Slc35a1 Cas9-CKO Strategy

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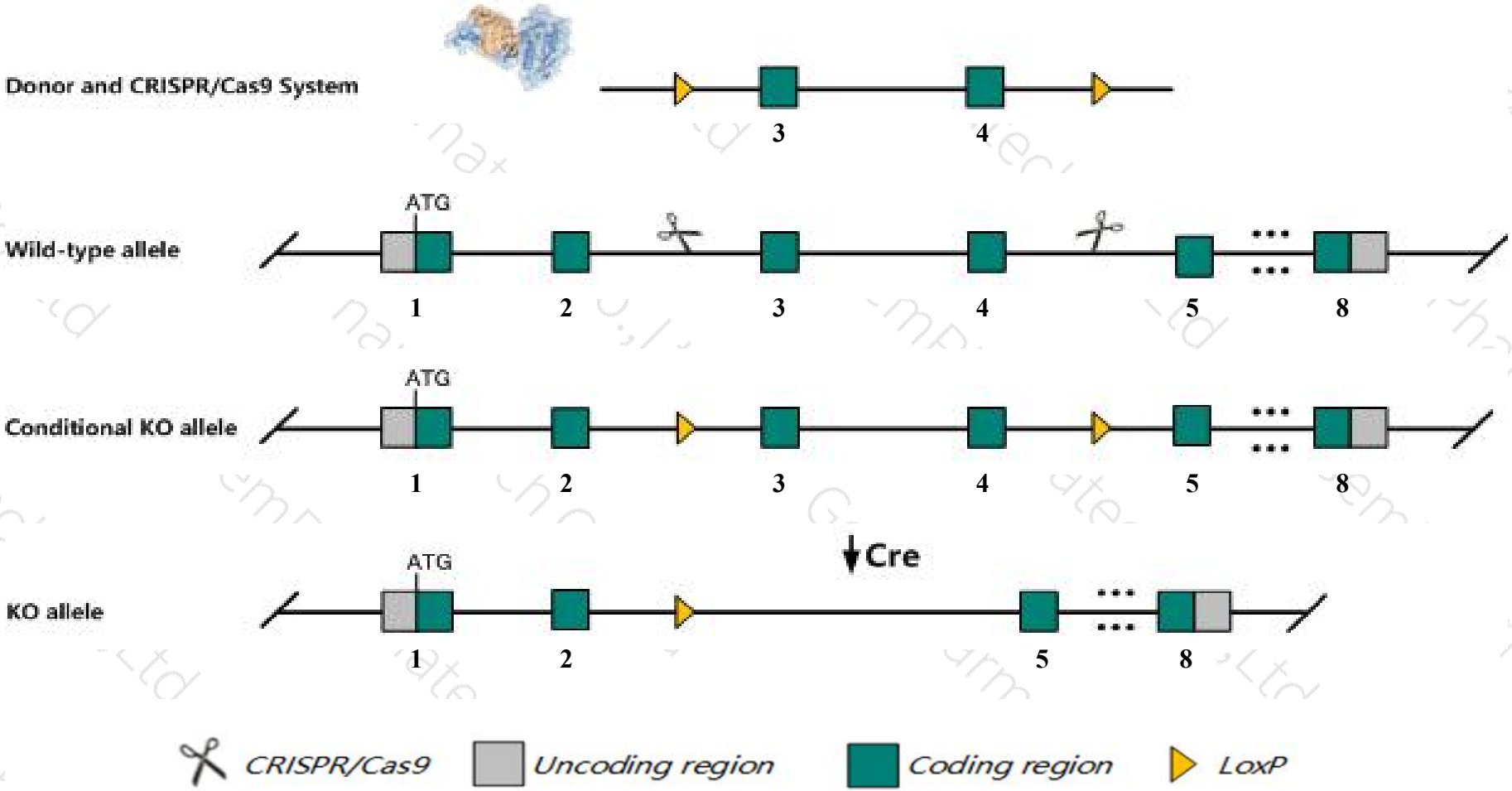
Project Overview

Project Name	***Slc35a1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Slc35a1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Slc35a1* gene has 5 transcripts. According to the structure of *Slc35a1* gene, exon3-exon4 of *Slc35a1-201*(ENSMUST00000029970.13) transcript is recommended as the knockout region.The region contains 313bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Slc35a1* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 mutation of this gene results in lethality before weaning.

