Mctp1 Cas9-CKO Strategy

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Project Overview

Project Name	***Mctp1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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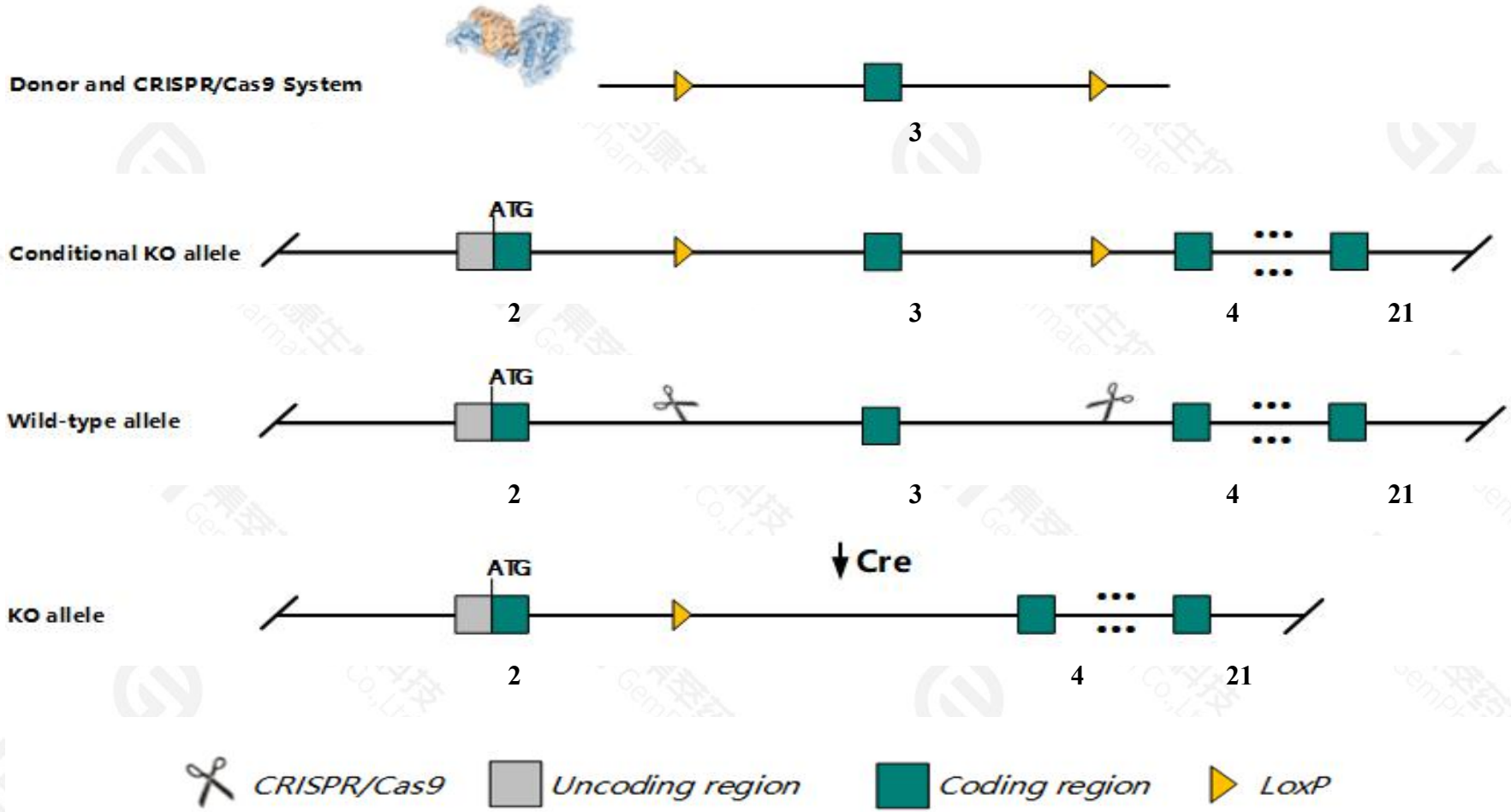
This model will use CRISPR/Cas9 technology to edit the *Mctp1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Mctp1* gene has 8 transcripts. According to the structure of *Mctp1* gene, exon3 of *Mctp1-201*(ENSMUST00000109583.9) transcript is recommended as the knockout region.The region contains 143bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Mctp1* 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 have normal hearing and inner ear structures. Mice homozygous for a spontaneous deletion that encompasses a cis-regulatory region crucial for Nr2f1 expression show circling, fused saccule and utricle, hearing loss, inner ear dysmorphology and disorganized cochlear hair cells.

