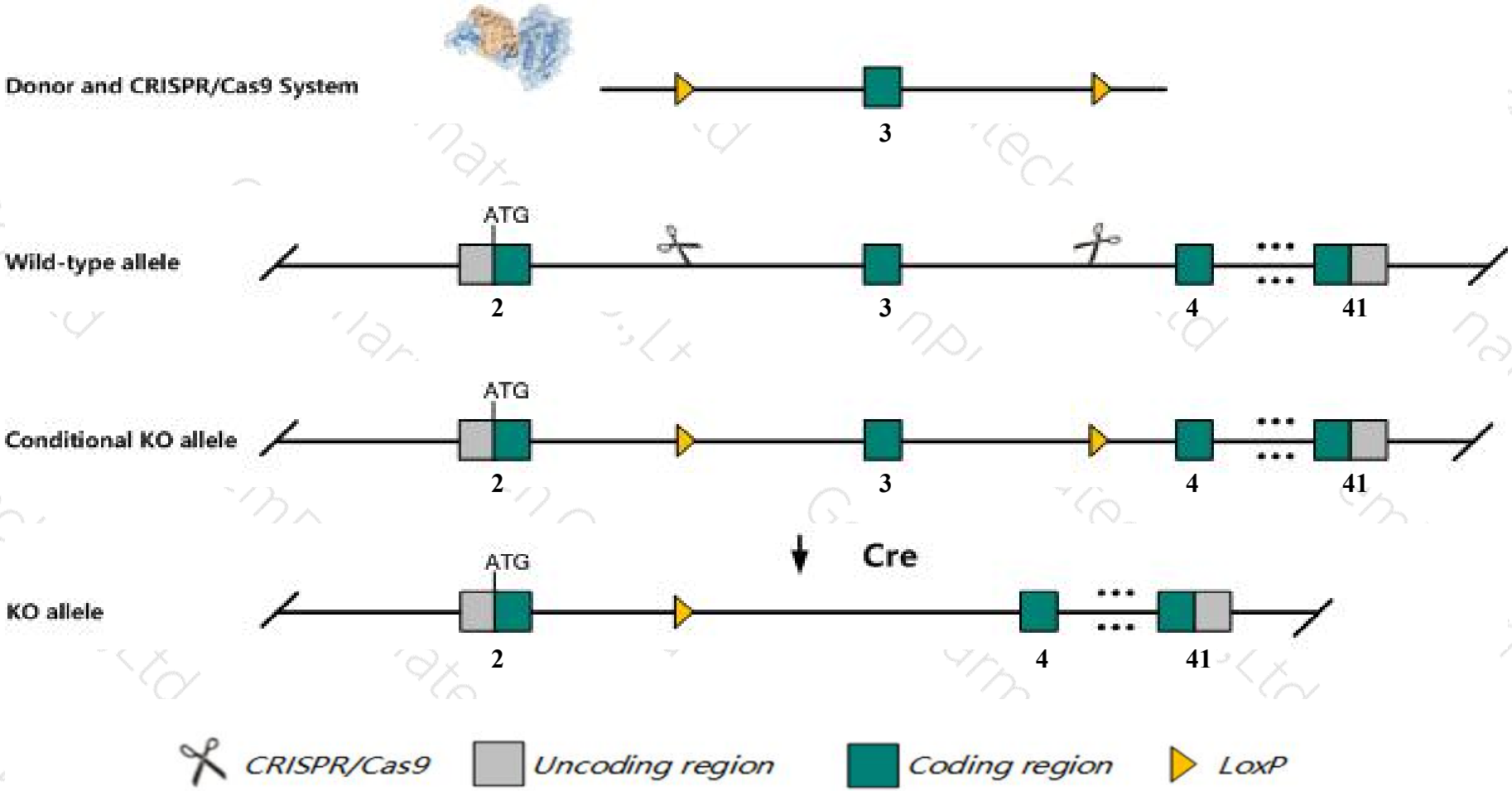
Project Overview

Project Name	***Myh9***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Myh9* gene. The schematic diagram is as follows:



Technical routes

- The *Myh9* gene has 9 transcripts. According to the structure of *Myh9* gene, exon3 of *Myh9-201* (ENSMUST00000016771.12) transcript is recommended as the knockout region.The region contains 157bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Myh9* 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,Homozygous null mice display embryonic lethality. Heterozygous null mice display hearing loss with incomplete penetrance. Mice homozygous or heterozygous for one of several knock-in alleles exhibit macrothrombocytopenia, nephritis, cataracts and deafness.

