Abcd1 Cas9-KO Strategy

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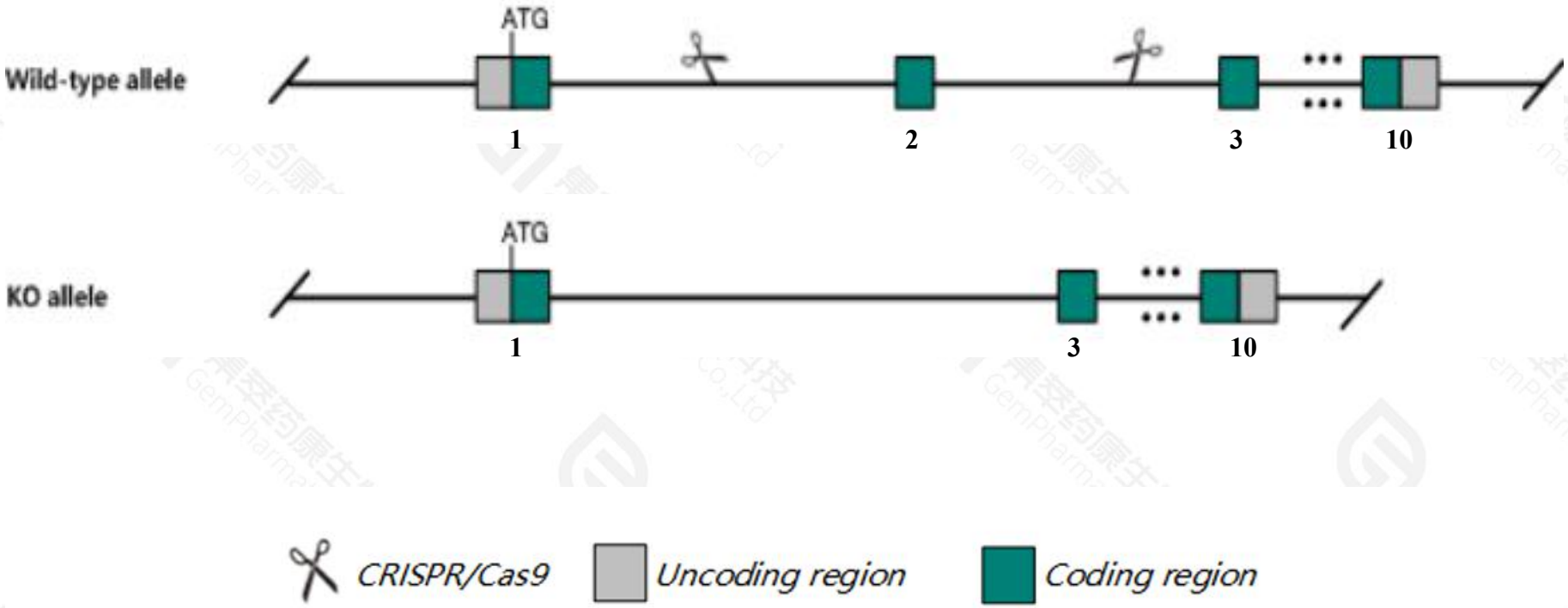
Design Date: 2021-4-16

Project Overview

Project Name	***Abcd1***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Abcd1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Abcd1* gene has 2 transcripts. According to the structure of *Abcd1* gene, exon2 of *Abcd1-201*(ENSMUST00000002084.14) transcript is recommended as the knockout region.The region contains 181bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Abcd1* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice

➢ According to the existing MGI data,mice homozygous/hemizygous for disruptions in this gene develop a late onset of neurodegenerative disease (no neurological symptoms up to 6 months of age), with an accumulation of very long chain fatty acids.

➢ The N-terminal of *Abcd1* gene will remain several amino acids, it may remain the partial function of *Abcd1* gene.

