

***Gnat1* Cas9-CKO Strategy**

Designer: Xueting Zhang

Reviewer: Yanhua Shen

Date: 2019-12-13

Project Overview

Project Name

Gnat1

Project type

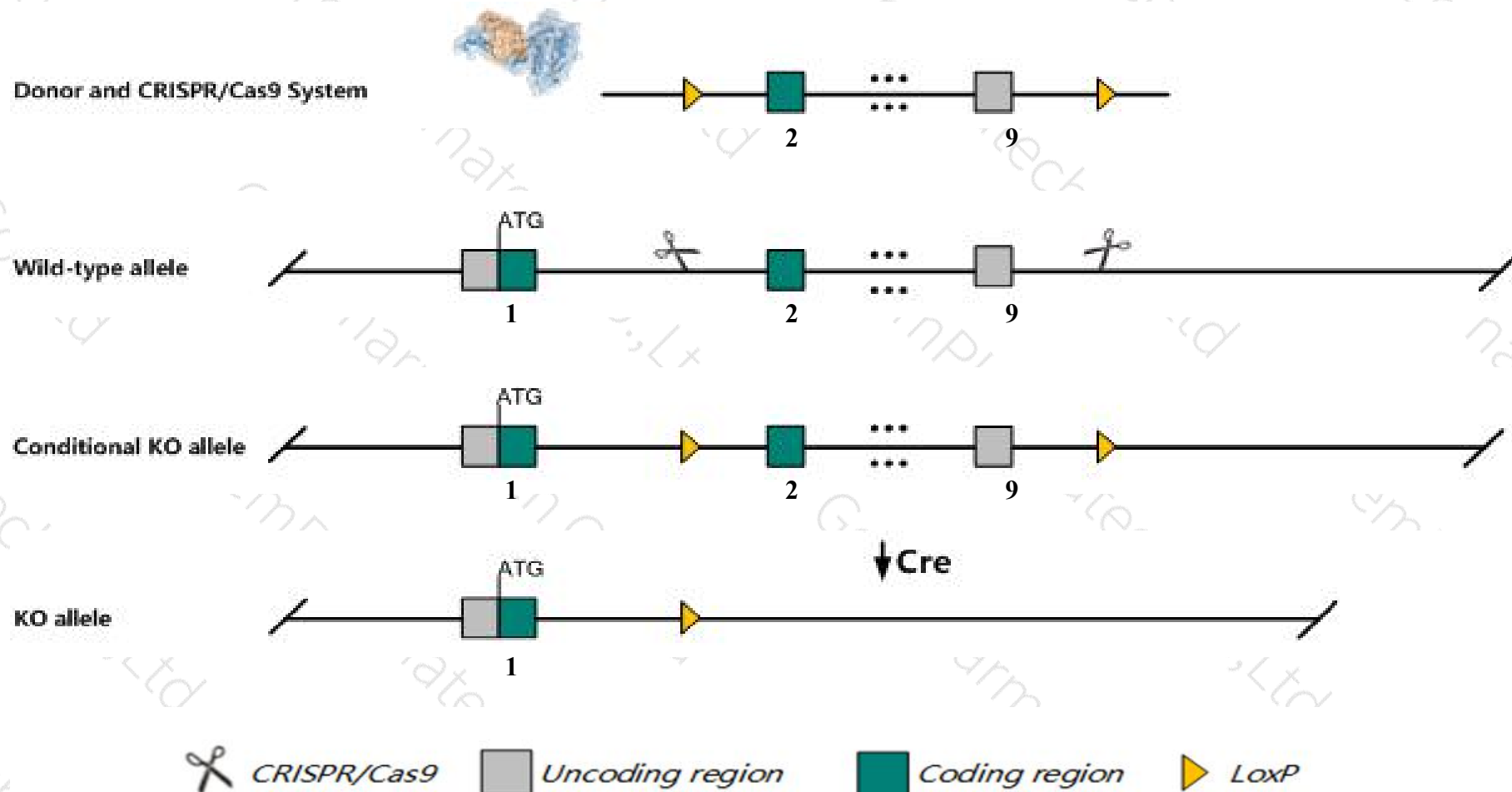
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gnat1* gene. The schematic diagram is as follows:



- The *Gnat1* gene has 7 transcripts. According to the structure of *Gnat1* gene, exon2-exon9 of *Gnat1*-201 (ENSMUST00000010205.8) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gnat1* 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 disruption of this gene display retinal degeneration with age and abnormal electrophysiology of the rods.
- The floxed region is near to the N-terminal of *Slc38a3* and *A930036K24Rik* gene, this strategy may influence the regulatory function of the N-terminal of these genes.
- The *Gnat1* gene is located on the Chr9. 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)

Gnat1 guanine nucleotide binding protein, alpha transducing 1 [*Mus musculus* (house mouse)]

Gene ID: 14685, updated on 10-Dec-2019

Summary

Official Symbol	Gnat1 provided by MGI
Official Full Name	guanine nucleotide binding protein, alpha transducing 1 provided by MGI
Primary source	MGI:MGI:95778
See related	Ensembl:ENSMUSG00000034837
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	Ird1; Ird2; irdc; irdr; Gnat-1; Tralpa; transducin
Expression	Biased expression in liver adult (RPKM 8.7) and liver E18 (RPKM 1.6) See more
Orthologs	human all

Genomic context

Location: 9 F1; 9 58.86 cM

See Gnat1 in [Genome Data Viewer](#)

Exon count: 9

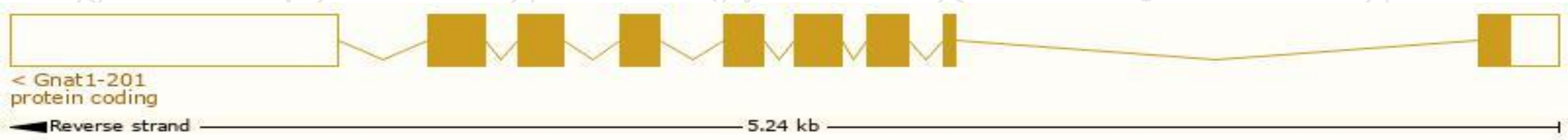
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	9	NC_000075.6 (107674437..107679634, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	9	NC_000075.5 (107576805..107581923, complement)

Transcript information (Ensembl)

