Chfr Cas9-CKO Strategy

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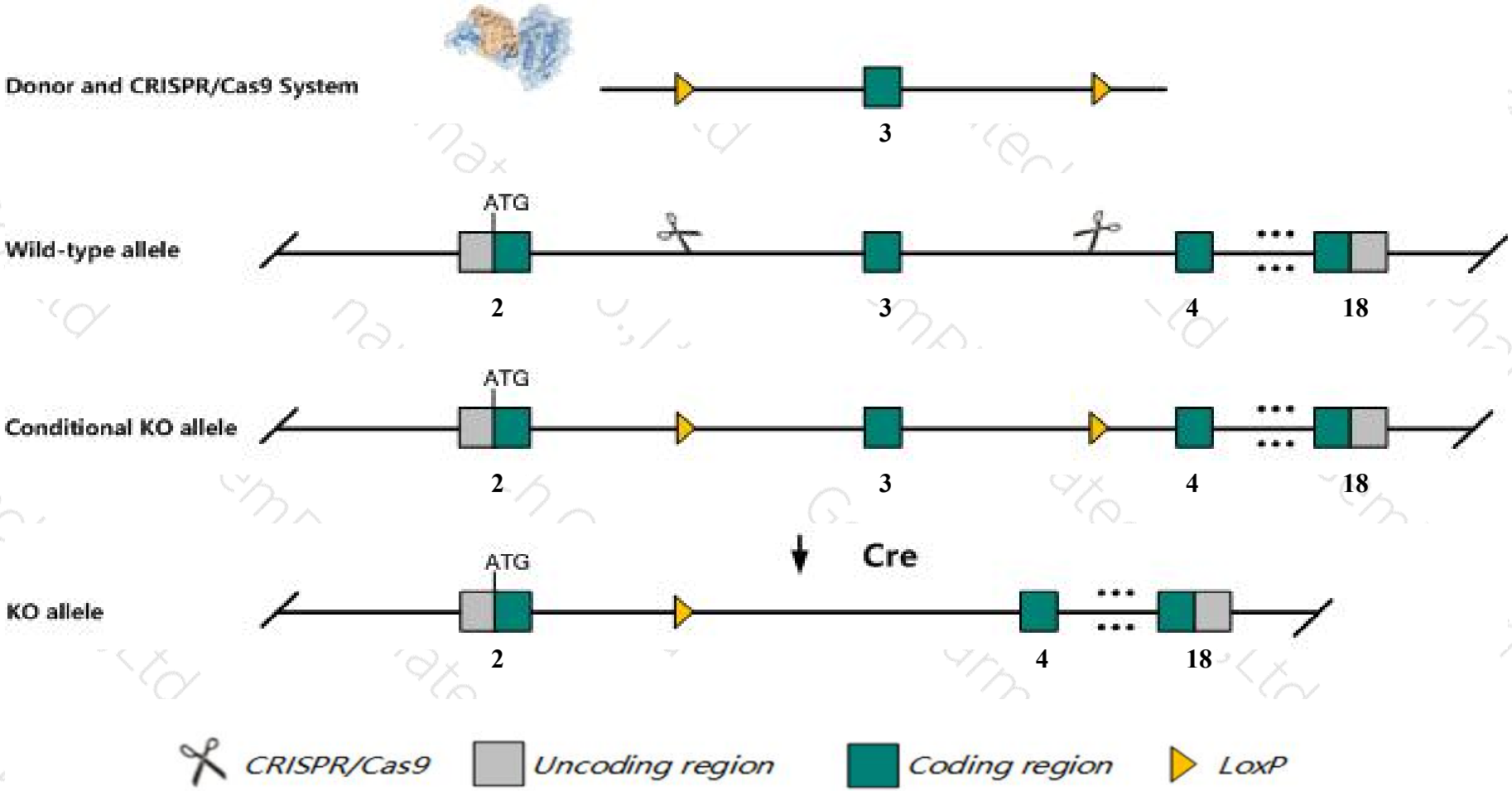
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Chfr* gene has 12 transcripts. According to the structure of *Chfr* gene, exon3 of *Chfr-202* (ENSMUST00000112519.8) transcript is recommended as the knockout region.The region contains 100bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Chfr* 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 and mefs display increased tumor incidence and inducibility, premature death, increased chromosomal instability, and cell cycle abnormalities.
- Transcripts Chfr-203,Chfr-205,Chfr-212 may not be affected.
- The *Chfr* gene is located on the Chr5. 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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Chfr checkpoint with forkhead and ring finger domains [Mus musculus (house mouse)]