➢ The *Slc35a1* gene is located on the Chr4. 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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Slc35a1 solute carrier family 35 (CMP-sialic acid transporter), member 1 [Mus musculus (house mouse)]

Gene ID: 24060, updated on 14-Mar-2020

Summary

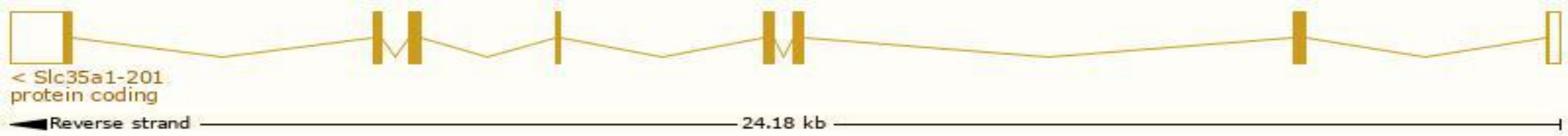
Official Symbol	Slc35a1 provided by MGI
Official Full Name	solute carrier family 35 (CMP-sialic acid transporter), member 1 provided byMGI
Primary source	MGI:MGI:1345622
See related	Ensembl:ENSMUSG00000028293
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AA408150, AI314851, CST
Expression	Ubiquitous expression in colon adult (RPKM 7.9), bladder adult (RPKM 4.2) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

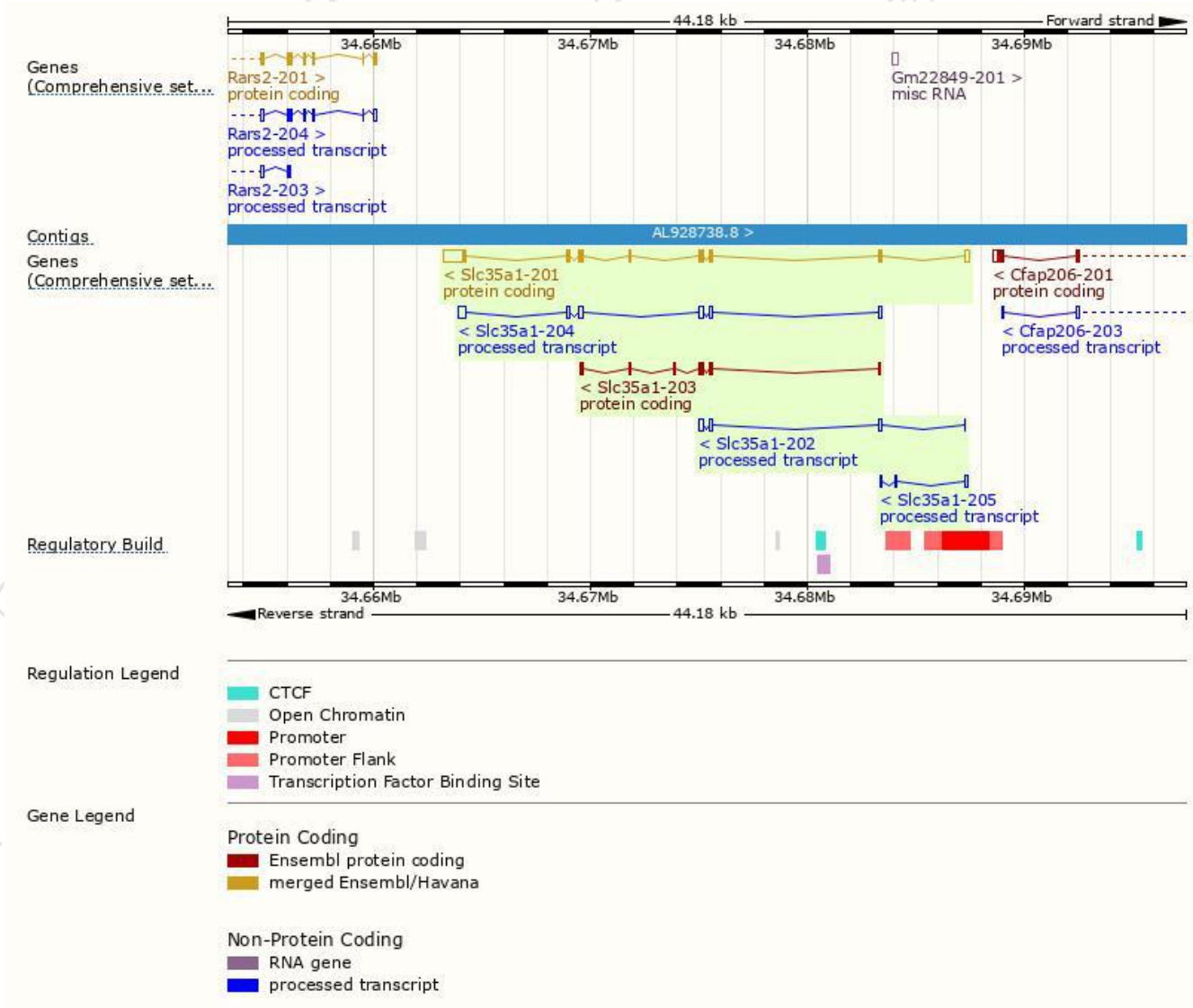
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Slc35a1-201	ENSMUST00000029970.13	2039	336aa	Protein coding	CCDS18030	Q61420	TSL:1 GENCODE basic APPRIS P1
Slc35a1-203	ENSMUST00000126033.1	751	250aa	Protein coding	-	F6R7Y0	CDS 5' and 3' incomplete TSL:3
Slc35a1-204	ENSMUST00000151549.7	1141	No protein	Processed transcript	-	-	TSL:5
Slc35a1-202	ENSMUST00000124456.1	554	No protein	Processed transcript	-	-	TSL:2
Slc35a1-205	ENSMUST00000154020.1	358	No protein	Processed transcript	-	-	TSL:3

