Wrn Cas9-CKO Strategy

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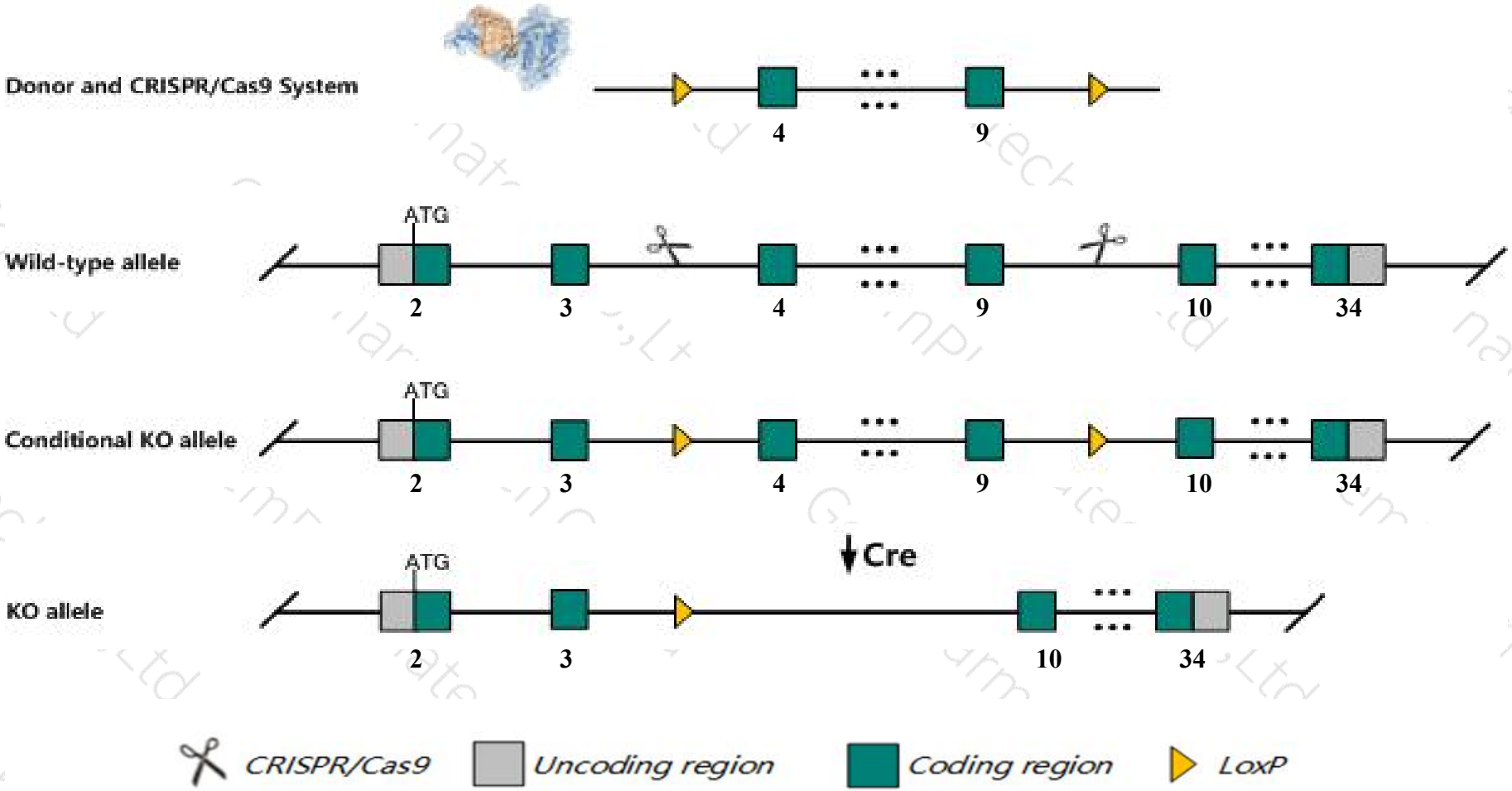
Project Overview

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

Conditional Knockout strategy

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



Technical routes

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- The *Wrn* gene has 4 transcripts. According to the structure of *Wrn* gene, exon4-exon9 of *Wrn-202* (ENSMUST00000033991.12) transcript is recommended as the knockout region.The region contains 1057bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Wrn* 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 mutants show enhanced frequency and variety of tumors in conjunction with Trp53 knockout alleles. Homozygotes also have an elevated frequency of somatic reversion of the pink-eyed dilution unstable mutation.