➢ The *Mctp1* gene is located on the Chr13. 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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Mctp1 multiple C2 domains, transmembrane 1 [Mus musculus (house mouse)]

Gene ID: 78771, updated on 17-Feb-2021

Summary

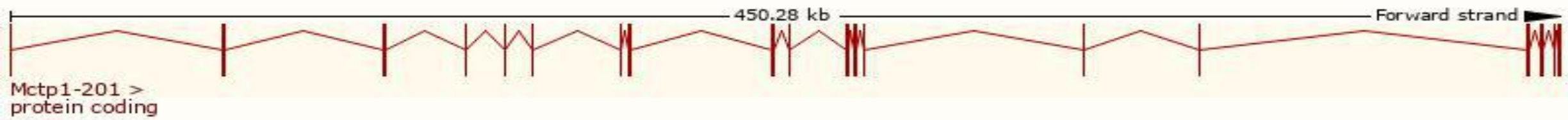
Official Symbol	Mctp1 provided by MGI
Official Full Name	multiple C2 domains, transmembrane 1 provided by MGI
Primary source	MGI:MGI:1926021
See related	Ensembl:ENSMUSG00000021596
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2810465F10Rik
Expression	Biased expression in cerebellum adult (RPKM 5.3), ovary adult (RPKM 4.5) and 9 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

