Agl Cas9-CKO Strategy

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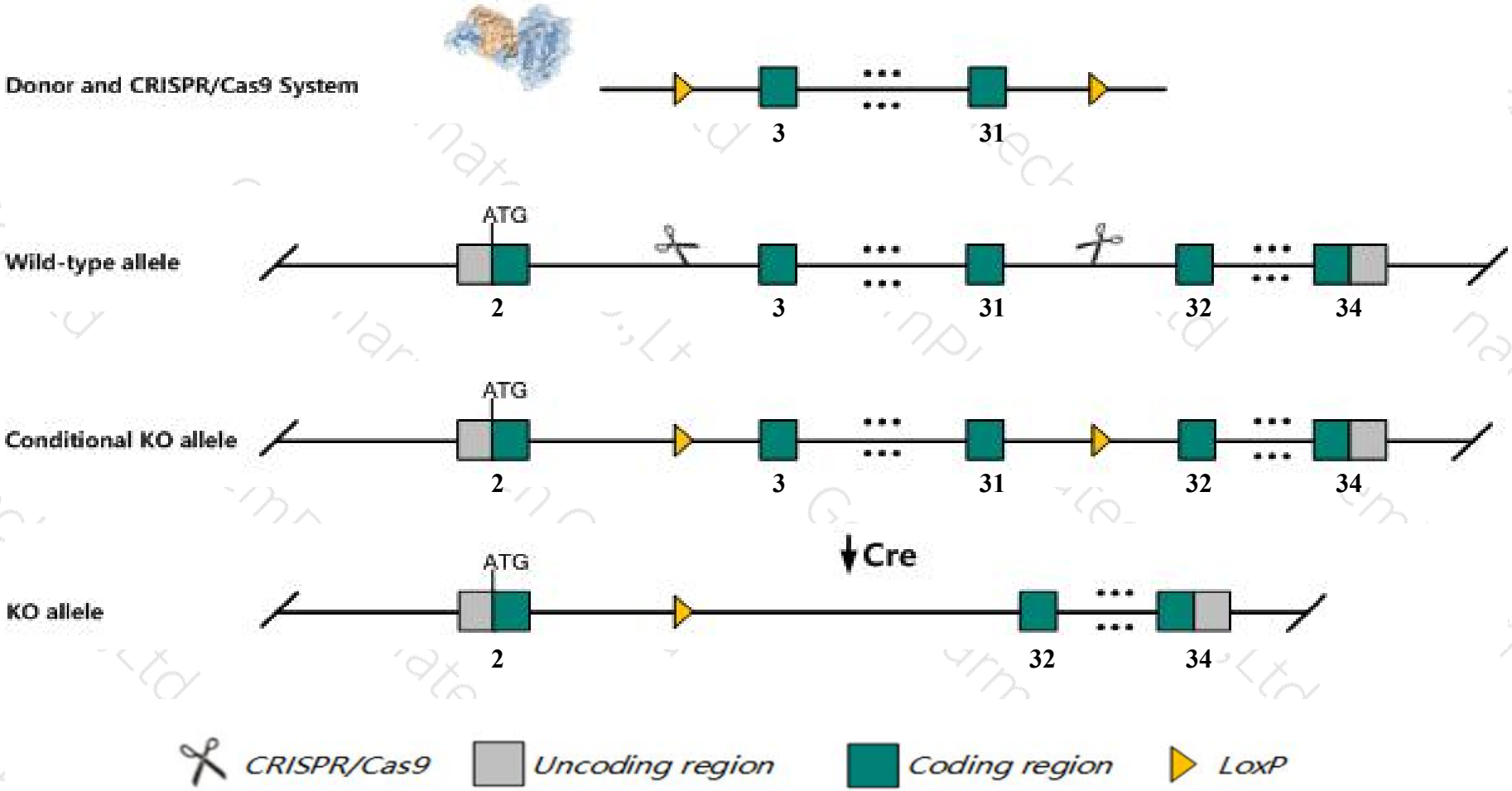
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Agl* gene has 8 transcripts. According to the structure of *Agl* gene, exon3-exon31 of *Agl-201* (ENSMUST00000040603.13) transcript is recommended as the knockout region.The region contains 4177bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Agl* 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 inactivation of this gene leads to hypoglycemia, altered blood biochemistry, severe hepatomegaly, glycogen accumulation in the liver, heart, skeletal muscle and other tissues, motor impairment, and premature death.