- The *Wrn* gene is located on the Chr8. 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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Wrn Werner syndrome RecQ like helicase [Mus musculus (house mouse)]

Gene ID: 22427, updated on 31-Jan-2019

Summary

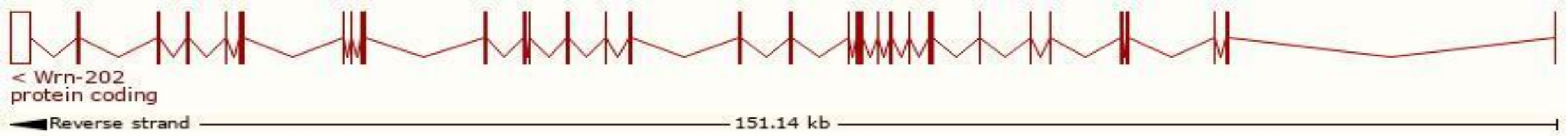
Official Symbol	Wrn provided by MGI
Official Full Name	Werner syndrome RecQ like helicase provided by MGI
Primary source	MGI:MGI:109635
See related	Ensembl:ENSMUSG00000031583
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI846146
Expression	Ubiquitous expression in liver E14 (RPKM 4.4), liver E14.5 (RPKM 3.9) and 25 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

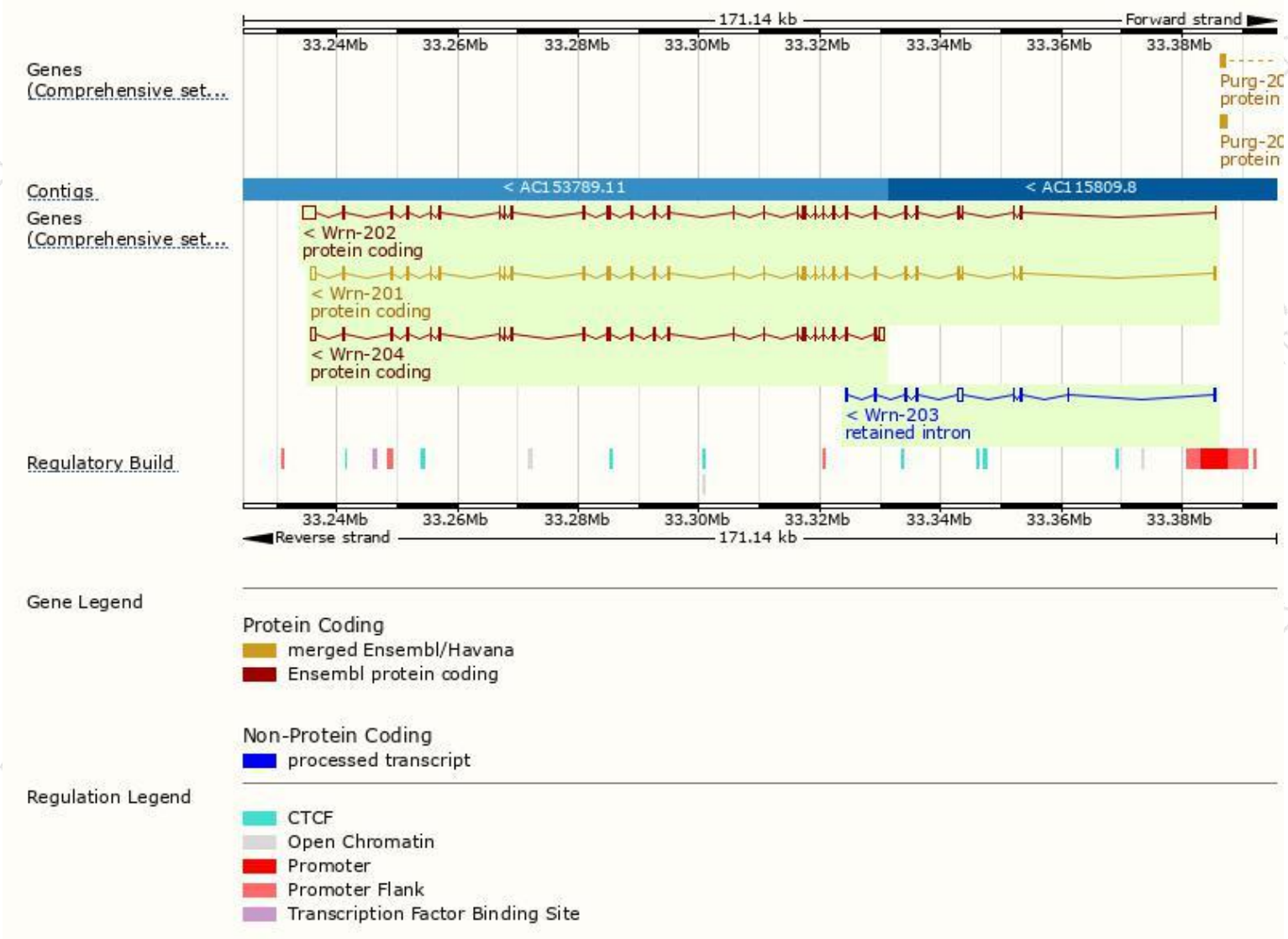
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Wrn-202	ENSMUST00000033991.12	6262	1401aa	Protein coding	CCDS22229	O09053	TSL:5 GENCODE basic APPRIS P1
Wrn-201	ENSMUST00000033990.6	5019	1401aa	Protein coding	CCDS22229	O09053	TSL:1 GENCODE basic APPRIS P1
Wrn-204	ENSMUST00000211498.1	4856	1158aa	Protein coding	-	A0A1B0GR54	TSL:1 GENCODE basic
Wrn-203	ENSMUST00000209293.1	1921	No protein	Retained intron	-	-	TSL:1