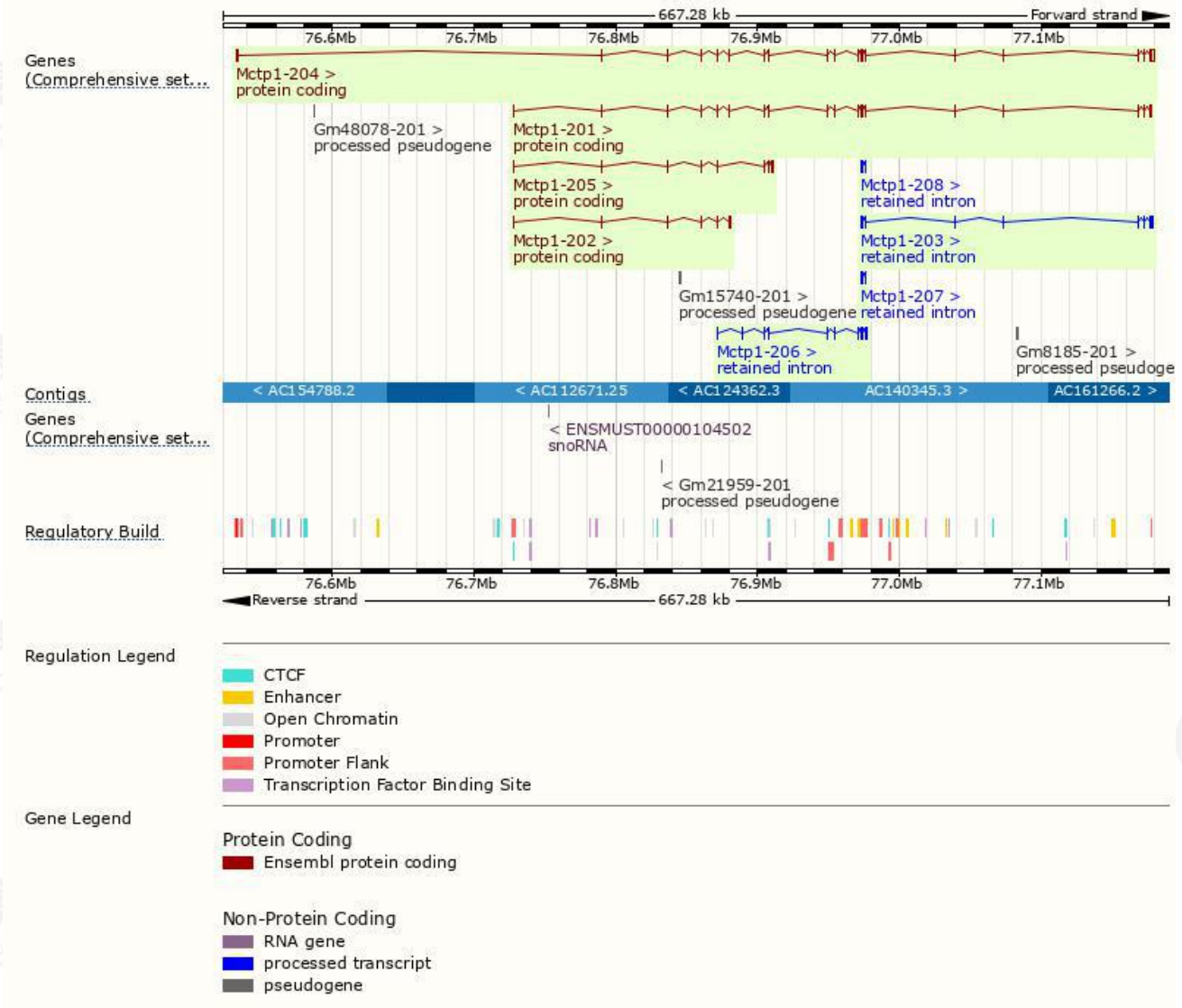
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mctp1-201	ENSMUST00000109583.9	2283	694aa	Protein coding	CCDS26656		TSL:5 , GENCODE basic , APPRIS P2 ,
Mctp1-204	ENSMUST00000125209.8	4715	951aa	Protein coding	-		TSL:1 , GENCODE basic , APPRIS ALT2 ,
Mctp1-205	ENSMUST00000126960.8	1780	319aa	Protein coding	-		TSL:1 , GENCODE basic ,
Mctp1-202	ENSMUST00000109589.3	1653	187aa	Protein coding	-		TSL:1 , GENCODE basic ,
Mctp1-206	ENSMUST00000137052.8	2278	No protein	Retained intron	-		TSL:1 ,
Mctp1-203	ENSMUST00000122843.2	1830	No protein	Retained intron	-		TSL:1 ,
Mctp1-207	ENSMUST00000149028.2	917	No protein	Retained intron	-		TSL:3 ,
Mctp1-208	ENSMUST00000155275.2	571	No protein	Retained intron	-		TSL:3 ,

