Gnptab Cas9-CKO Strategy

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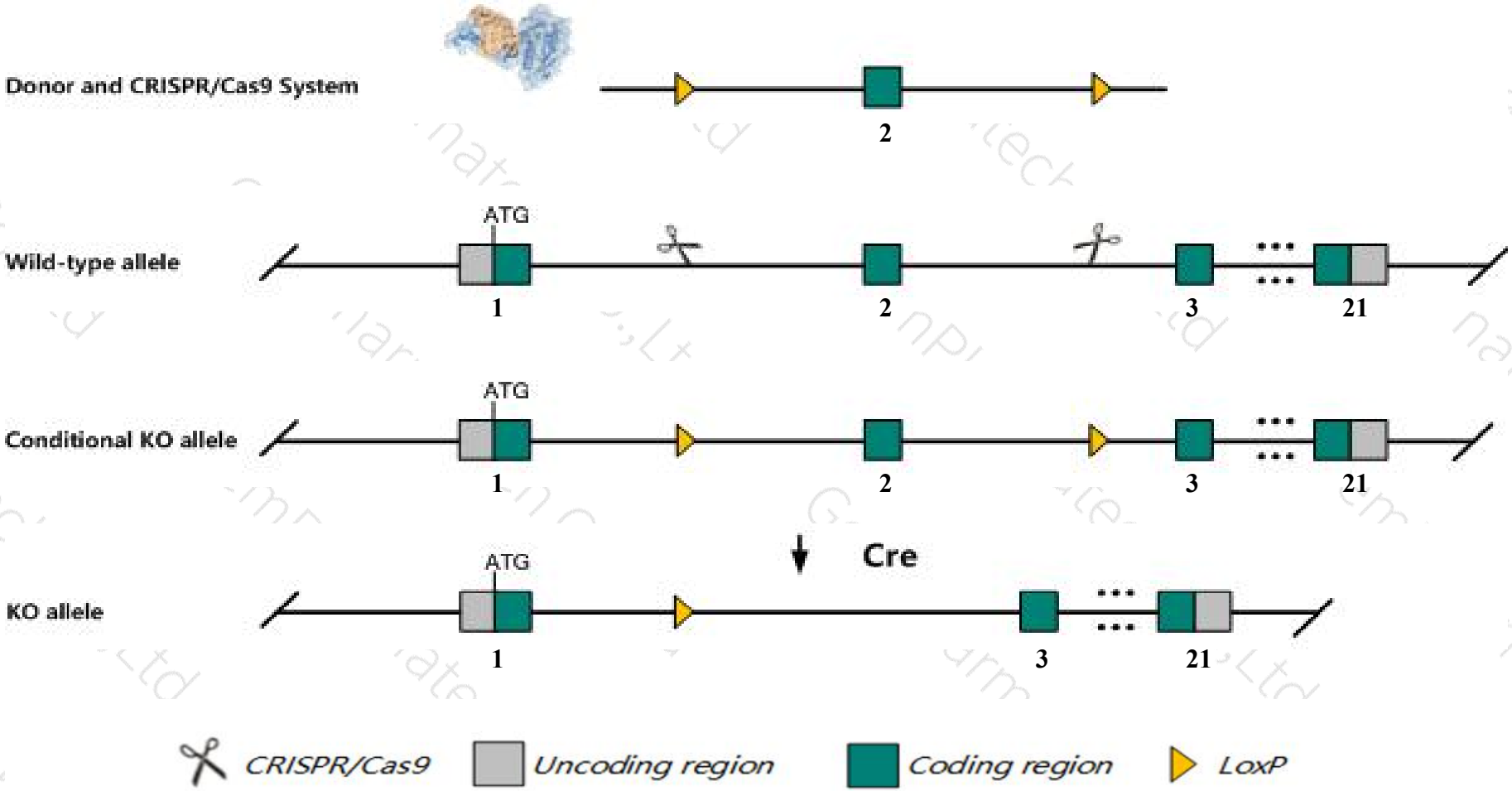
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Gnptab* gene has 7 transcripts. According to the structure of *Gnptab* gene, exon2 of *Gnptab-201* (ENSMUST00000020251.9) transcript is recommended as the knockout region.The region contains 86bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Gnptab* 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 mutations cause stunted growth, high lysosomal enzyme levels, skeletal defects, retinal degeneration and secretory cell lesions. Homozygotes for an ENU allele show skeletal and facial defects, altered enzymatic activities, lysosomal storage, Purkinje cell loss, ataxia and premature death.

