Ap3d1 Cas9-CKO Strategy

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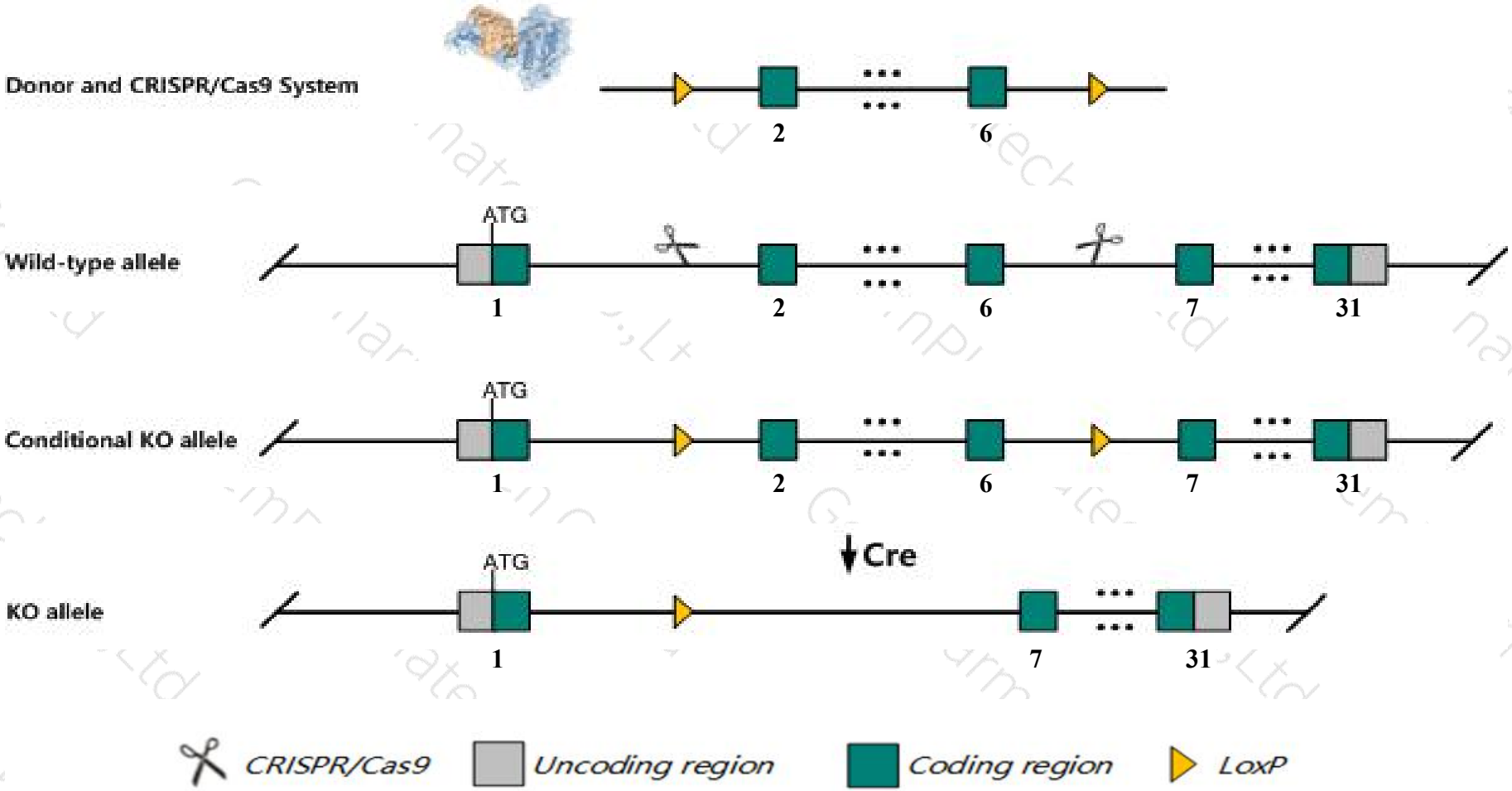
Project Overview

Project Name	***Ap3d1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ap3d1* gene. The schematic diagram is as follows:



Technical routes

- The *Ap3d1* gene has 7 transcripts. According to the structure of *Ap3d1* gene, exon2-exon6 of *Ap3d1-201* (ENSMUST00000020420.8) transcript is recommended as the knockout region.The region contains 496bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Ap3d1* 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,Mutant mice show coat and eye color dilution, platelet defects, lysosomal abnormalities, inner ear degeneration and neurological defects and model Hermansky-Pudlak storage pool deficiency syndrome.

