

***Gnb3* Cas9-CKO Strategy**

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Reviewer:

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Design Date:

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Project Overview

Project Name

Gnb3

Project type

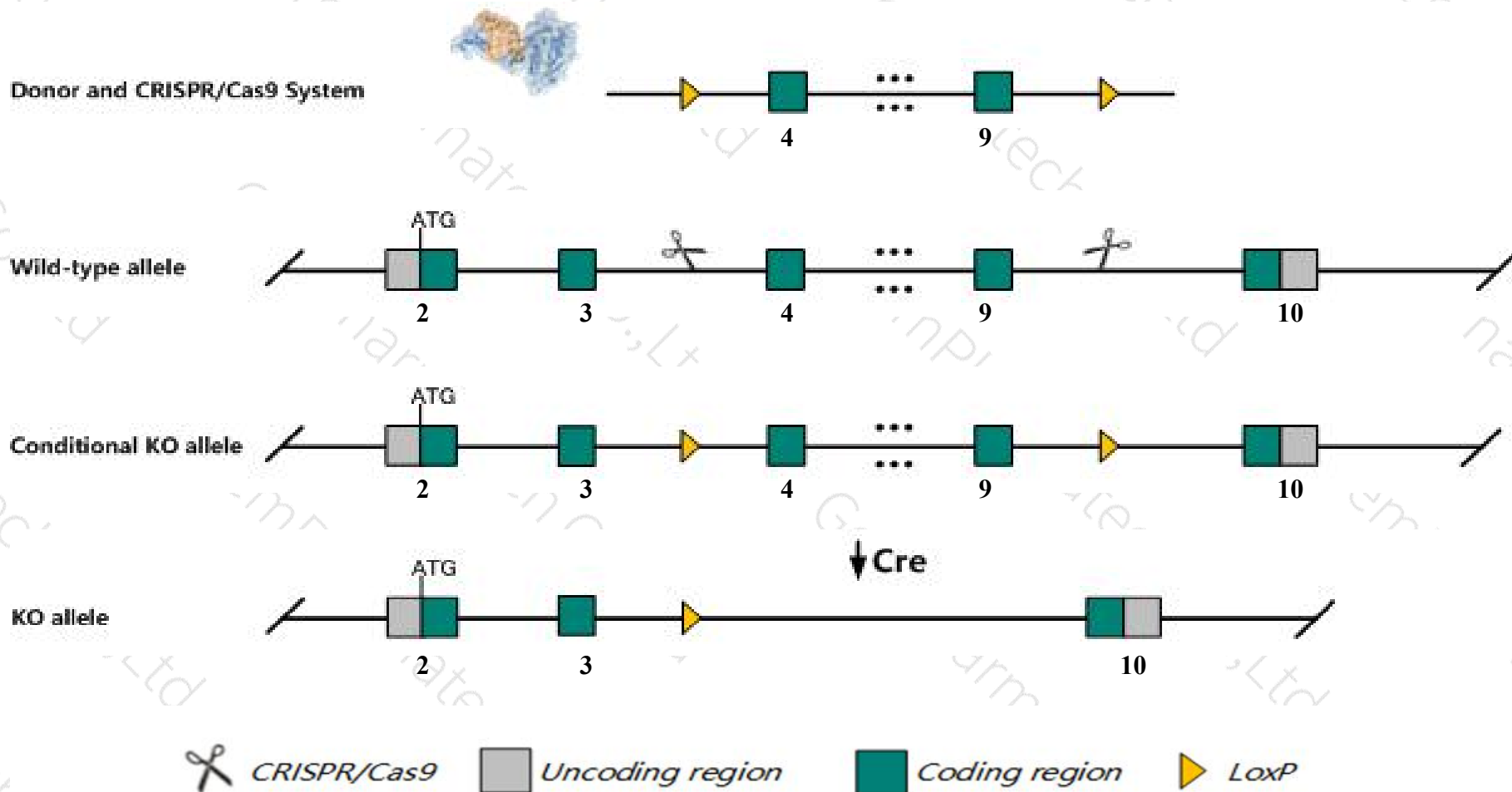
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gnb3* gene. The schematic diagram is as follows:



Technical routes

- The *Gnb3* gene has 4 transcripts. According to the structure of *Gnb3* gene, exon4-exon9 of *Gnb3-201* (ENSMUST00000024206.5) transcript is recommended as the knockout region. The region contains 820bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gnb3* 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.

- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit abnormal light ON response and synaptic maintenance.
- The *Gnb3* gene is located on the Chr6. 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)

Gnb3 guanine nucleotide binding protein (G protein), beta 3 [Mus musculus (house mouse)]

Gene ID: 14695, updated on 31-Jan-2019

Summary



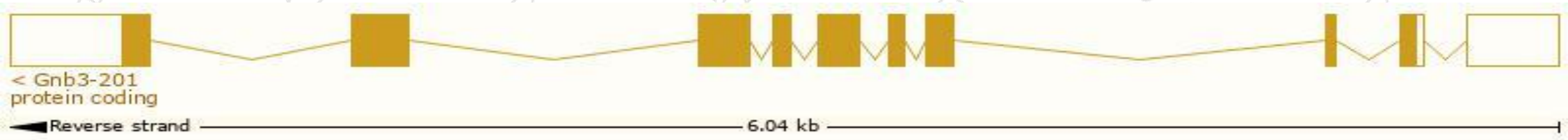
Official Symbol	Gnb3 provided by MGI
Official Full Name	guanine nucleotide binding protein (G protein), beta 3 provided by MGI
Primary source	MGI:MGI:95785
See related	Ensembl:ENSMUSG00000023439
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gnb-3
Expression	Biased expression in adrenal adult (RPKM 3.8), heart adult (RPKM 3.5) and 9 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

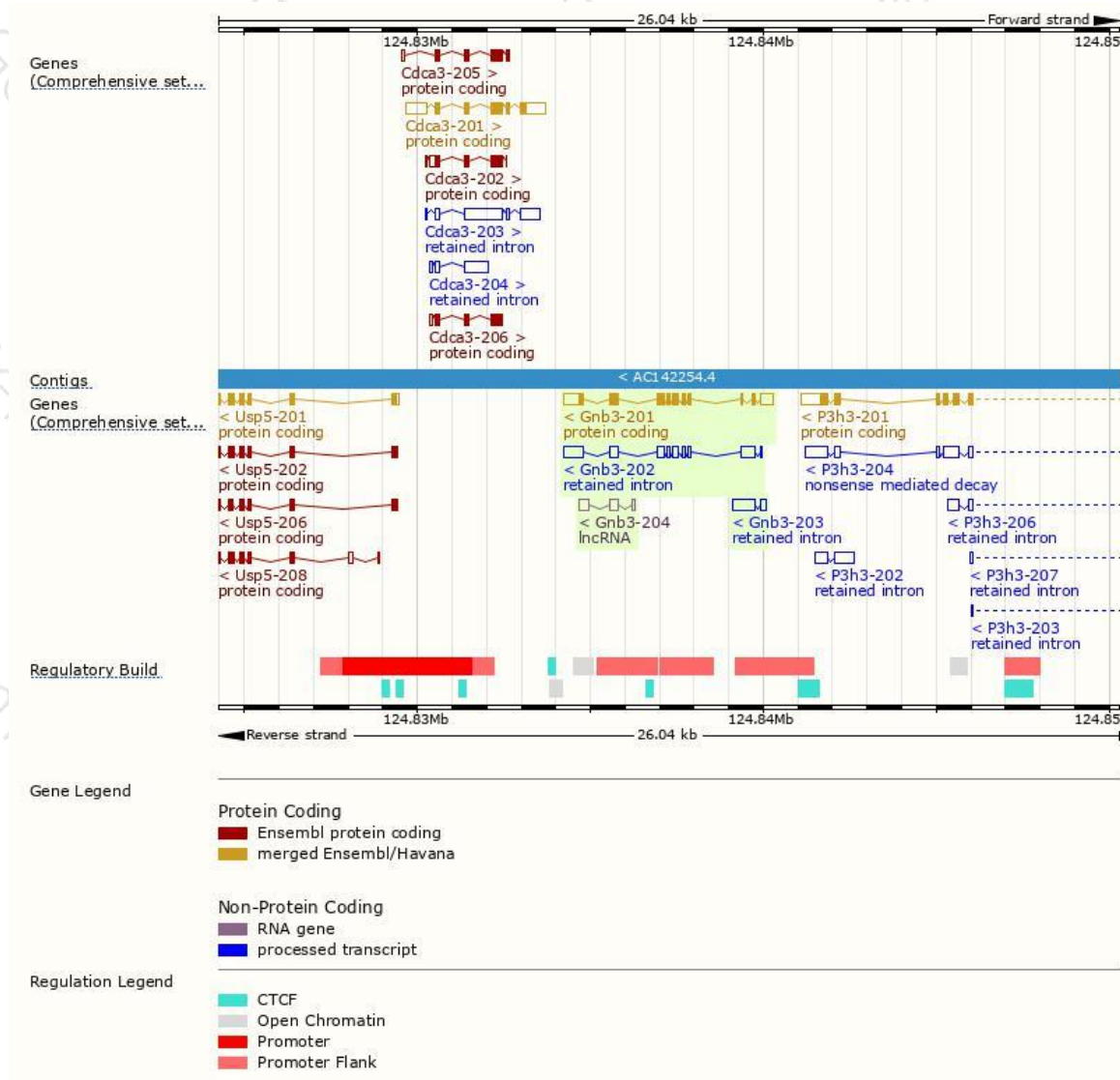
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gnb3-201	ENSMUST00000024206.5	1846	340aa	Protein coding	CCDS20532	Q54AE3 Q61011	TSL:1 GENCODE basic APPRIS P1
Gnb3-202	ENSMUST00000129251.7	1779	No protein	Retained intron	-	-	TSL:1
Gnb3-203	ENSMUST00000135996.1	816	No protein	Retained intron	-	-	TSL:3
Gnb3-204	ENSMUST00000140233.1	582	No protein	lncRNA	-	-	TSL:3