➢ The *Gnptab* 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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Gnptab N-acetylglucosamine-1-phosphate transferase, alpha and beta subunits [Mus musculus (house mouse)]

Gene ID: 432486, updated on 31-Jan-2019

Summary

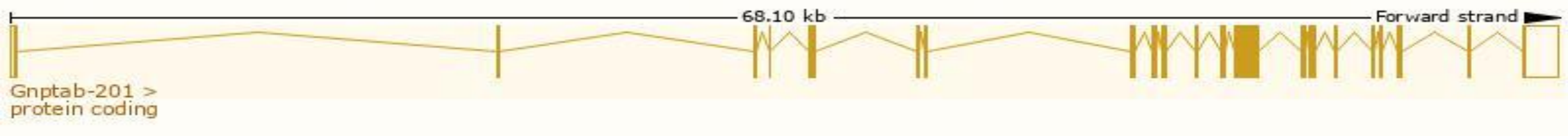
Official Symbol	Gnptab provided by MGI
Official Full Name	N-acetylglucosamine-1-phosphate transferase, alpha and beta subunits provided by MGI
Primary source	MGI:MGI:3643902
See related	Ensembl:ENSMUSG00000035311
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	EG432486
Expression	Ubiquitous expression in CNS E18 (RPKM 16.6), subcutaneous fat pad adult (RPKM 10.3) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

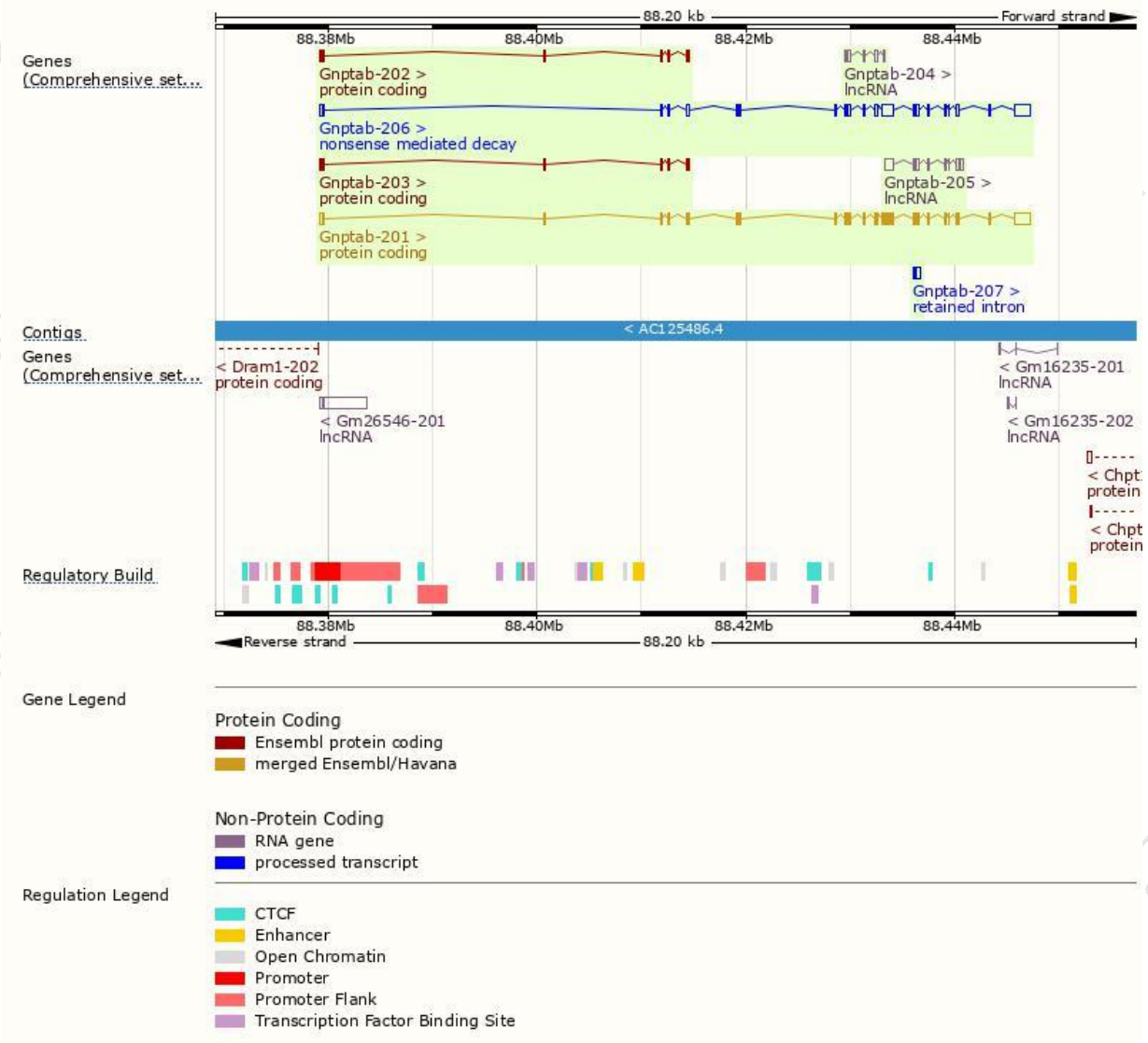
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gnptab-201	ENSMUST00000020251.9	5390	1235aa	Protein coding	CCDS24110	Q69ZN6	TSL:1 GENCODE basic APPRIS P1
Gnptab-202	ENSMUST00000127615.7	686	174aa	Protein coding	-	D3Z1C3	CDS 3' incomplete TSL:2
Gnptab-203	ENSMUST00000130301.7	569	166aa	Protein coding	-	D3YXC6	CDS 3' incomplete TSL:5
Gnptab-206	ENSMUST00000151273.7	5297	56aa	Nonsense mediated decay	-	D6RJ30	TSL:1
Gnptab-207	ENSMUST00000155306.1	536	No protein	Retained intron	-	-	TSL:3
Gnptab-205	ENSMUST00000141343.1	1856	No protein	lncRNA	-	-	TSL:1
Gnptab-204	ENSMUST00000132738.1	726	No protein	lncRNA	-	-	TSL:3