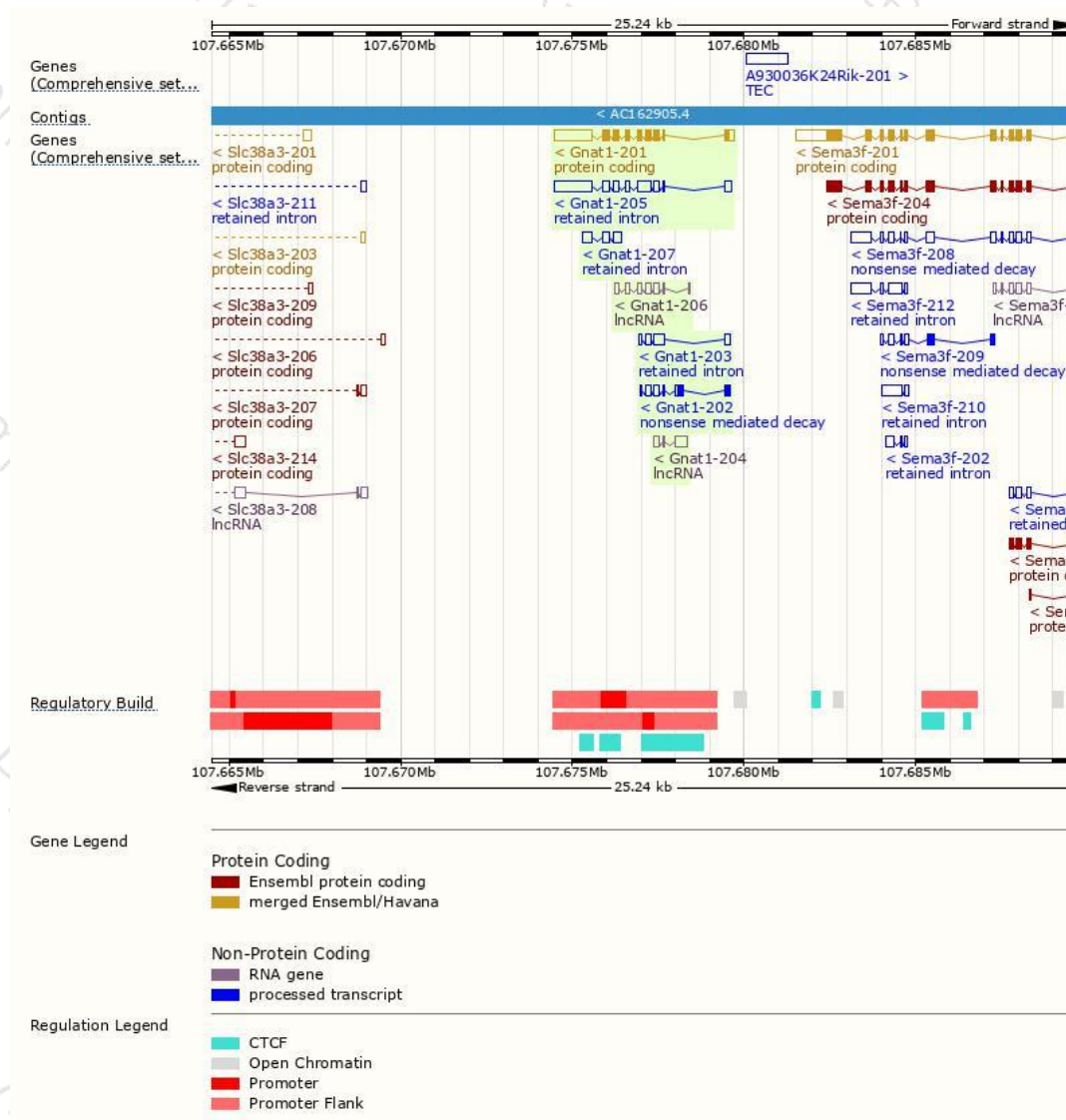
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gnat1-201	ENSMUST00000010205.8	2328	350aa	Protein coding	CCDS23504	P20612	TSL:1 GENCODE basic APPRIS P1
Gnat1-202	ENSMUST000000192271.5	782	87aa	Nonsense mediated decay	-	A0A0A6YWJ0	TSL:3
Gnat1-205	ENSMUST000000194802.5	2354	No protein	Retained intron	-	-	TSL:1
Gnat1-207	ENSMUST000000195849.1	725	No protein	Retained intron	-	-	TSL:2
Gnat1-203	ENSMUST000000193188.5	717	No protein	Retained intron	-	-	TSL:2
Gnat1-206	ENSMUST000000195129.5	754	No protein	lncRNA	-	-	TSL:3
Gnat1-204	ENSMUST000000194153.1	525	No protein	lncRNA	-	-	TSL:5

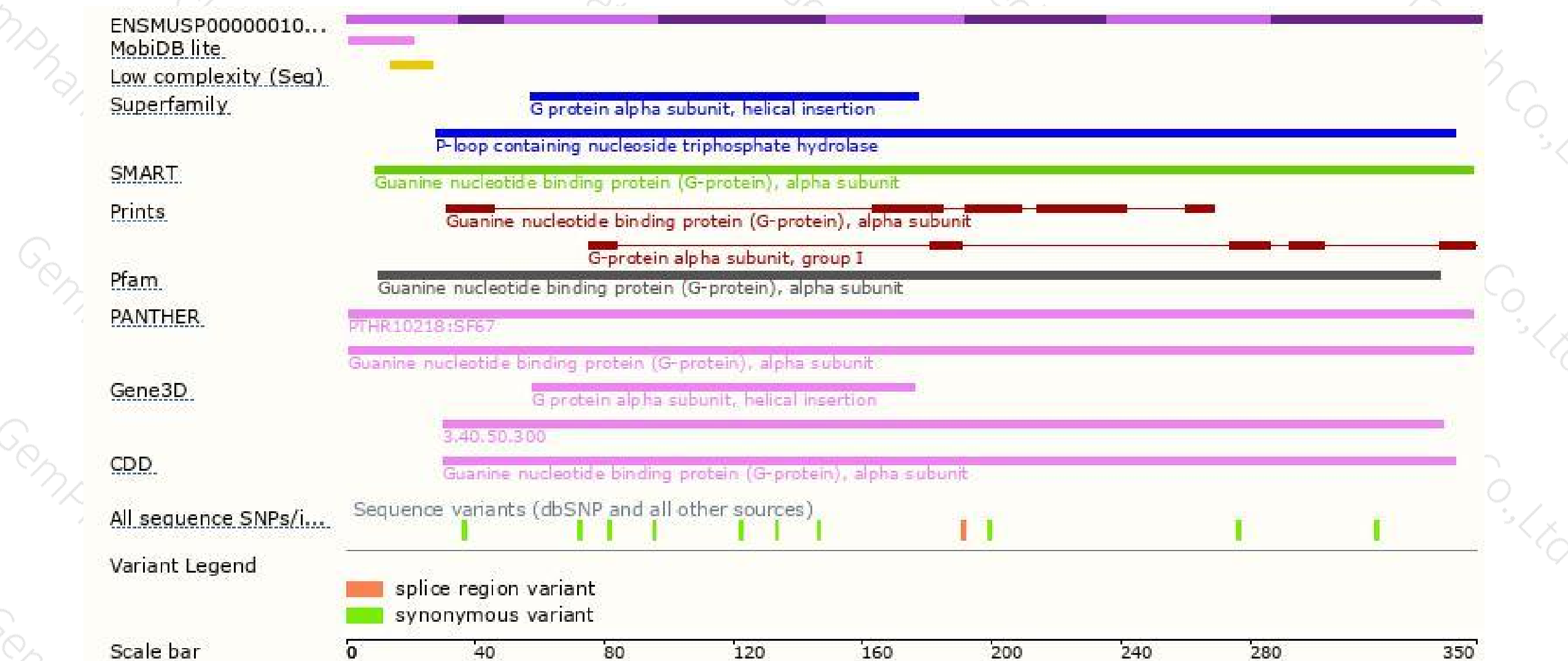
The strategy is based on the design of *Gnat1-201* transcript,The transcription is shown below