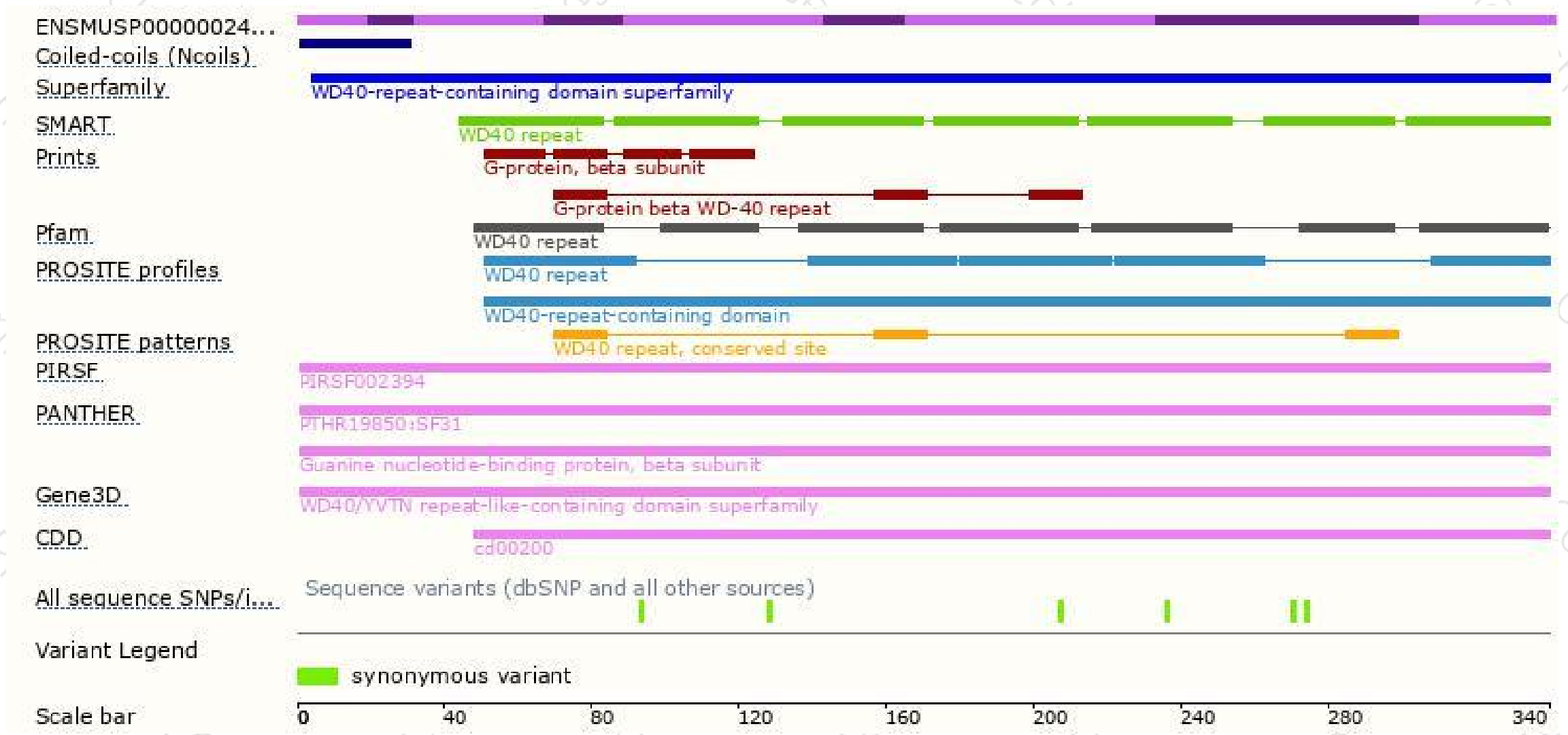
The strategy is based on the design of *Gnb3-201* transcript,The transcription is shown below