Gene ID: 231600, updated on 13-Mar-2020

Summary

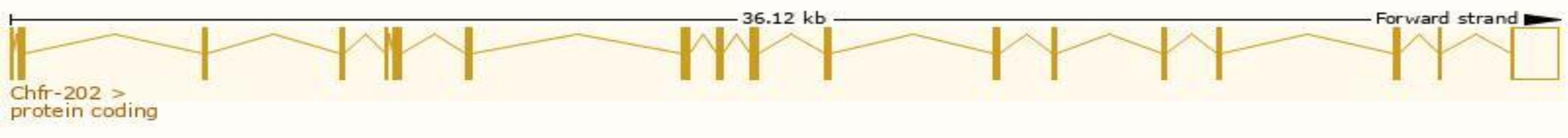
Official Symbol	Chfr provided by MGI
Official Full Name	checkpoint with forkhead and ring finger domains provided by MGI
Primary source	MGI:MGI:2444898
See related	Ensembl:ENSMUSG00000014668
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	5730484M20Rik, C230082M18, RNF116
Expression	Ubiquitous expression in thymus adult (RPKM 17.7), CNS E14 (RPKM 15.6) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

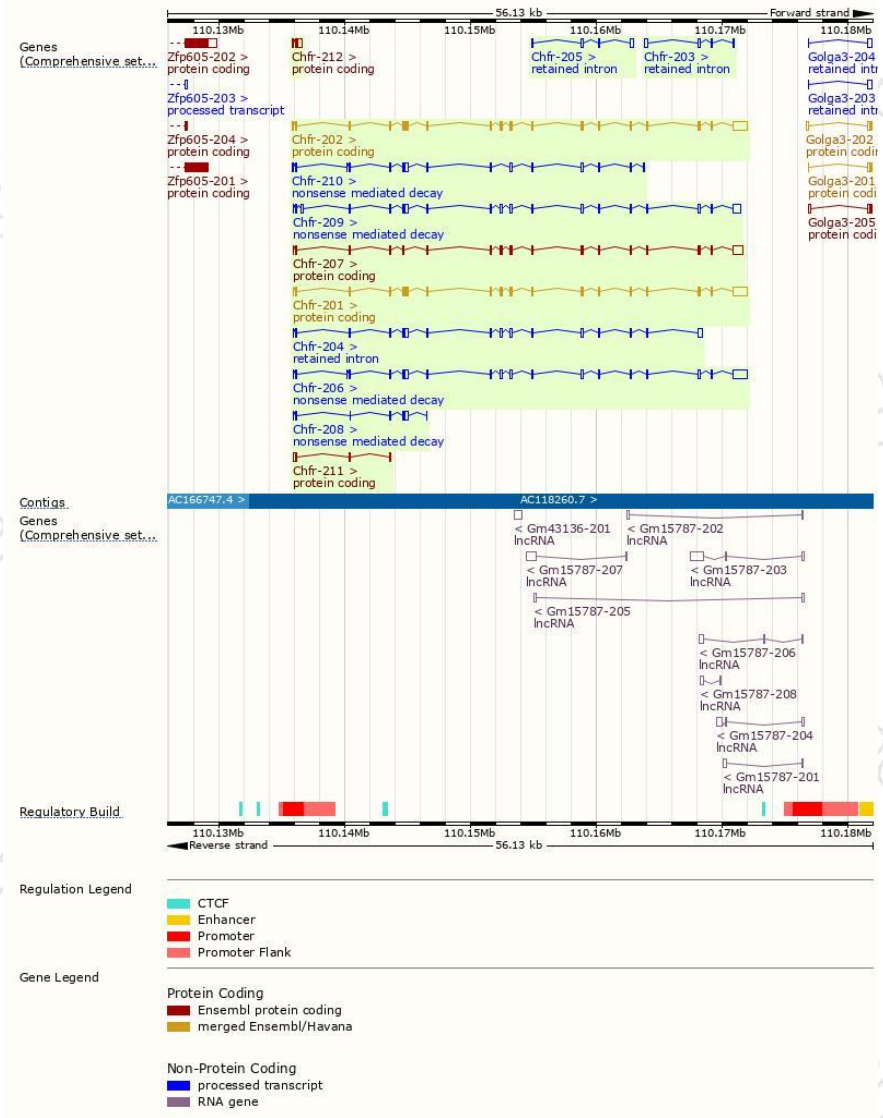
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Chfr-202	ENSMUST00000112519.8	3159	664aa	Protein coding	CCDS71637	Q810L3	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 ALT2
Chfr-201	ENSMUST00000014812.12	3146	663aa	Protein coding	CCDS19522	Q810L3	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 P3
Chfr-207	ENSMUST00000198633.4	2623	592aa	Protein coding	CCDS80358	Q810L3	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 ALT2
Chfr-212	ENSMUST00000200038.1	606	51aa	Protein coding	-	A0A0G2JDG0	TSL:1 GENCODE basic
Chfr-211	ENSMUST00000199811.2	480	114aa	Protein coding	-	A0A0G2JGX6	CDS 3' incomplete TSL:5
Chfr-206	ENSMUST00000198066.4	3201	49aa	Nonsense mediated decay	-	A0A0G2JFU4	TSL:1
Chfr-209	ENSMUST00000199557.4	2873	50aa	Nonsense mediated decay	-	A0A0G2JFC1	TSL:1
Chfr-210	ENSMUST00000199672.4	1774	49aa	Nonsense mediated decay	-	A0A0G2JFU4	TSL:1
Chfr-208	ENSMUST00000199283.1	714	64aa	Nonsense mediated decay	-	A0A0G2JG18	TSL:3
Chfr-204	ENSMUST00000197010.4	2135	No protein	Retained intron	-	-	TSL:2
Chfr-203	ENSMUST00000197005.1	613	No protein	Retained intron	-	-	TSL:2
Chfr-205	ENSMUST00000197968.1	597	No protein	Retained intron	-	-	TSL:2