- The *Agl* 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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Agl amylo-1,6-glucosidase, 4-alpha-glucanotransferase [Mus musculus (house mouse)]

Gene ID: 77559, updated on 31-Jan-2019

Summary

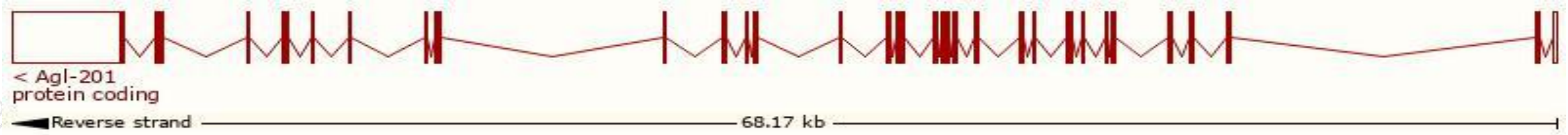
Official Symbol	Agl provided by MGI
Official Full Name	amylo-1,6-glucosidase, 4-alpha-glucanotransferase provided by MGI
Primary source	MGI:MGI:1924809
See related	Ensembl:ENSMUSG00000033400
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1110061O17Rik, 9430004C13Rik, 9630046L06Rik, AI850929, C77197
Expression	Ubiquitous expression in heart adult (RPKM 13.2), liver E18 (RPKM 6.5) and 26 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

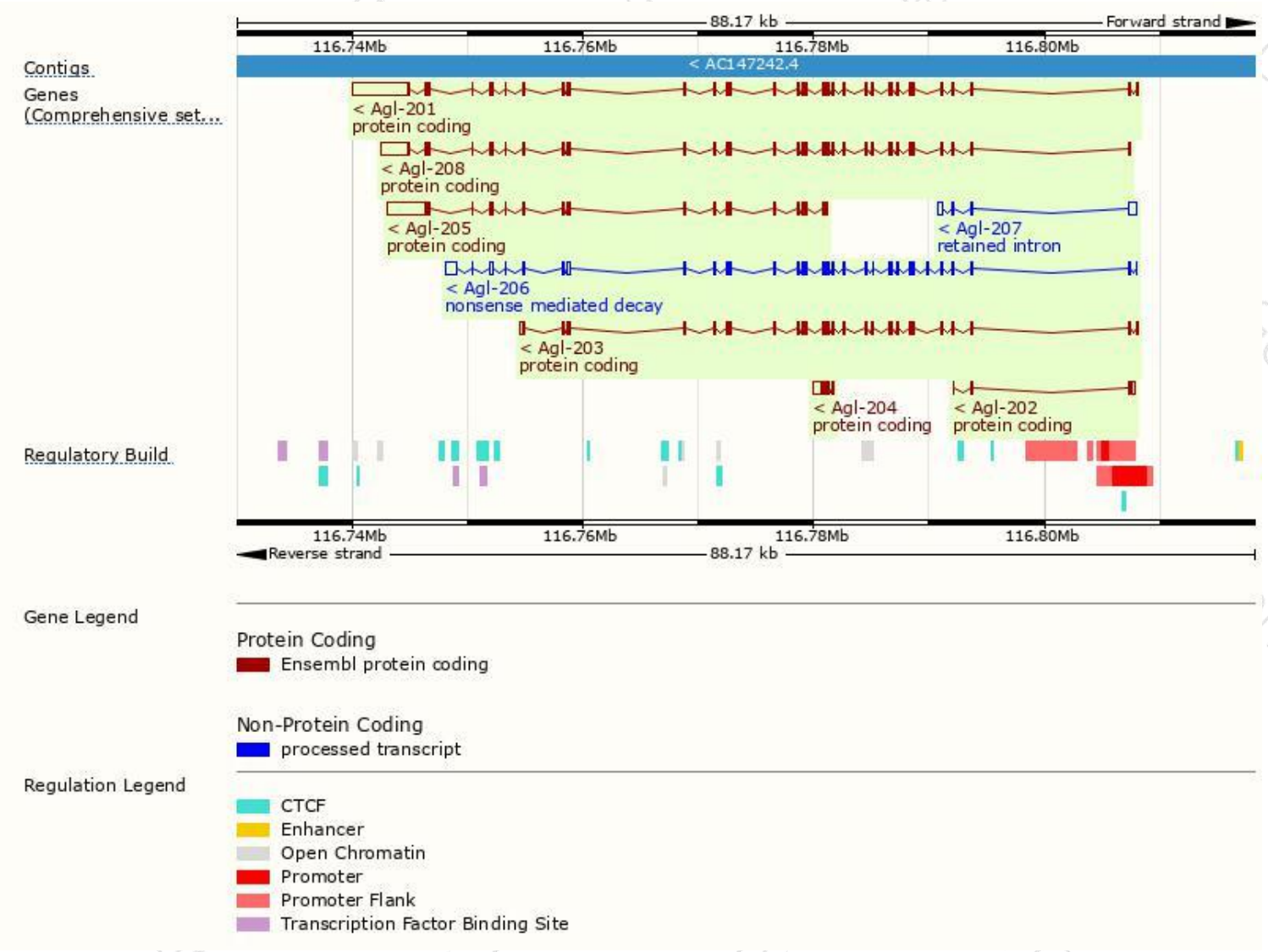
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Agl-201	ENSMUST00000040603.13	9625	1532aa	Protein coding	CCDS38613	F8VPN4	TSL:5 GENCODE basic APPRIS P1
Agl-208	ENSMUST00000162792.7	6978	1532aa	Protein coding	CCDS38613	F8VPN4	TSL:2 GENCODE basic APPRIS P1
Agl-205	ENSMUST00000160484.5	5817	830aa	Protein coding	-	F6XXE6	CDS 5' incomplete TSL:1
Agl-203	ENSMUST00000159742.7	4344	1279aa	Protein coding	-	A0A0G2JGI9	TSL:1 GENCODE basic
Agl-204	ENSMUST00000159995.1	1285	198aa	Protein coding	-	F7CSZ6	CDS 5' incomplete TSL:1
Agl-202	ENSMUST00000159670.2	722	116aa	Protein coding	-	E0CX86	CDS 3' incomplete TSL:5
Agl-206	ENSMUST00000161336.7	5365	233aa	Nonsense mediated decay	-	E0CYU6	TSL:5
Agl-207	ENSMUST00000162040.1	1572	No protein	Retained intron	-	-	TSL:1