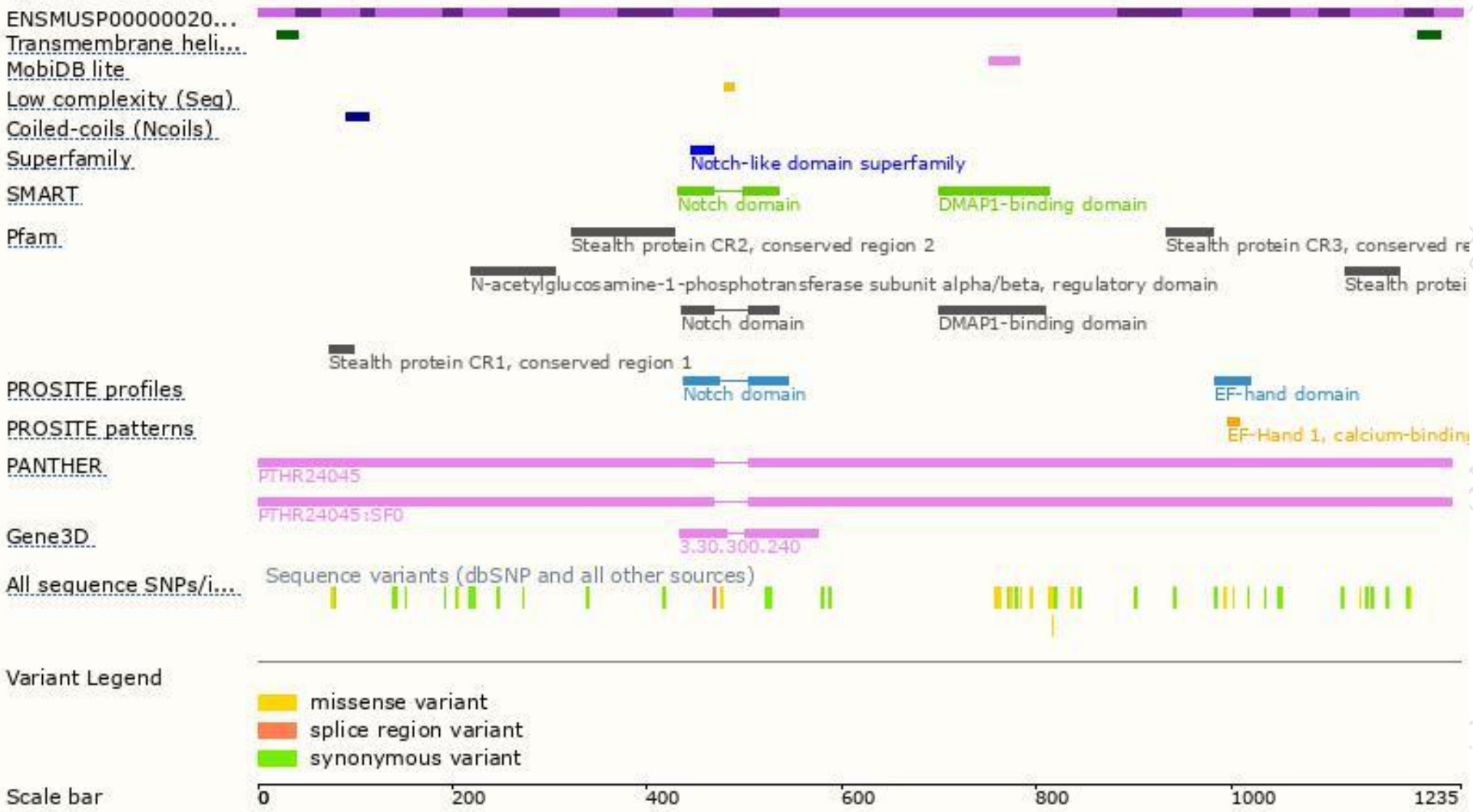
The strategy is based on the design of *Gnptab-201* transcript,The transcription is shown below