➢ The *Abcd1* gene is located on the ChrX. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

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Abcd1 ATP-binding cassette, sub-family D (ALD), member 1 [Mus musculus (house mouse)]

Gene ID: 11666, updated on 14-Jan-2021

Summary

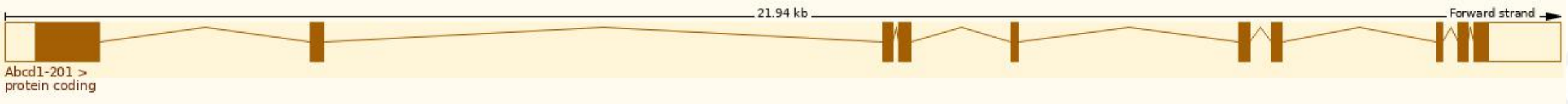
Official Symbol	Abcd1 provided by MGI
Official Full Name	ATP-binding cassette, sub-family D (ALD), member 1 provided byMGI
Primary source	MGI:MGI:1349215
See related	Ensembl:ENSMUSG00000031378
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	A, ALDP, Ald, Aldgh
Summary	The membrane-associated protein encoded by this gene is a member of the superfamily of ATP-binding cassette (ABC) transporters. ABC proteins transport various molecules across extra- and intra-cellular membranes. ABC genes are divided into seven distinct subfamilies (ABC1, MDR/TAP, MRP, ALD, OABP, GCN20, White). This protein is a member of the ALD subfamily, which is involved in peroxisomal import of fatty acids and/or fatty acyl-CoAs in the organelle. All known peroxisomal ABC transporters are half transporters which require a partner half transporter molecule to form a functional homodimeric or heterodimeric transporter. This peroxisomal membrane protein is likely involved in the peroxisomal transport or catabolism of very long chain fatty acids. Defects in the human gene have been identified as the underlying cause of adrenoleukodystrophy, an X-chromosome recessively inherited demyelinating disorder of the nervous system. [provided by RefSeq, Jul 2008]
Expression	Ubiquitous expression in duodenum adult (RPKM 45.9), small intestine adult (RPKM 33.3) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

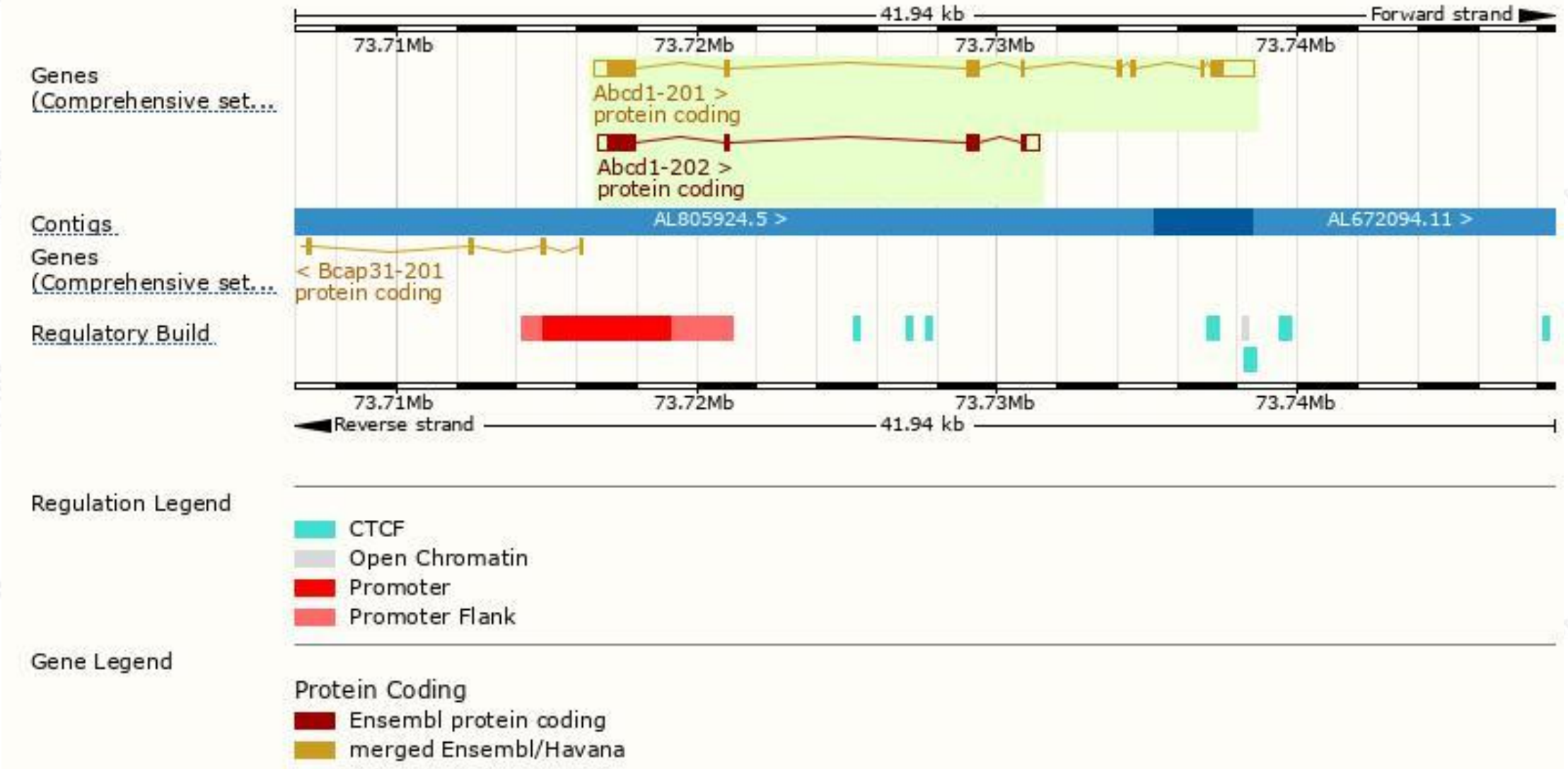
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Abcd1-201	ENSMUST00000002084.14	3648	736aa	Protein coding	CCDS30210		TSL:1 , GENCODE basic , APPRIS P1 ,
Abcd1-202	ENSMUST00000114461.3	2323	504aa	Protein coding	-		TSL:1 , GENCODE basic ,

