Sec24b Cas9-CKO Strategy

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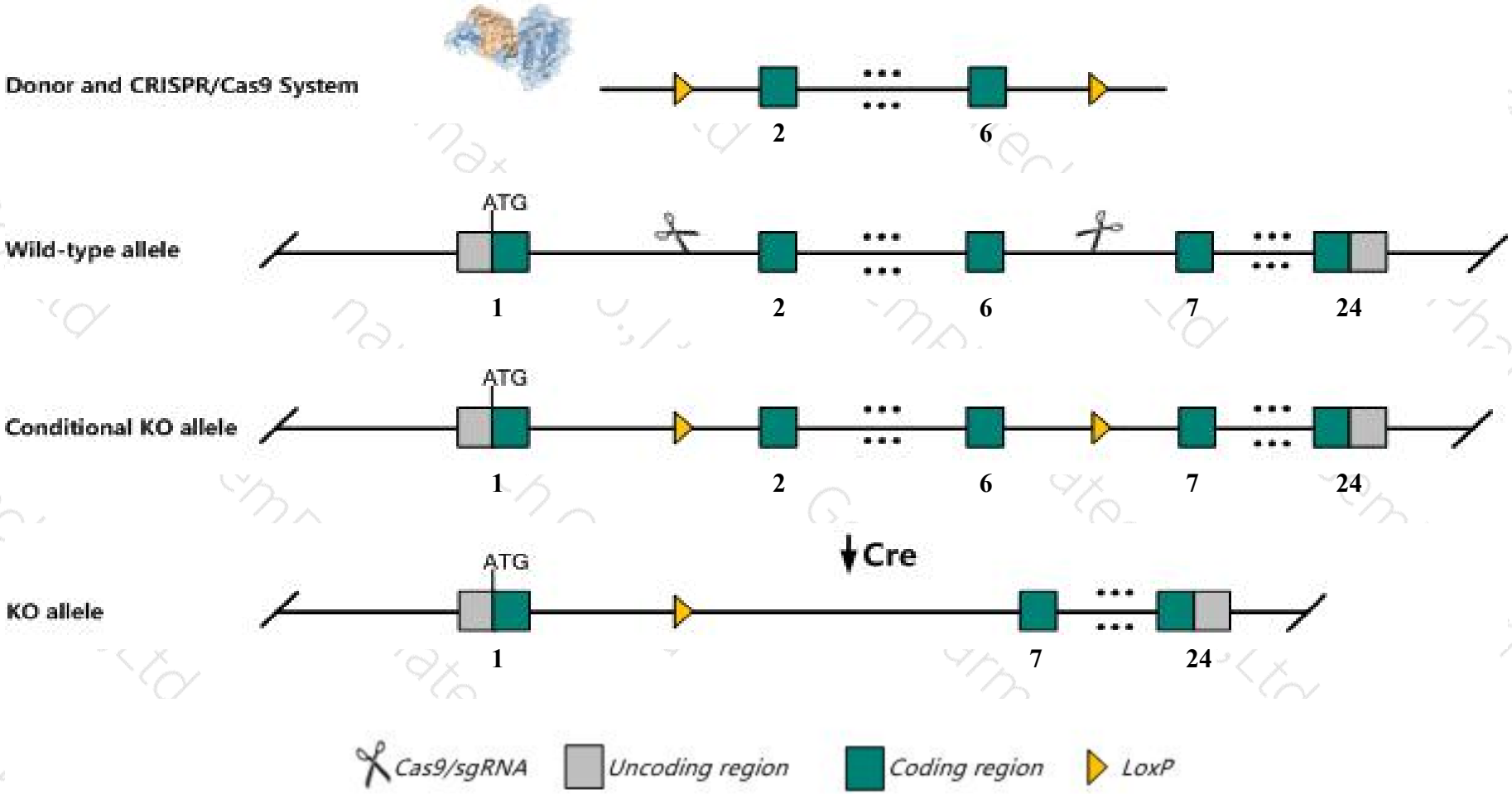
Project Overview

Project Name	***Sec24b***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sec24b* gene. The schematic diagram is as follows:



Technical routes

➢ The *Sec24b* gene has 9 transcripts. According to the structure of *Sec24b* gene, exon2-exon6 of *Sec24b-201*(ENSMUST00000001079.14) transcript is recommended as the knockout region.The region contains 1337bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Sec24b* 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/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 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 an ENU induced mutation exhibit craniorachischisis, abnormal embryo shape, omphalocele, disoriented hair cells, and failure of eyelid fusion.