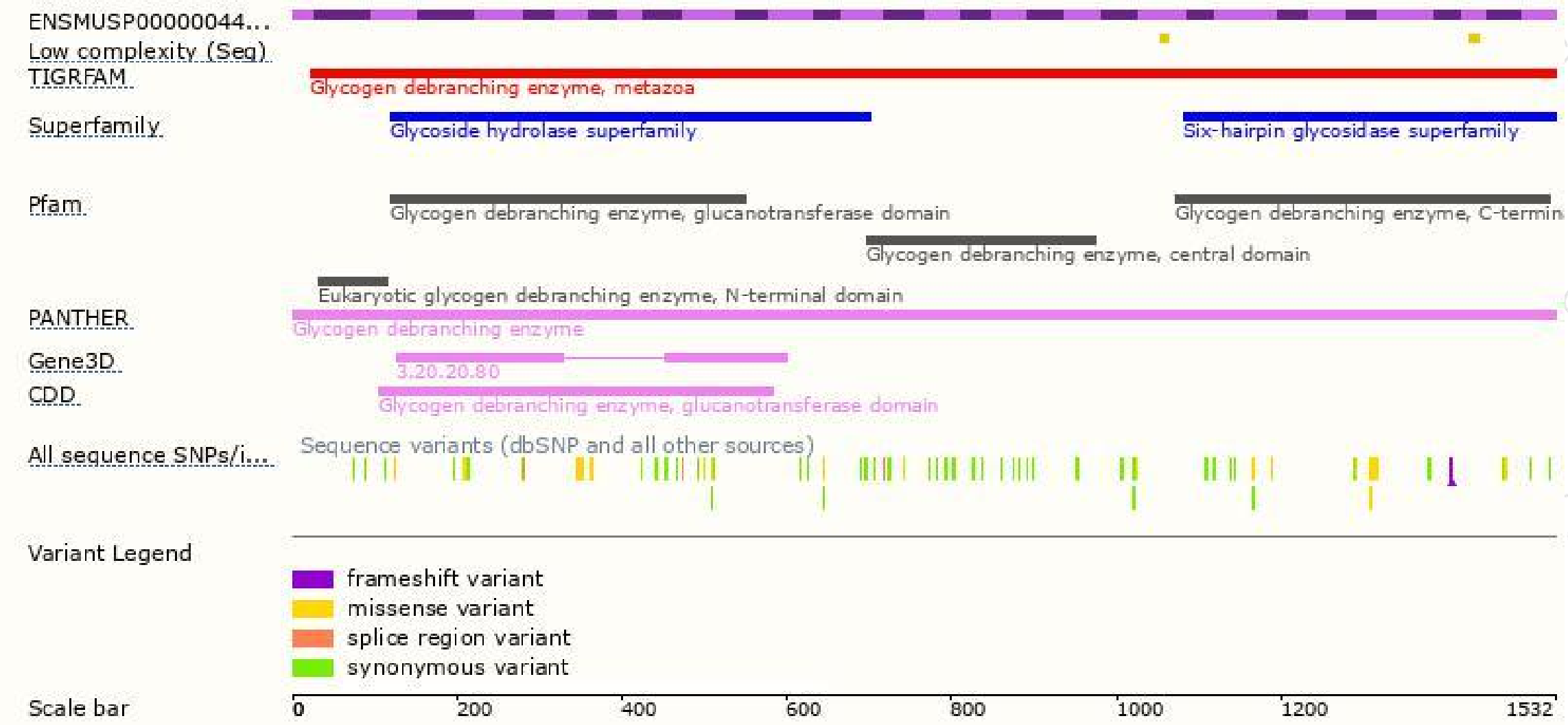
The strategy is based on the design of *Agl-201* transcript,The transcription is shown below