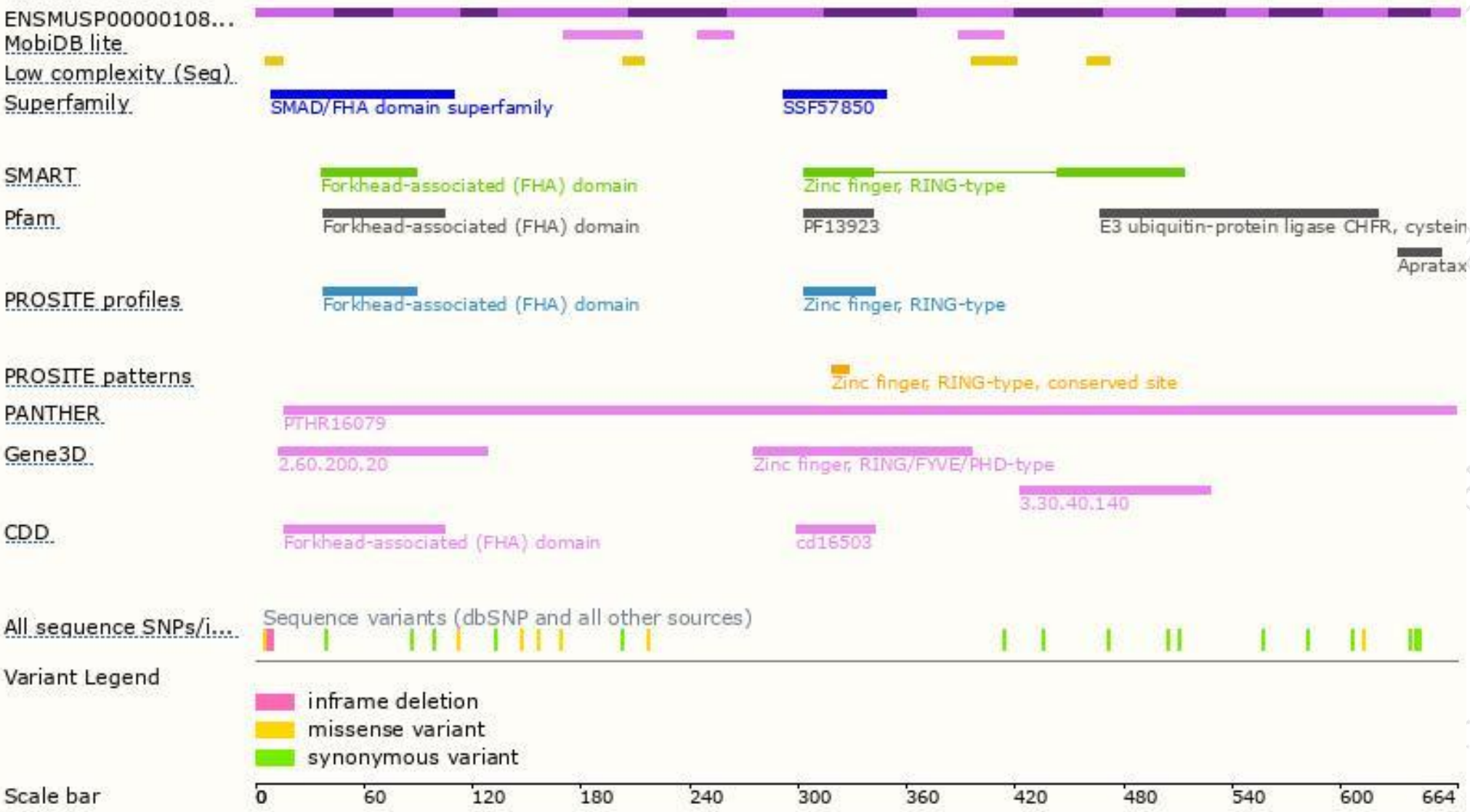
The strategy is based on the design of *Chfr-202* transcript,the transcription is shown below:



