Nfe2l2 Cas9-CKO Strategy

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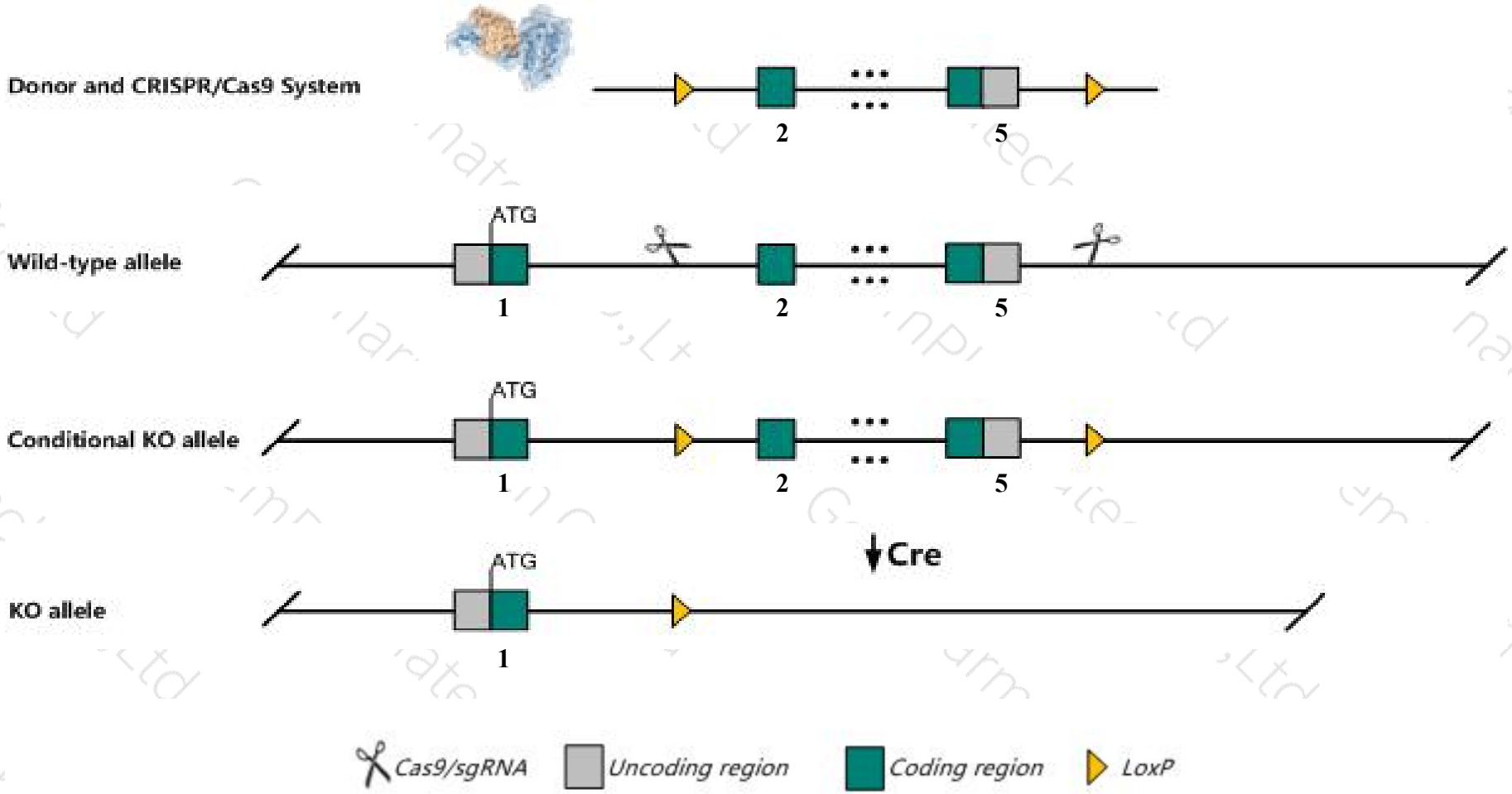
Project Overview

Project Name	***Nfe2l2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6J**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Nfe2l2* gene. The schematic diagram is as follows:



Technical routes

- The *Nfe2l2* gene has 2 transcripts. According to the structure of *Nfe2l2* gene, exon2-exon5 of *Nfe2l2-201* (ENSMUST00000102672.4) transcript is recommended as the knockout region.The region contains most of the coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Nfe2l2* 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/6J 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/6J 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.