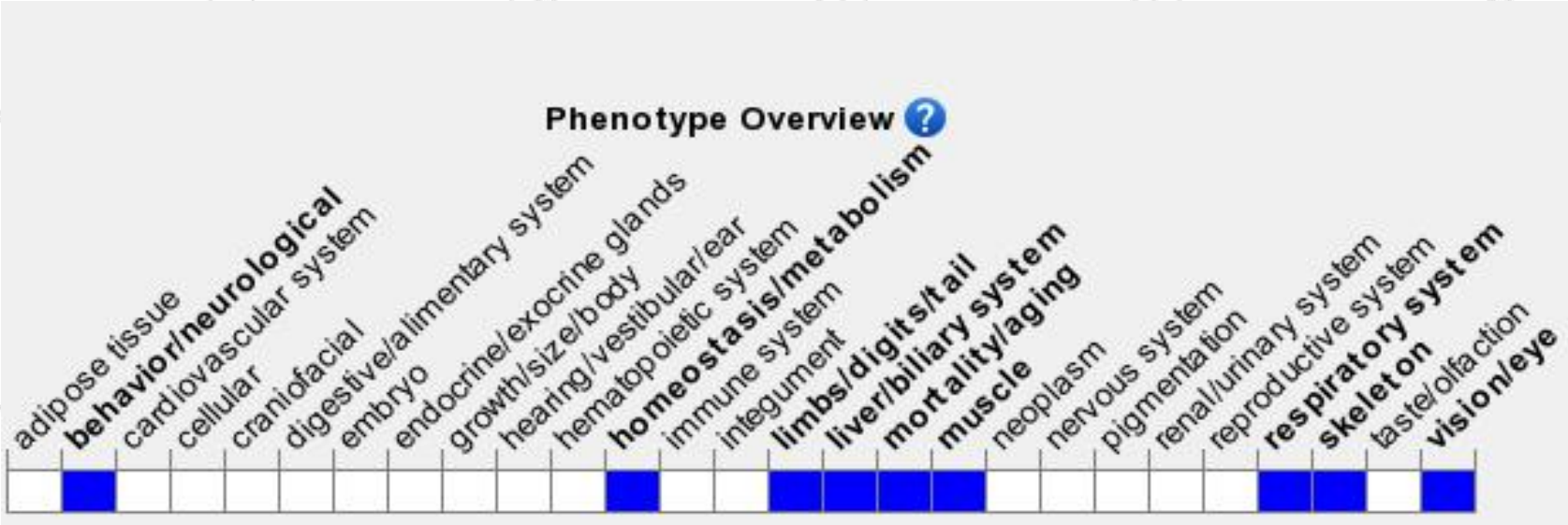
Genomic location distribution



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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