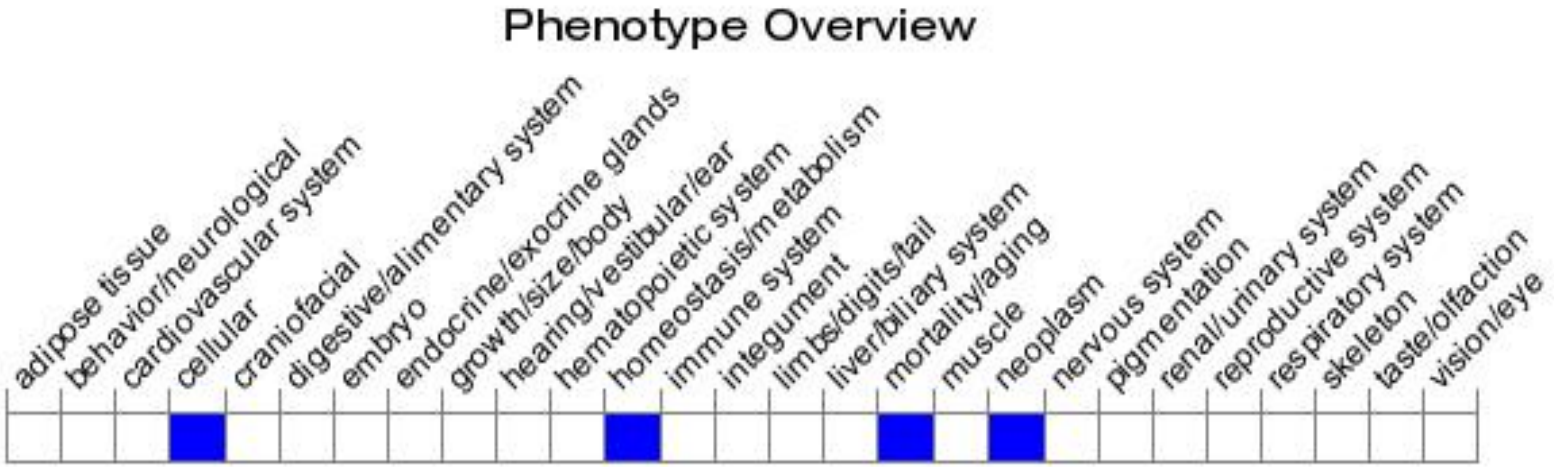
Genomic location distribution



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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