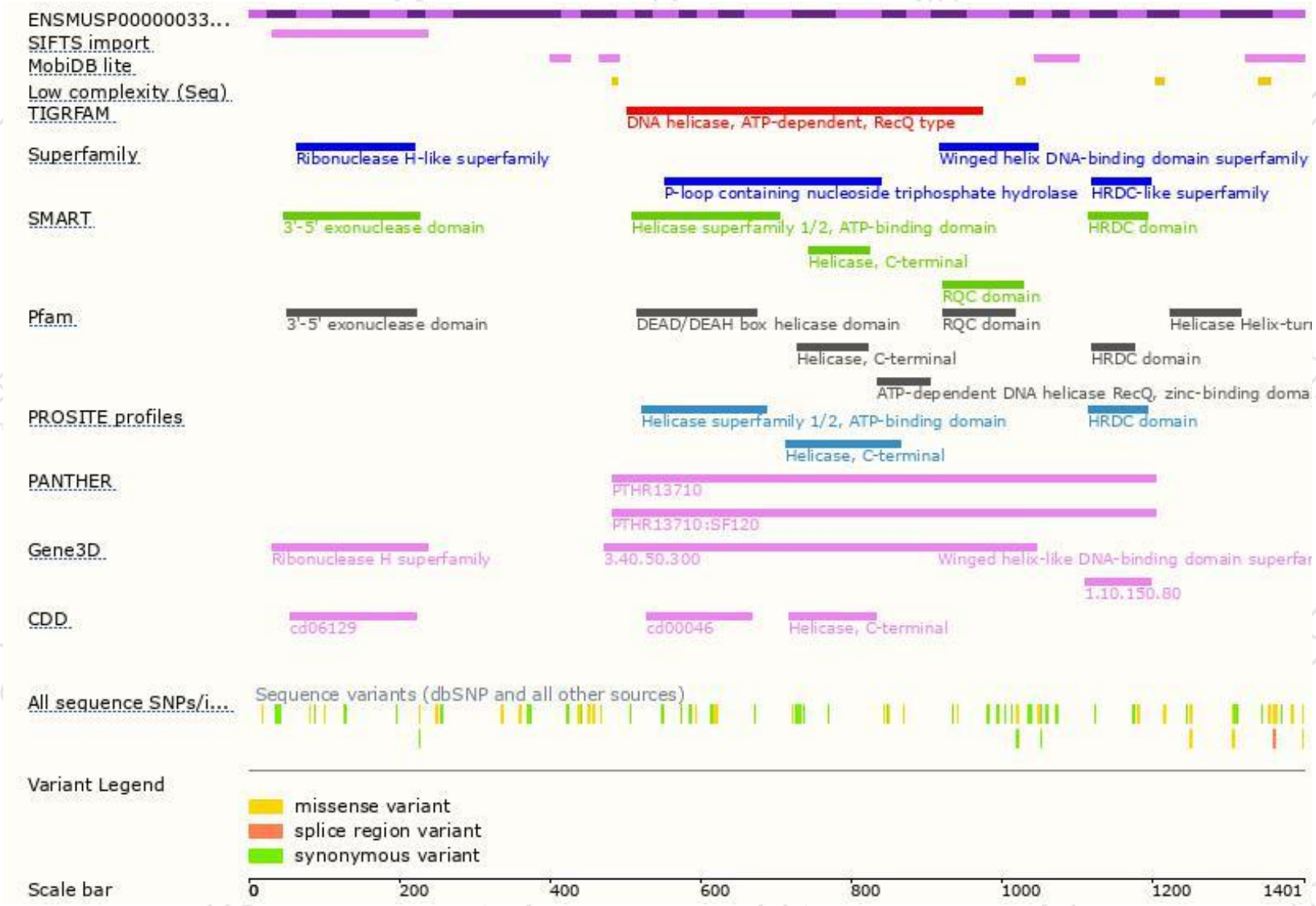
The strategy is based on the design of *Wrn-202* transcript,The transcription is shown below