- The *Ap3d1* gene is located on the Chr10. 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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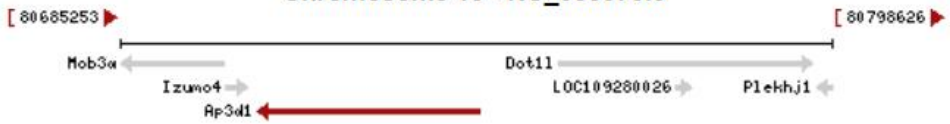
Ap3d1 adaptor-related protein complex 3, delta 1 subunit [*Mus musculus* (house mouse)]

Gene ID: 11776, updated on 12-Aug-2019

Summary

Official Symbol	Ap3d1 provided by MGI
Official Full Name	adaptor-related protein complex 3, delta 1 subunit provided by MGI
Primary source	MGI:MGI:107734
See related	Ensembl:ENSMUSG00000020198
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	mh; Ap3d; Bolvr; mocha; mBLVR1; AA407035
Expression	Ubiquitous expression in testis adult (RPKM 54.8), ovary adult (RPKM 50.0) and 28 other tissues See more
Orthologs	human all

Chromosome 10 - NC_000076.6

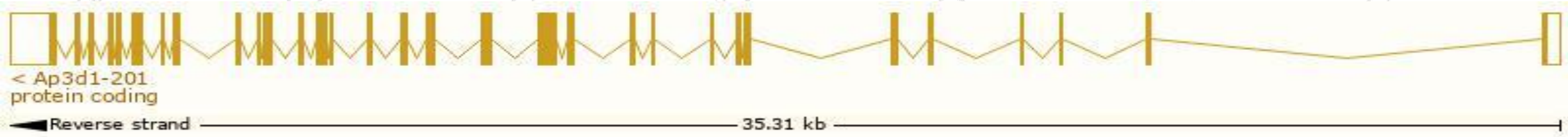


Transcript information（Ensembl）