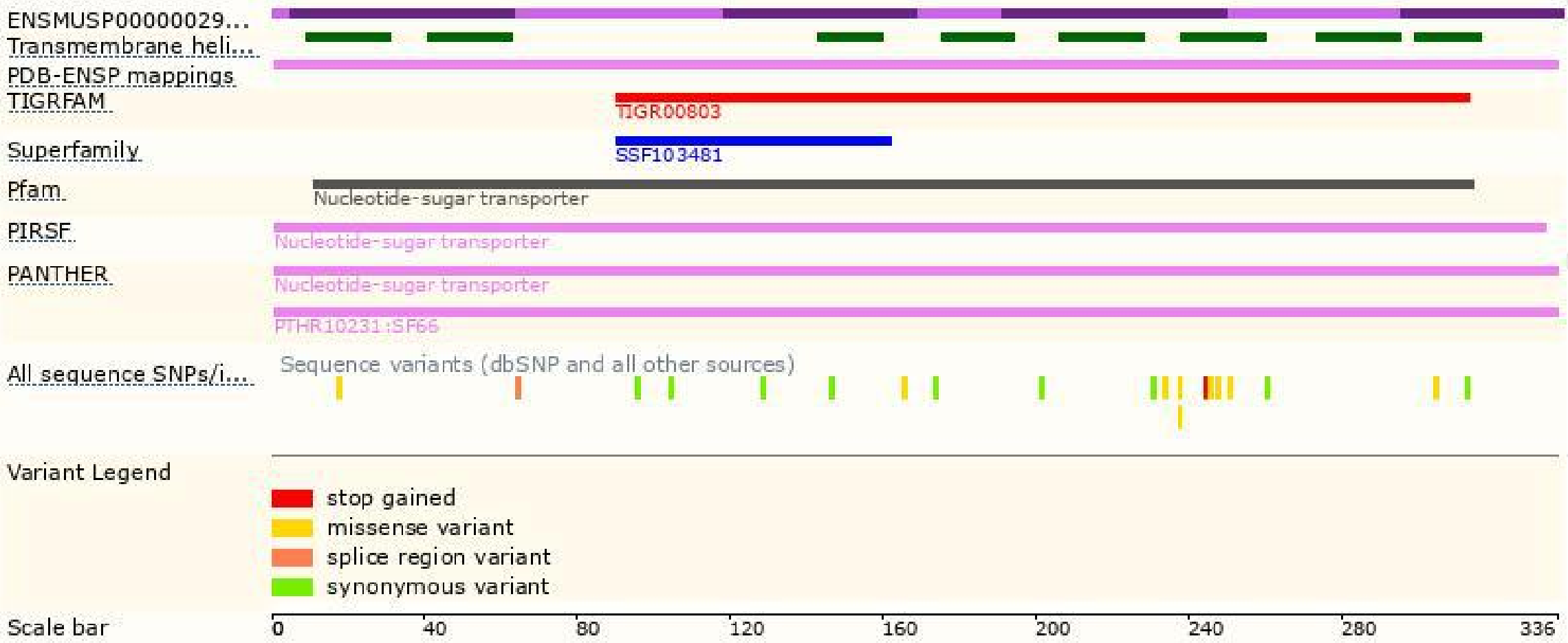
The strategy is based on the design of *Slc35a1-201* transcript,the transcription is shown below:



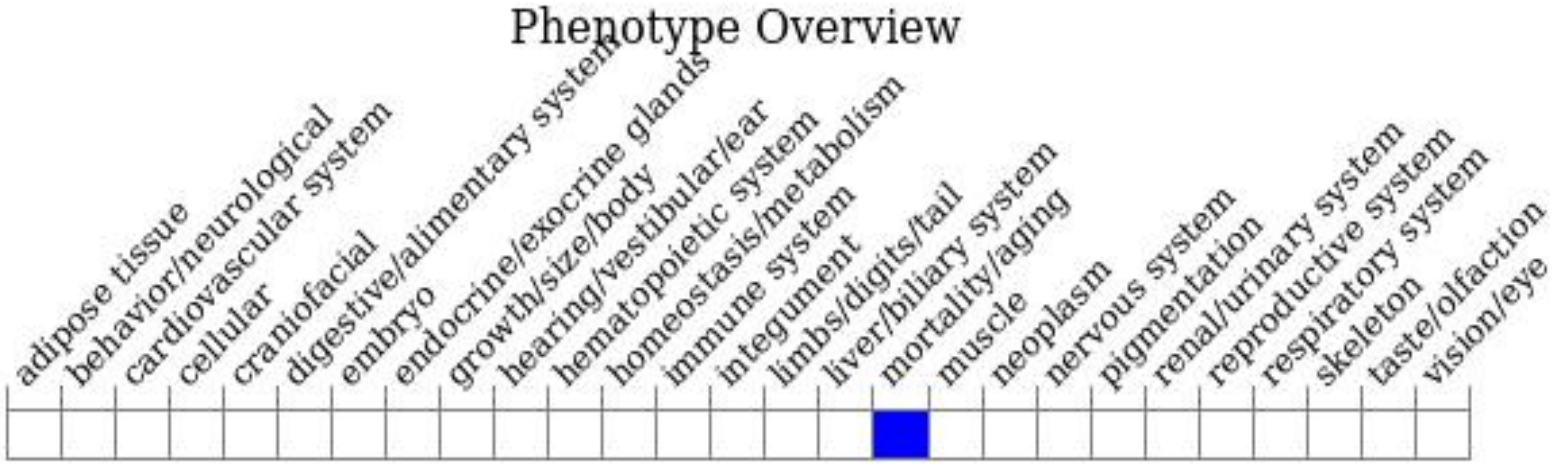
Genomic location distribution



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/).

According to the existing MGI data,homozygous mutation of this gene results in lethality before weaning.

If you have any questions, you are welcome to inquire.

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