Notice

- According to the existing MGI data,Mice homozygous for a knock-out allele exhibit increased sensitivity to oxidative stress in a variety of organs and cells including brain, liver, erythrocytes, and spleen, abnormal tooth enamel, and abnormal response to various injuries, chemical treatments, and induced inflammatory diseases.

- The *Nfe2l2* gene is located on the Chr2. 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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Nfe2l2 nuclear factor, erythroid derived 2, like 2 [Mus musculus (house mouse)]

Gene ID: 18024, updated on 9-Apr-2019

Summary

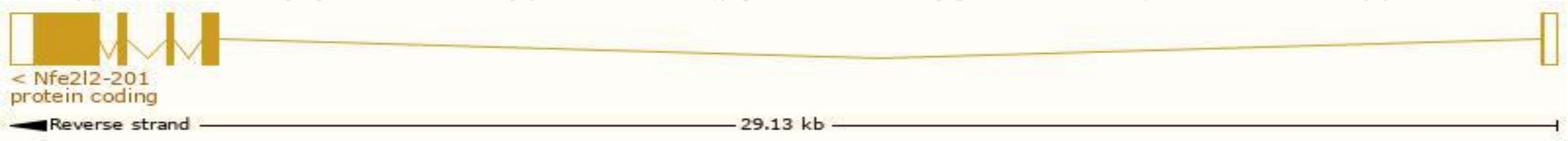
Official Symbol	Nfe2l2 provided by MGI
Official Full Name	nuclear factor, erythroid derived 2, like 2 provided by MGI
Primary source	MGI:MGI:108420
See related	Ensembl:ENSMUSG00000015839
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Nrf2
Summary	This gene encodes a transcription factor which is a member of a small family of basic leucine zipper (bZIP) proteins. The encoded transcription factor regulates genes which contain antioxidant response elements (ARE) in their promoters; many of these genes encode proteins involved in response to injury and inflammation which includes the production of free radicals. [provided by RefSeq, Sep 2015]
Expression	Ubiquitous expression in bladder adult (RPKM 57.1), duodenum adult (RPKM 26.8) and 26 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