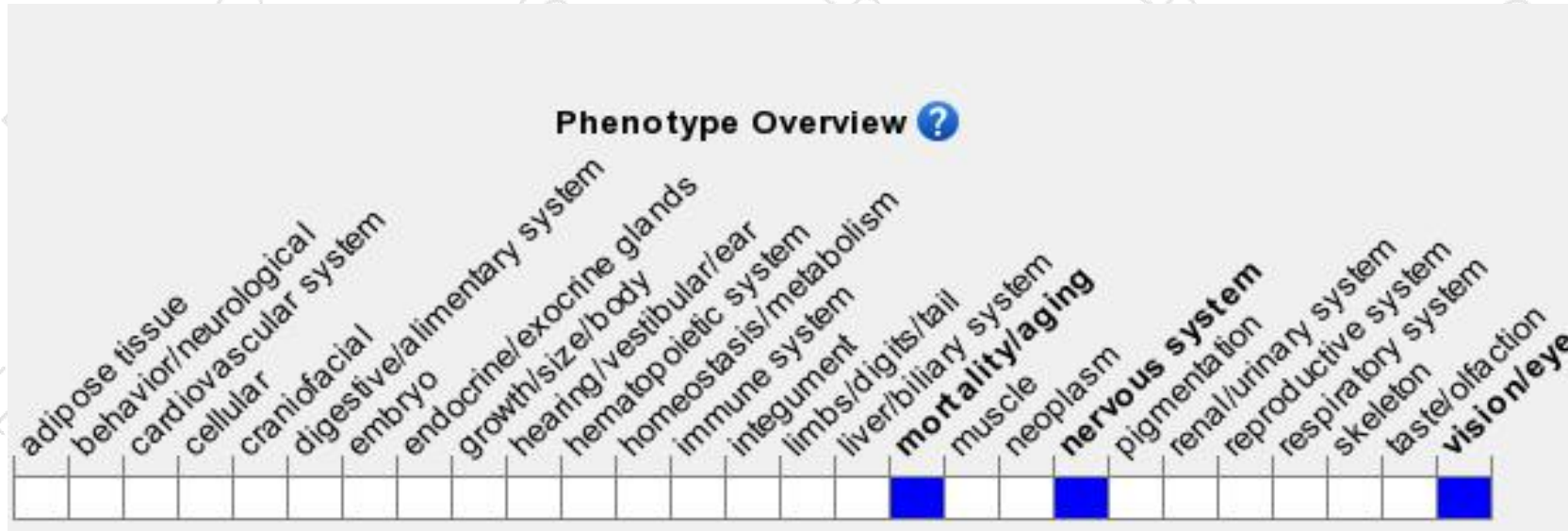
Genomic location distribution



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, Mice homozygous for a knock-out allele exhibit abnormal light ON response and synaptic maintenance.

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

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