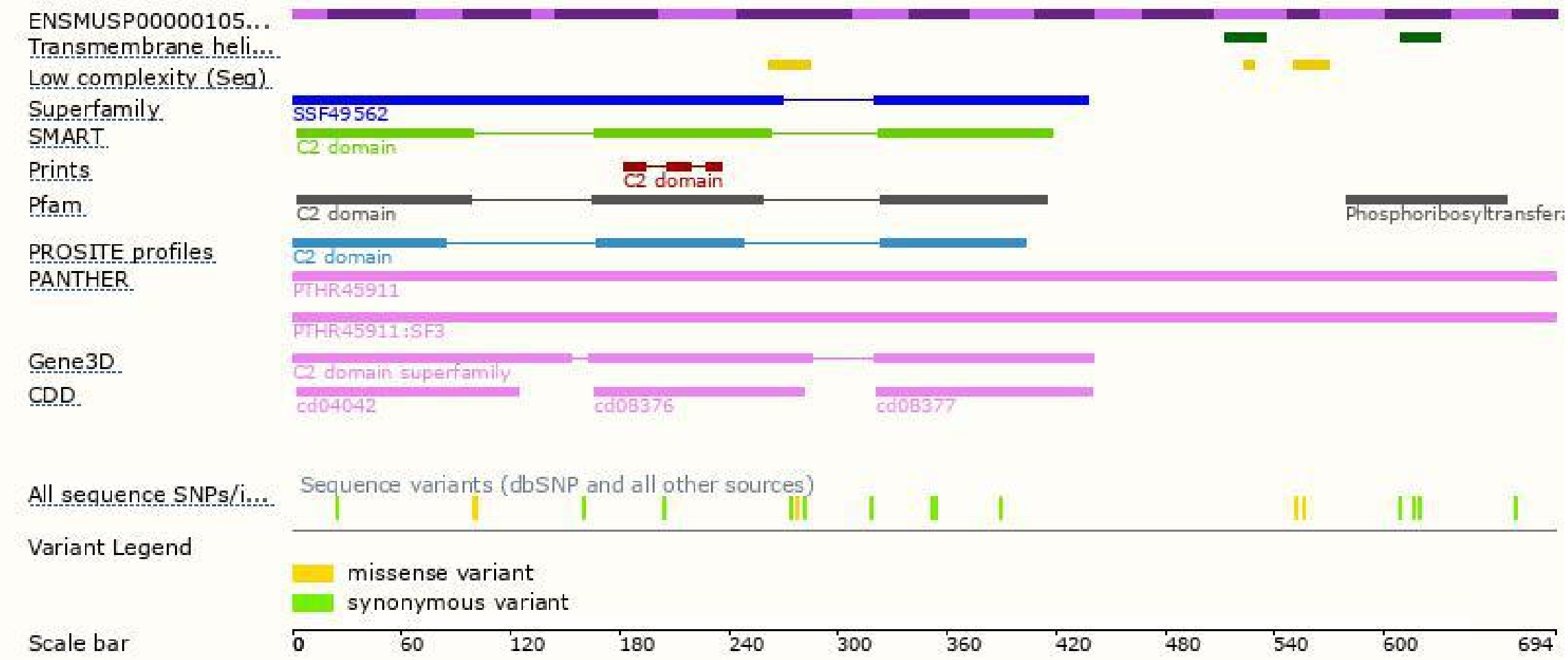
The strategy is based on the design of *Mctp1-201* transcript,the transcription is shown below:



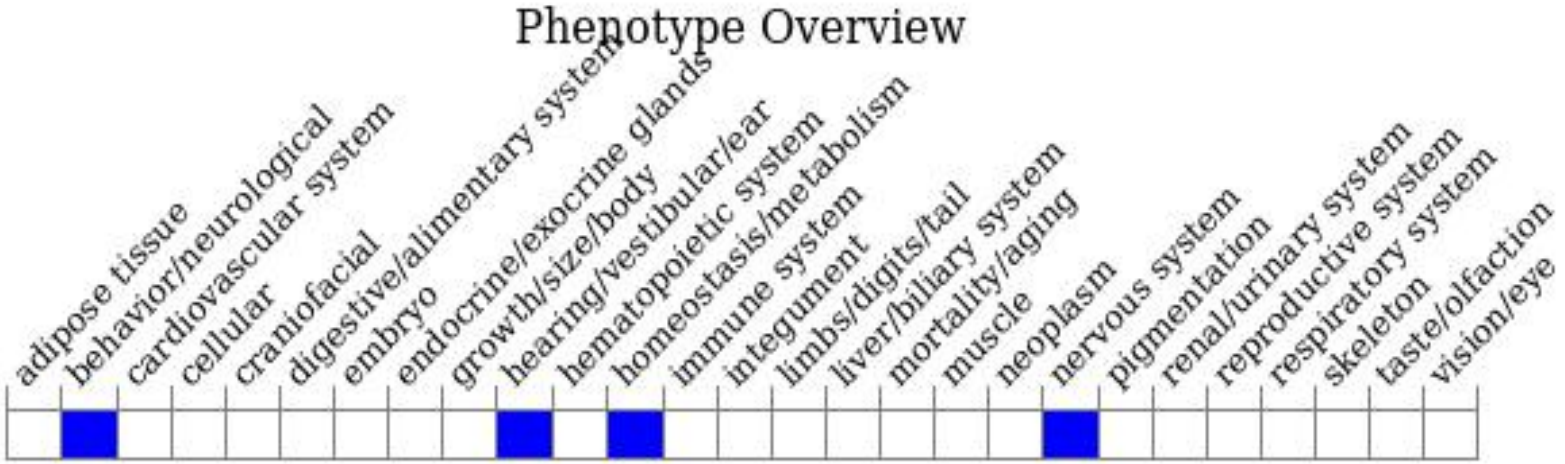
Genomic location distribution



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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