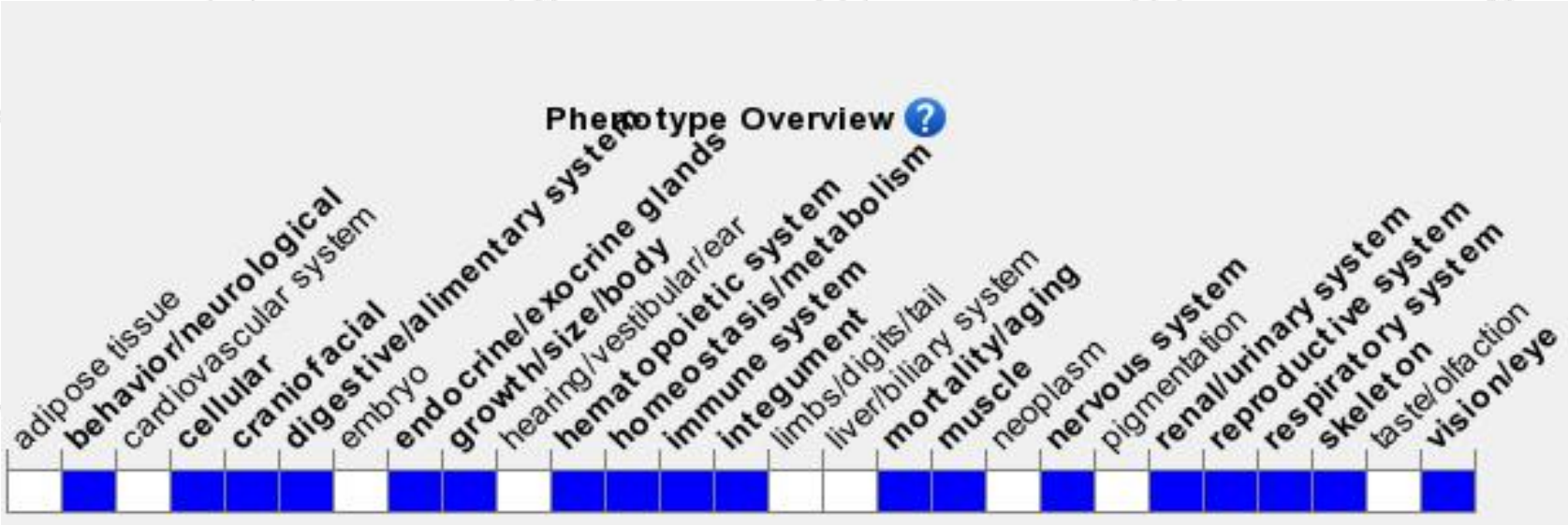
Genomic location distribution



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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