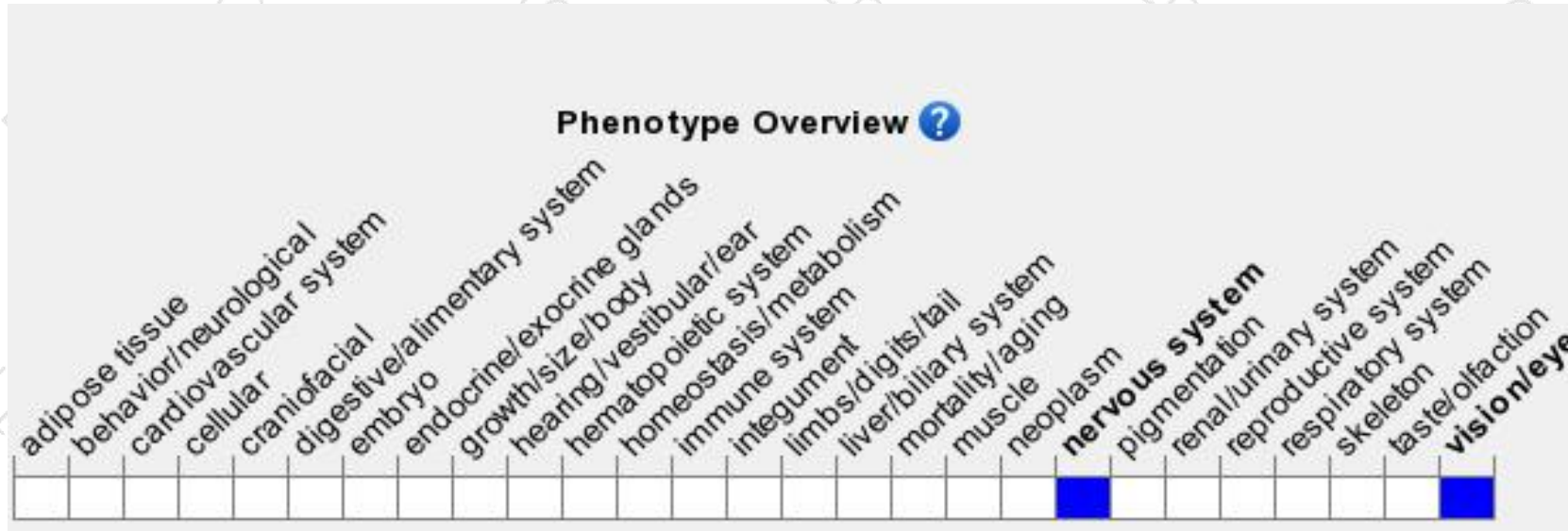
Genomic location distribution



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 disruption of this gene display retinal degeneration with age and abnormal electrophysiology of the rods.

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

Tel: 400-9660890