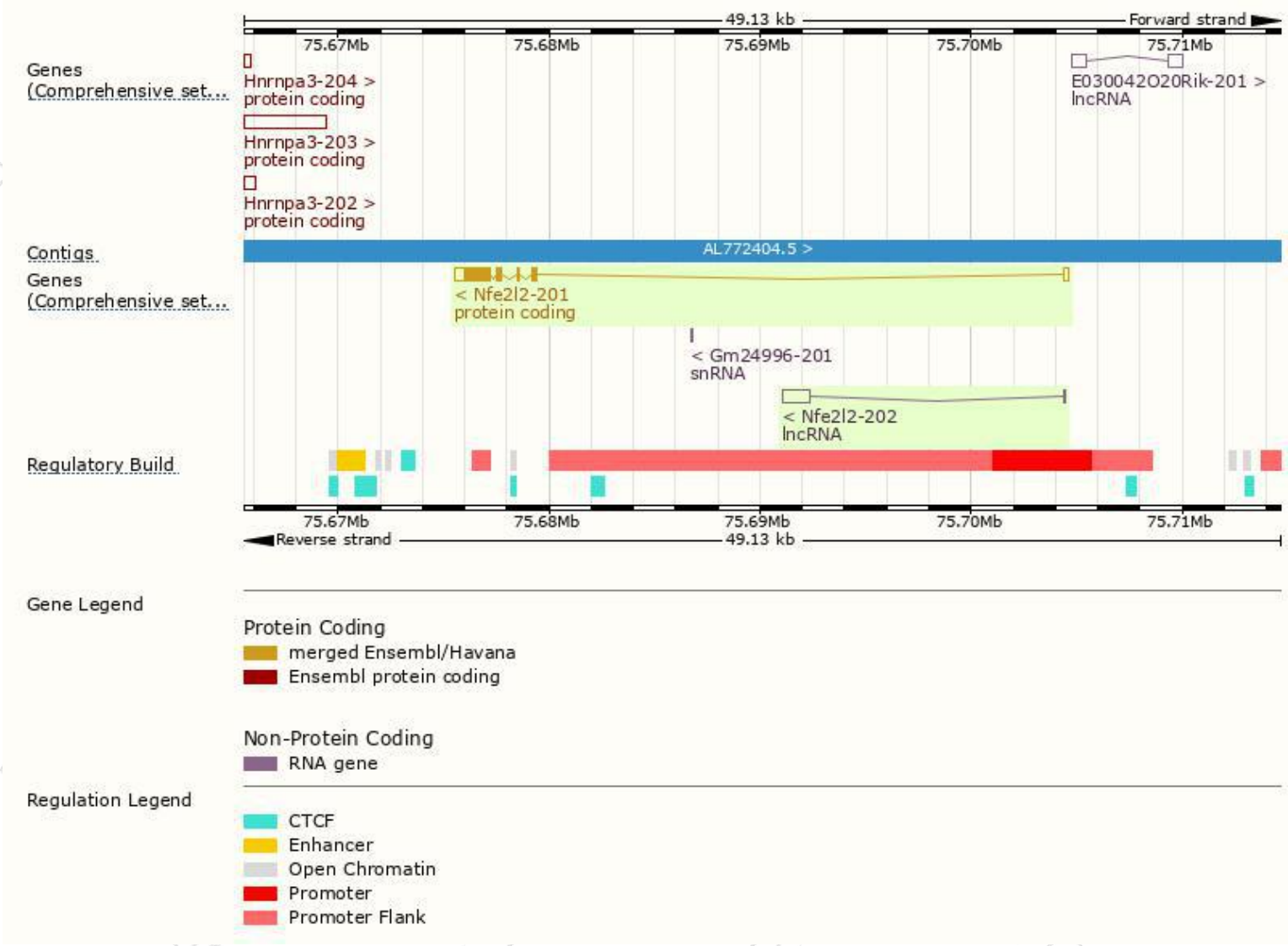
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nfe2l2-201	ENSMUST00000102672.4	2475	597aa	Protein coding	CCDS16150	Q60795	TSL:1 GENCODE basic APPRIS P1
Nfe2l2-202	ENSMUST00000152371.1	1447	No protein	Processed transcript	-	-	TSL:1

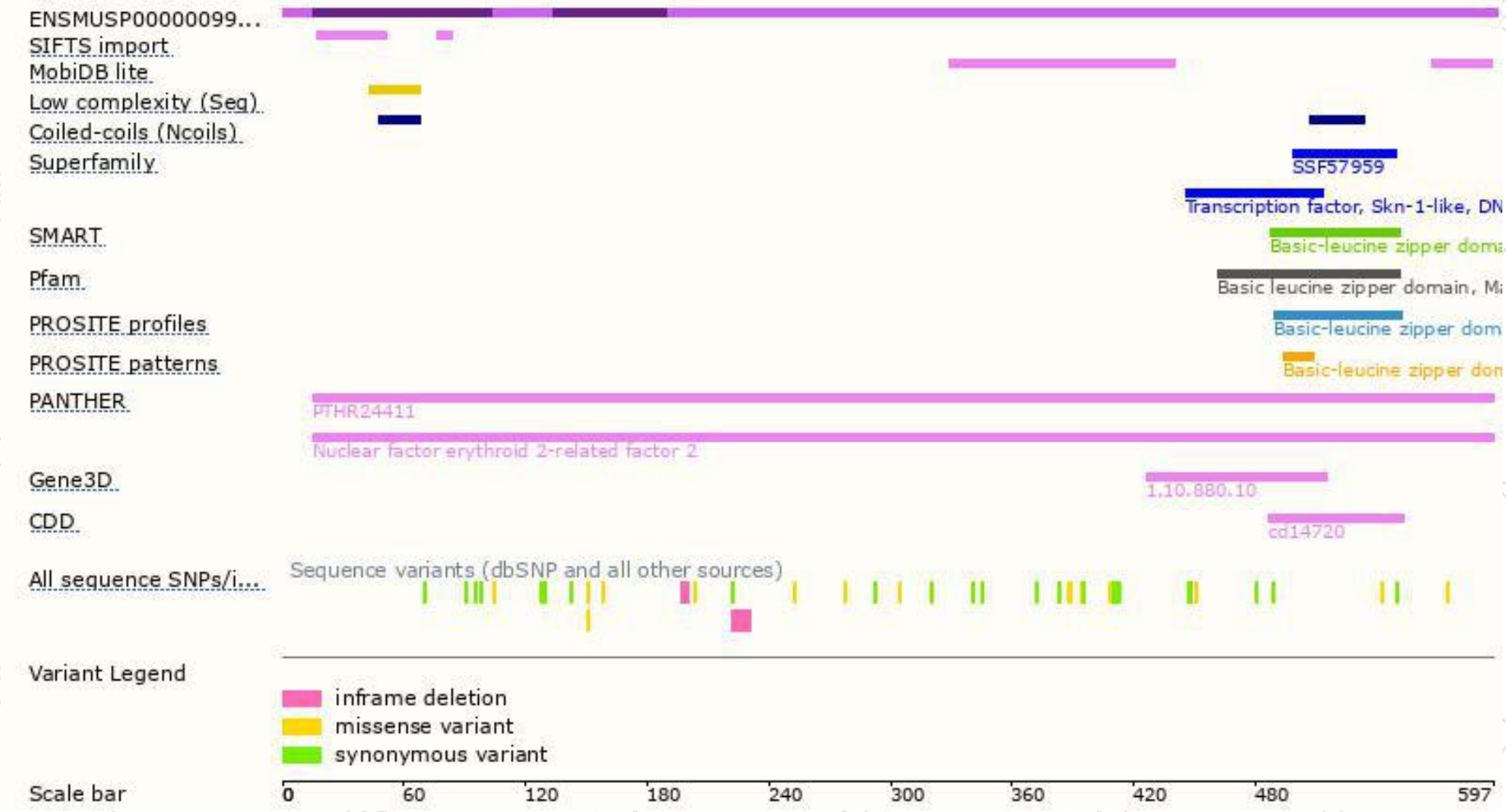
The strategy is based on the design of *Nfe2l2-201* transcript,The transcription is shown below