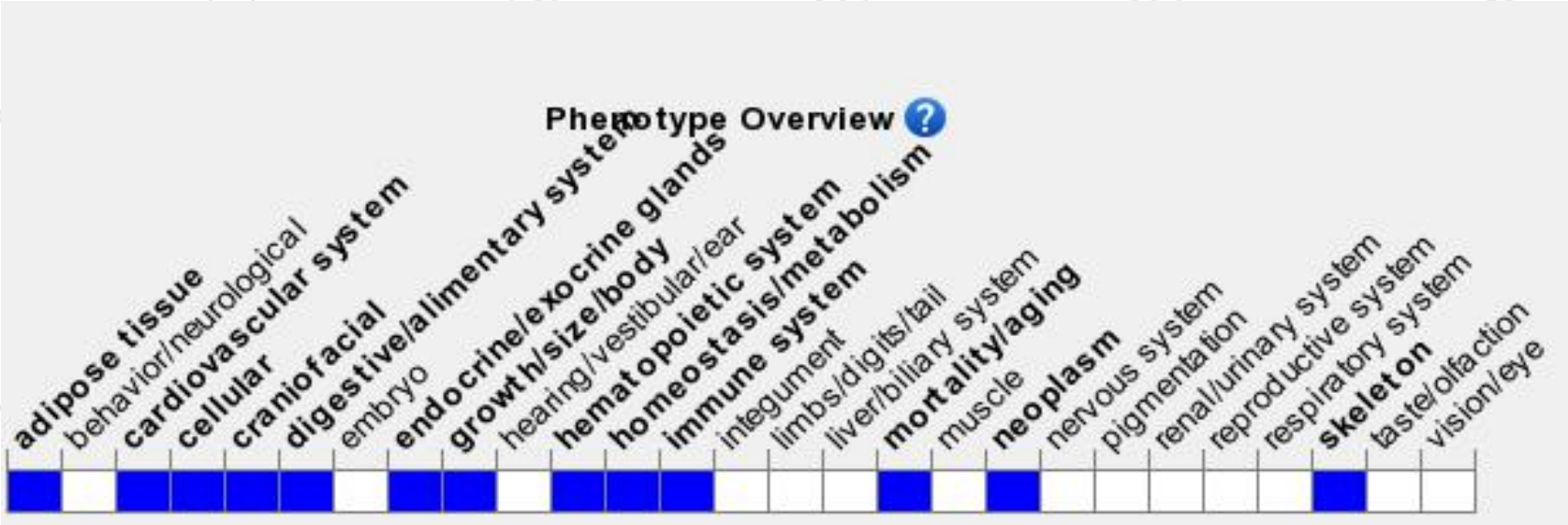
Genomic location distribution



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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