➢ The *Myh9* gene is located on the Chr15. 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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Myh9 myosin, heavy polypeptide 9, non-muscle [Mus musculus (house mouse)]

Gene ID: 17886, updated on 7-Apr-2019

Summary

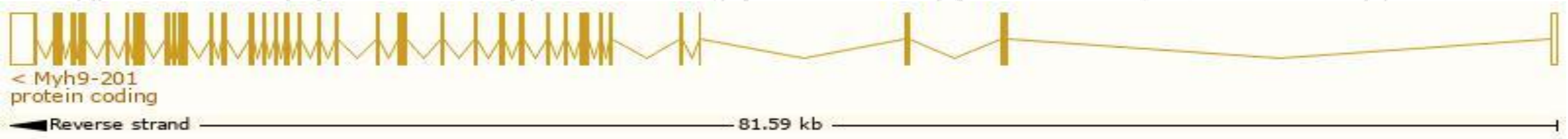
Official Symbol	Myh9 provided by MGI
Official Full Name	myosin, heavy polypeptide 9, non-muscle provided by MGI
Primary source	MGI:MGI:107717
See related	Ensembl:ENSMUSG00000022443
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Fltn, Myhn-1, Myhn1, NMHCIIA, NMMHC-A, NMMHC-IIA, TU72.6
Expression	Ubiquitous expression in lung adult (RPKM 64.5), spleen adult (RPKM 64.1) and 27 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

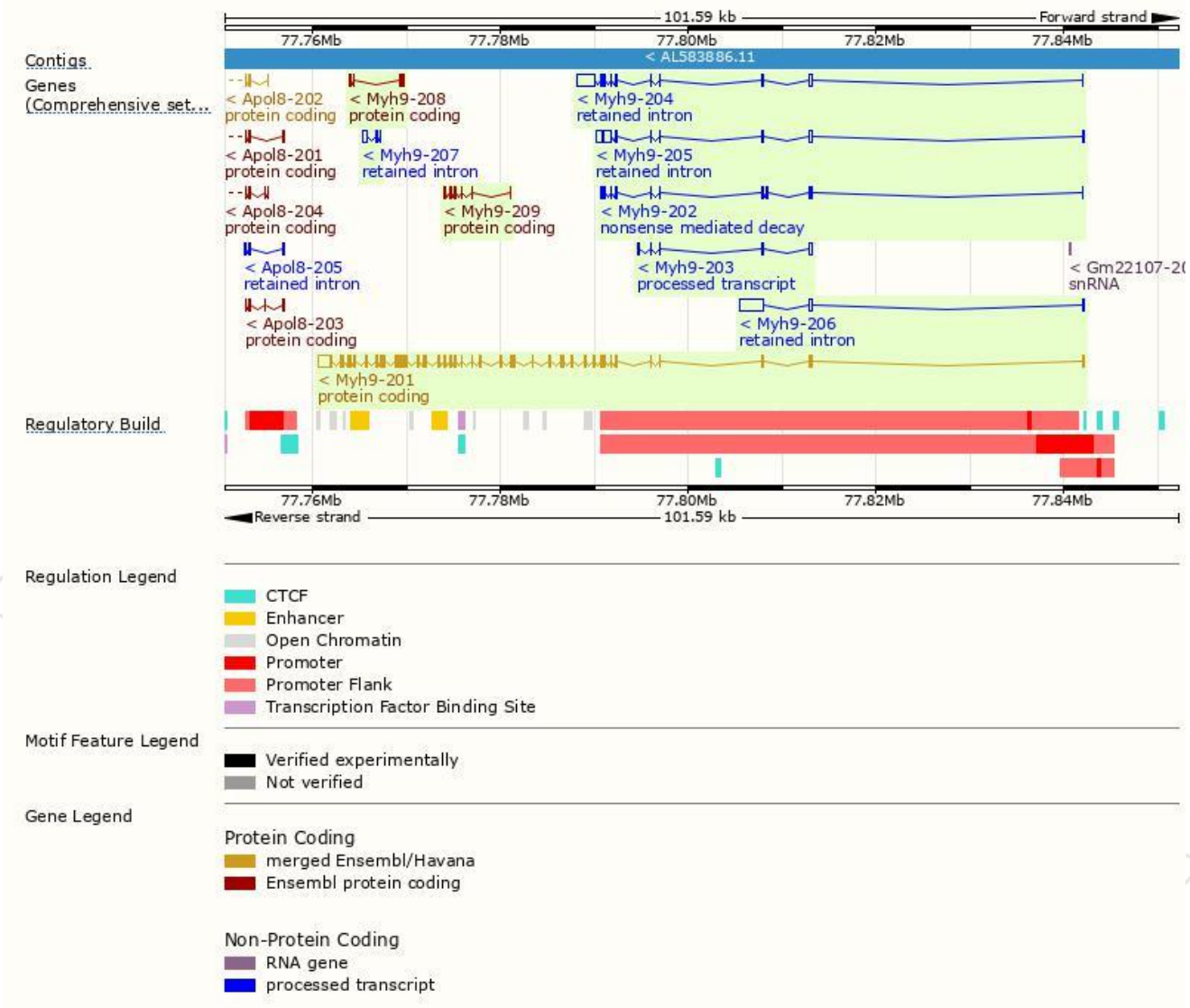
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Myh9-201	ENSMUST00000016771.12	7433	1960aa	Protein coding	CCDS27605	Q8VDD5	TSL:1 GENCODE basic APPRIS P1
Myh9-209	ENSMUST00000231192.1	633	211aa	Protein coding	-	A0A2R8VKI5	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Myh9-208	ENSMUST00000229259.1	329	110aa	Protein coding	-	A0A2R8W6V7	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Myh9-202	ENSMUST00000123101.7	1191	120aa	Nonsense mediated decay	-	F2Z494	TSL:5
Myh9-203	ENSMUST00000124844.7	720	No protein	Processed transcript	-	-	TSL:5
Myh9-206	ENSMUST00000134878.1	2980	No protein	Retained intron	-	-	TSL:1
Myh9-204	ENSMUST00000126796.7	2904	No protein	Retained intron	-	-	TSL:1
Myh9-205	ENSMUST00000129453.7	2228	No protein	Retained intron	-	-	TSL:2
Myh9-207	ENSMUST00000139729.1	723	No protein	Retained intron	-	-	TSL:3