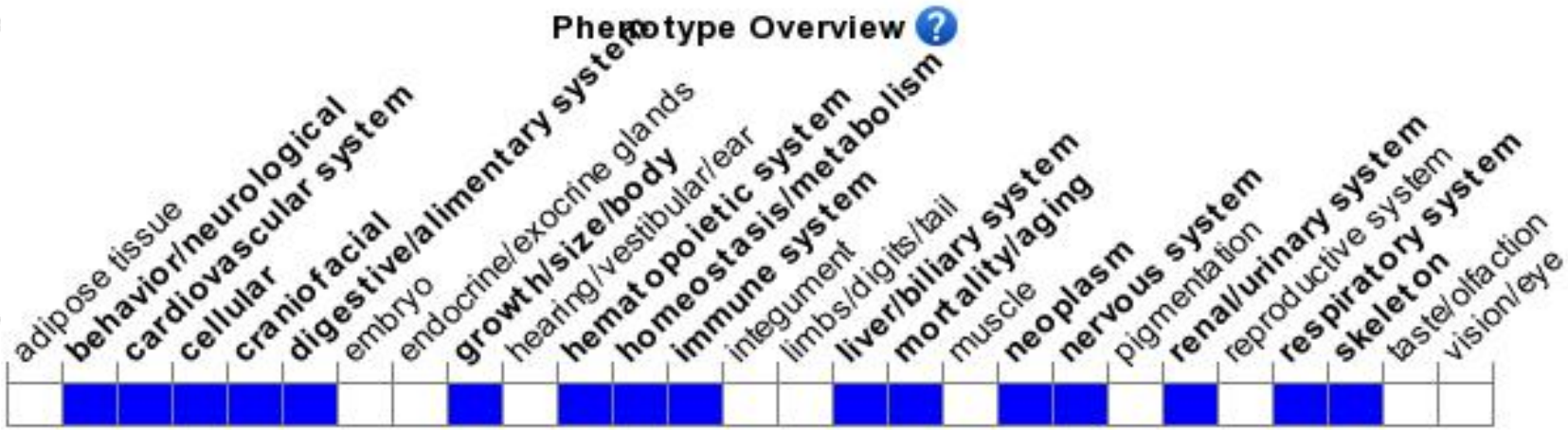
Genomic location distribution



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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