➢ The *Sec24b* gene is located on the Chr3. 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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Sec24b Sec24 related gene family, member B (S. cerevisiae) [*Mus musculus* (house mouse)]

Gene ID: 99683, updated on 26-Jun-2020

Summary

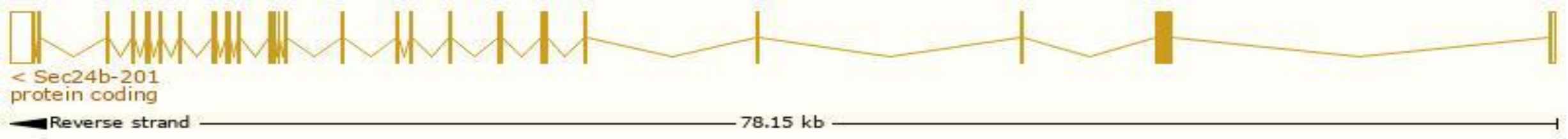
Official Symbol	Sec24b provided by MGI
Official Full Name	Sec24 related gene family, member B (S. cerevisiae) provided by MGI
Primary source	MGI:MGI:2139764
See related	Ensembl:ENSMUSG00000001052
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	SEC24; AI605202
Expression	Ubiquitous expression in testis adult (RPKM 37.4), thymus adult (RPKM 21.6) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

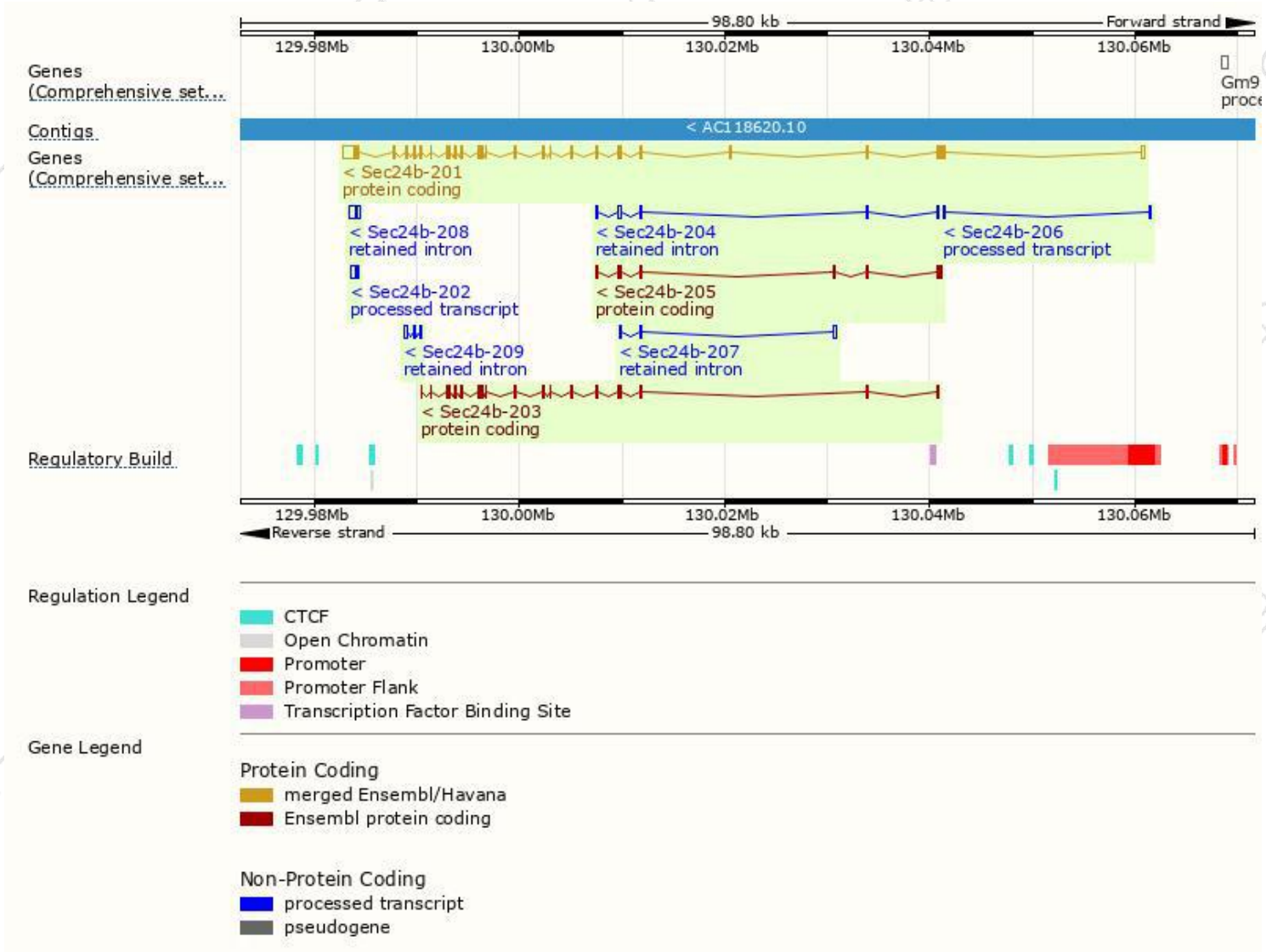
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sec24b-201	ENSMUST00000001079.14	5128	1251aa	Protein coding	CCDS17840	Q80ZX0	TSL:1 GENCODE basic APPRIS P1
Sec24b-203	ENSMUST00000165873.7	2295	765aa	Protein coding	-	F6VJC5	CDS 5' and 3' incomplete TSL:1
Sec24b-205	ENSMUST00000168644.2	1235	411aa	Protein coding	-	F6YIN5	CDS 5' and 3' incomplete TSL:5
Sec24b-202	ENSMUST00000164758.1	482	No protein	Processed transcript	-	-	TSL:5
Sec24b-206	ENSMUST00000168675.1	306	No protein	Processed transcript	-	-	TSL:5
Sec24b-208	ENSMUST00000170163.1	918	No protein	Retained intron	-	-	TSL:2
Sec24b-204	ENSMUST00000165889.7	829	No protein	Retained intron	-	-	TSL:5
Sec24b-209	ENSMUST00000172324.1	585	No protein	Retained intron	-	-	TSL:2
Sec24b-207	ENSMUST00000168908.1	527	No protein	Retained intron	-	-	TSL:3