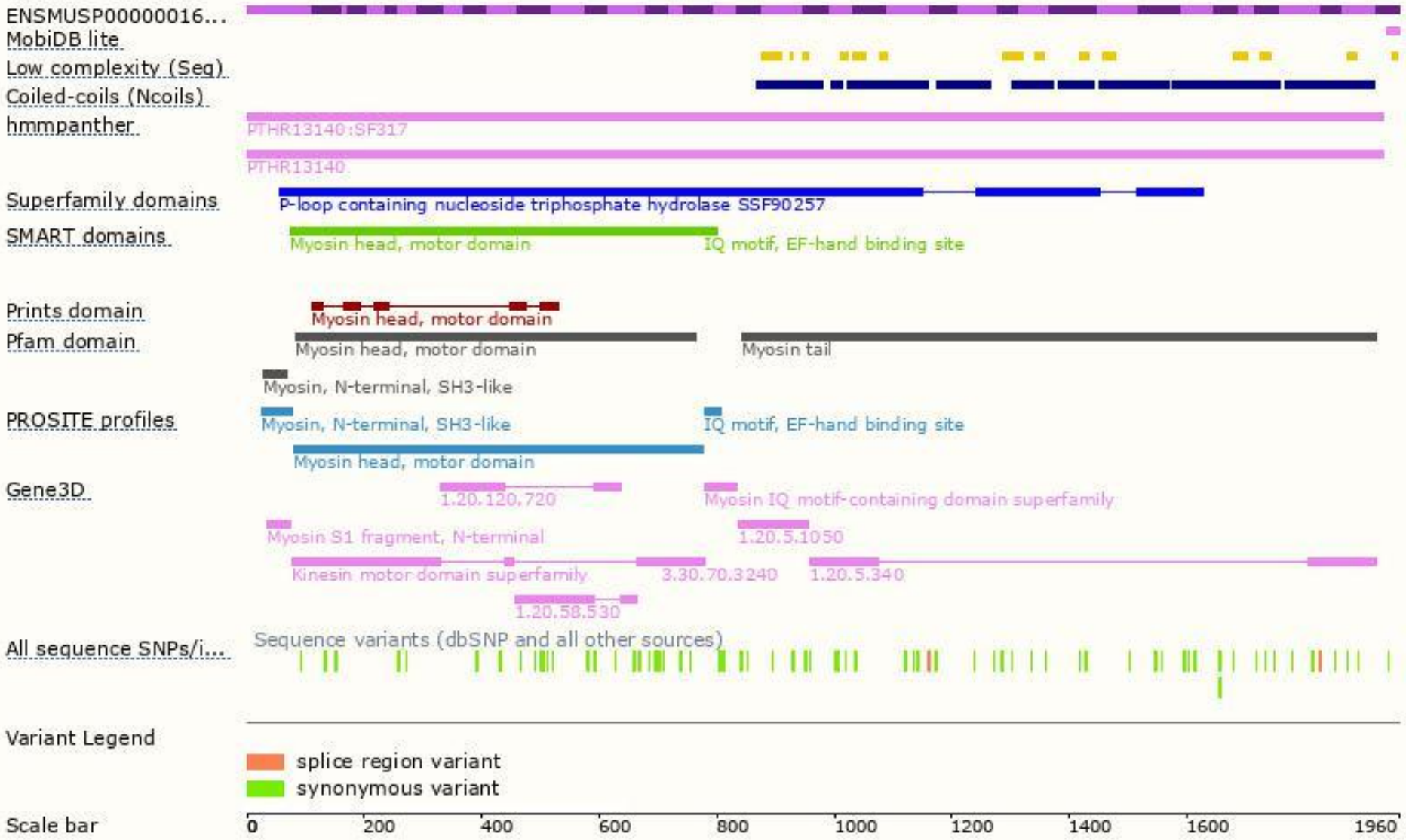
The strategy is based on the design of *Myh9-201* transcript,The transcription is shown below