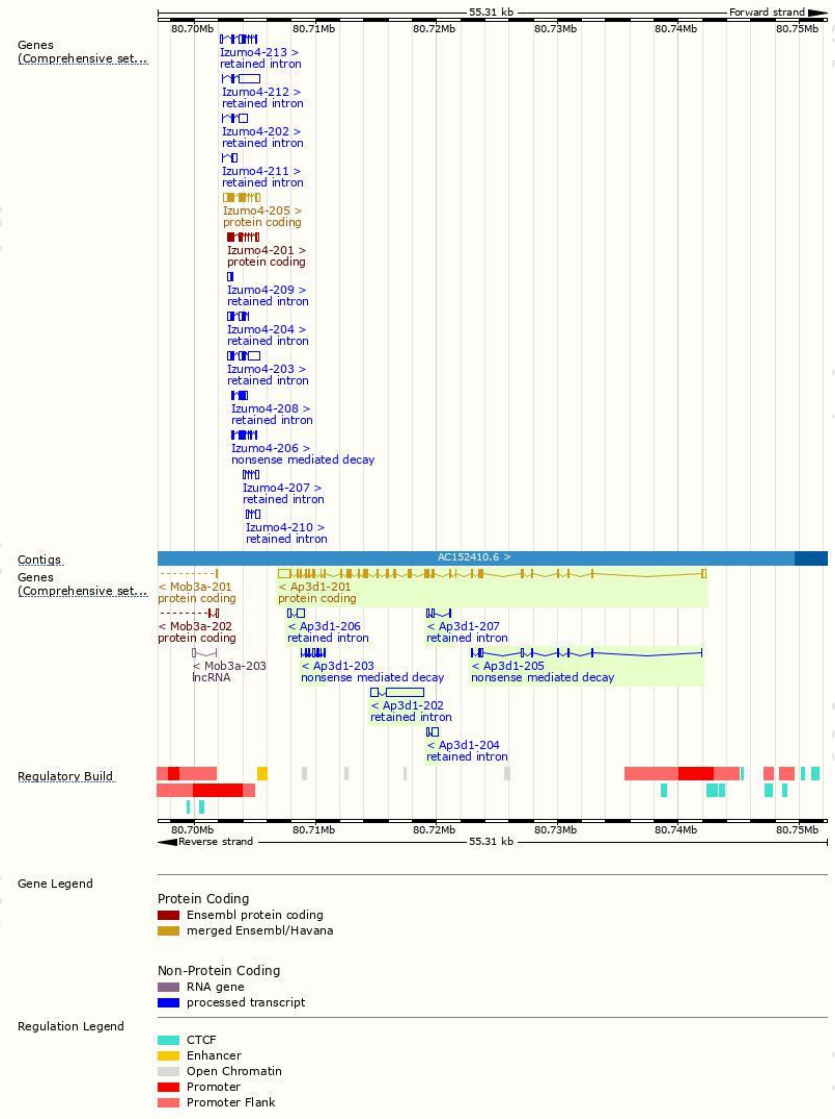
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ap3d1-201	ENSMUST00000020420.8	4805	1199aa	Protein coding	CCDS35984	O54774	TSL:1 GENCODE basic APPRIS P1
Ap3d1-203	ENSMUST00000218610.1	786	116aa	Nonsense mediated decay	-	A0A1W2P7Z2	CDS 5' incomplete TSL:3
Ap3d1-205	ENSMUST00000219356.1	766	122aa	Nonsense mediated decay	-	A0A1W2P6Q6	CDS 5' incomplete TSL:3
Ap3d1-202	ENSMUST00000218125.1	3728	No protein	Retained intron	-	-	TSL:1
Ap3d1-206	ENSMUST00000219816.1	858	No protein	Retained intron	-	-	TSL:2
Ap3d1-204	ENSMUST00000219253.1	668	No protein	Retained intron	-	-	TSL:2
Ap3d1-207	ENSMUST00000220183.1	422	No protein	Retained intron	-	-	TSL:2

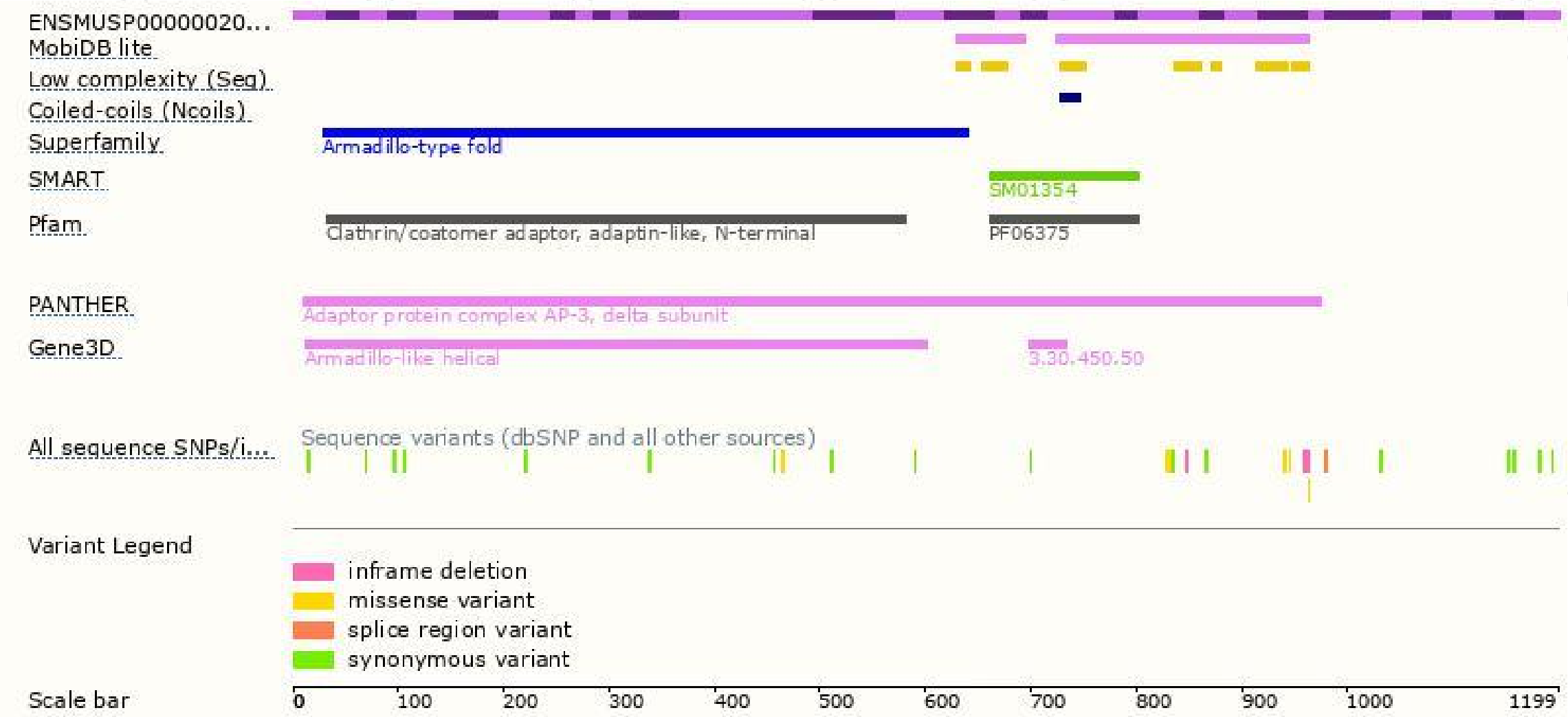
The strategy is based on the design of *Ap3d1-201* transcript,The transcription is shown below