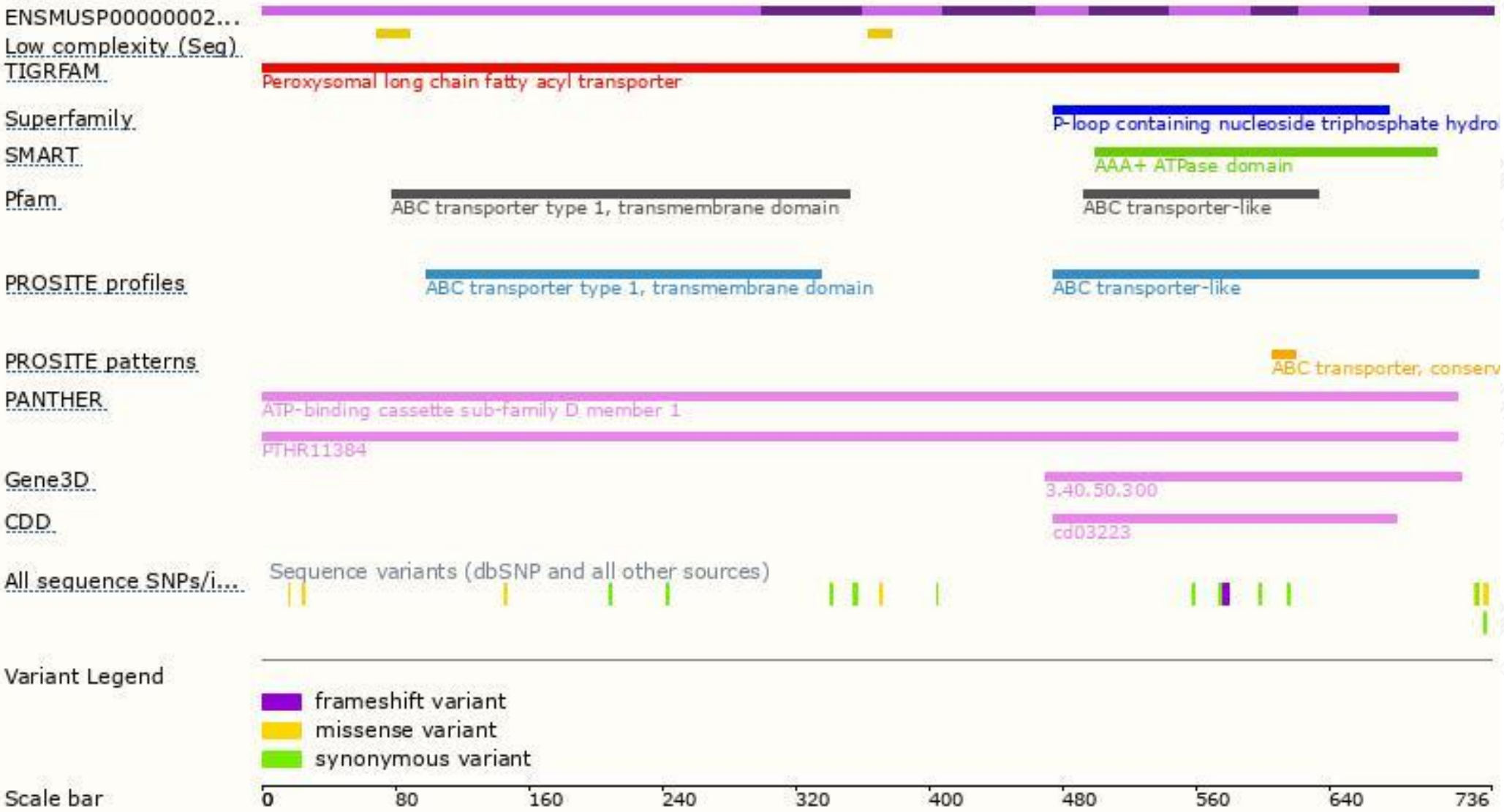
The strategy is based on the design of *Abcd1-201* transcript,the transcription is shown below:



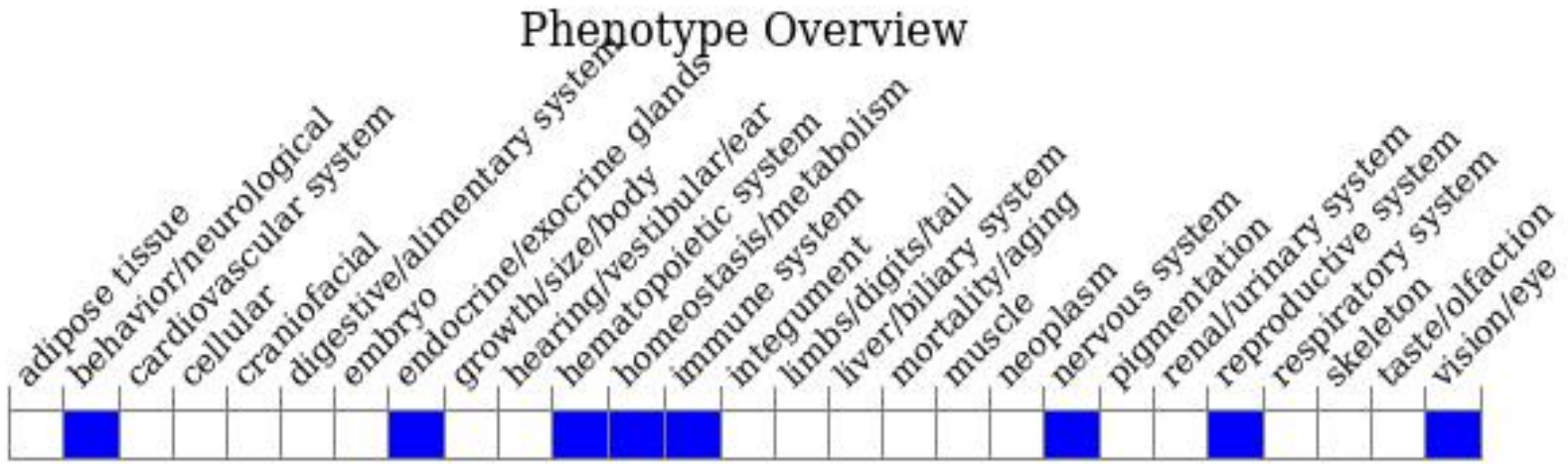
Genomic location distribution



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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