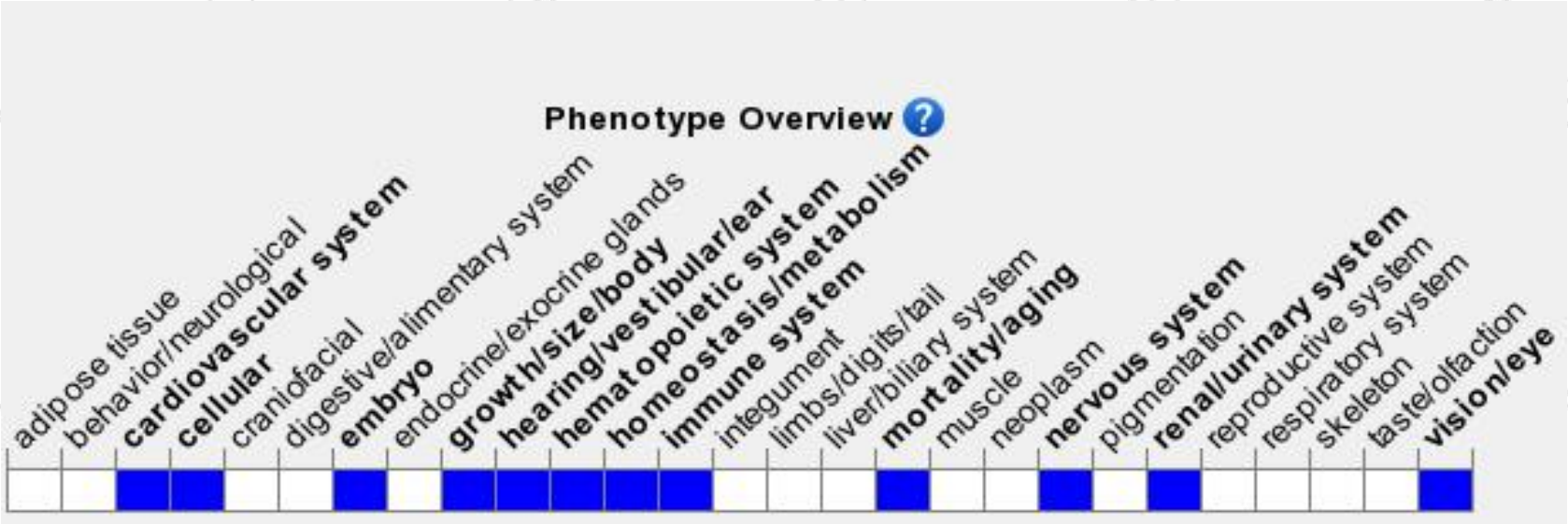
Genomic location distribution



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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