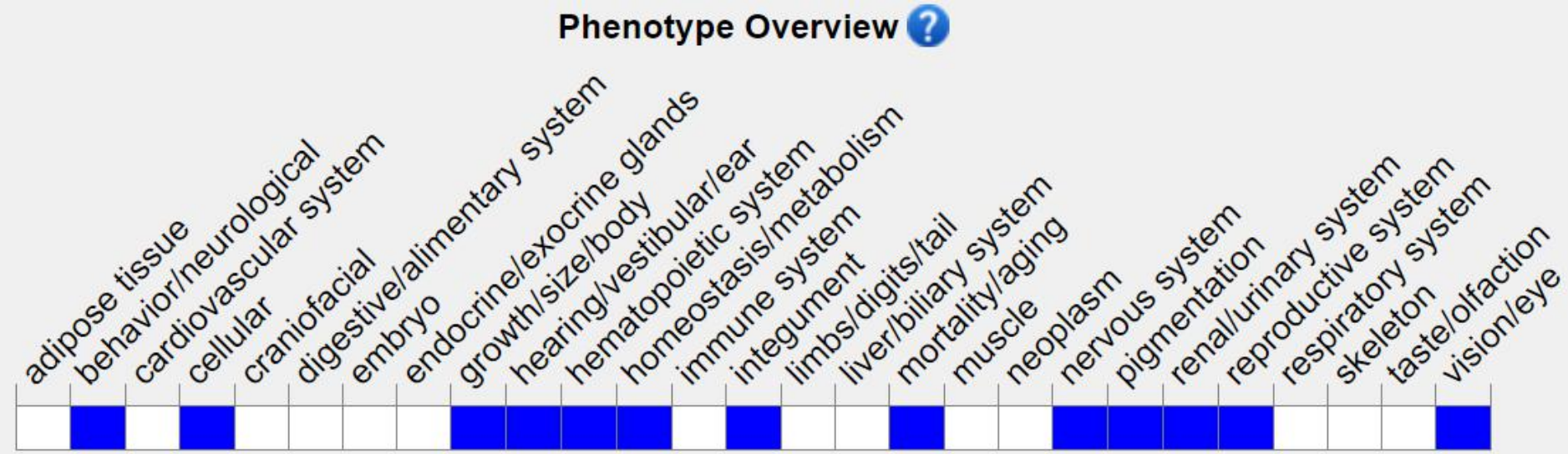
Genomic location distribution



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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