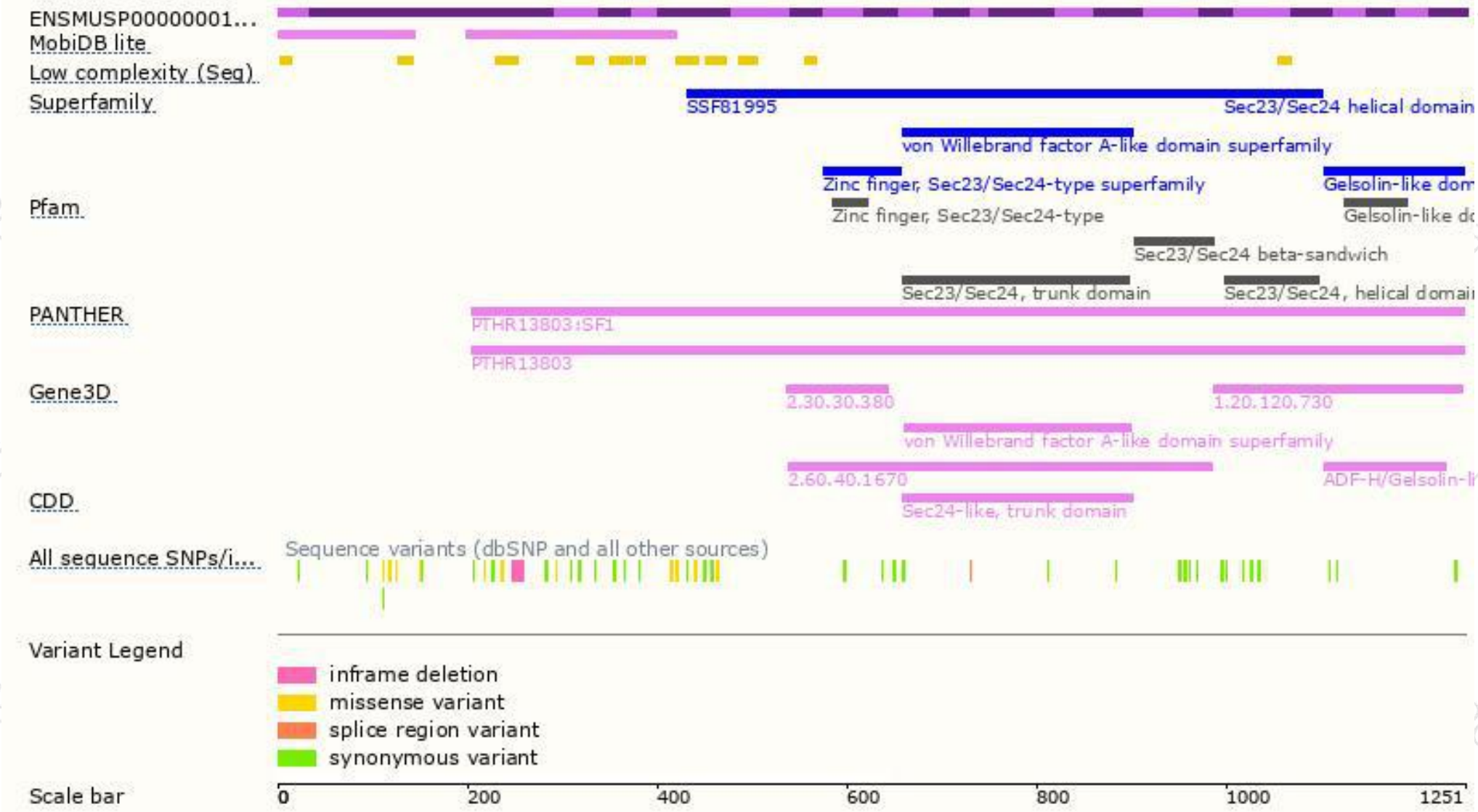
The strategy is based on the design of *Sec24b-201* transcript,the transcription is shown below:



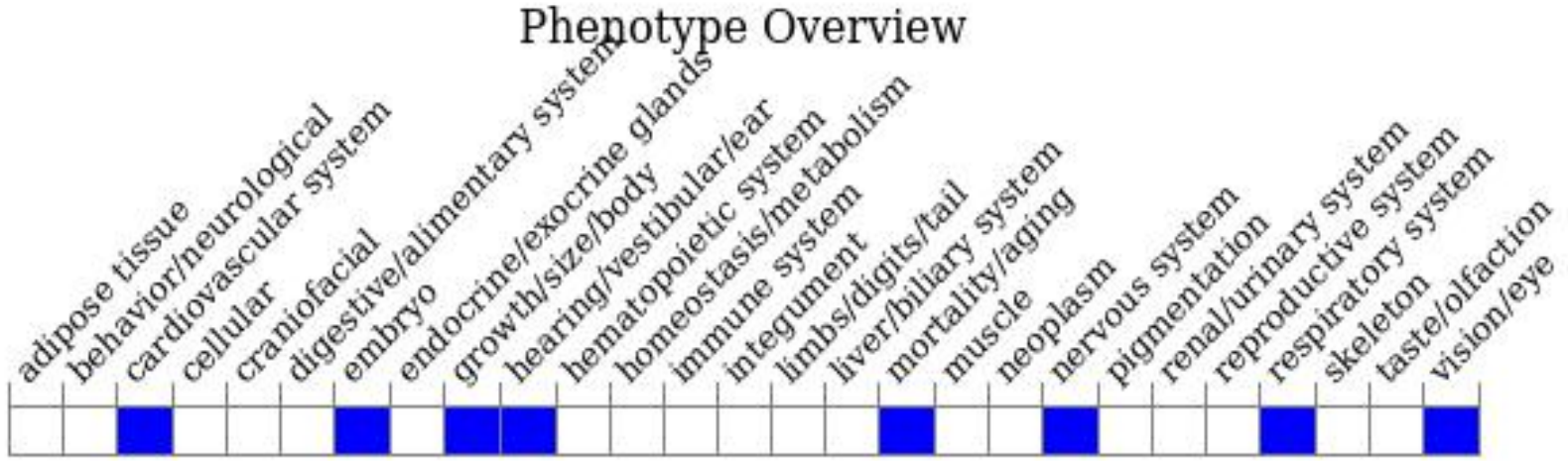
Genomic location distribution



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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