Grm2 Cas9-CKO *Strategy*

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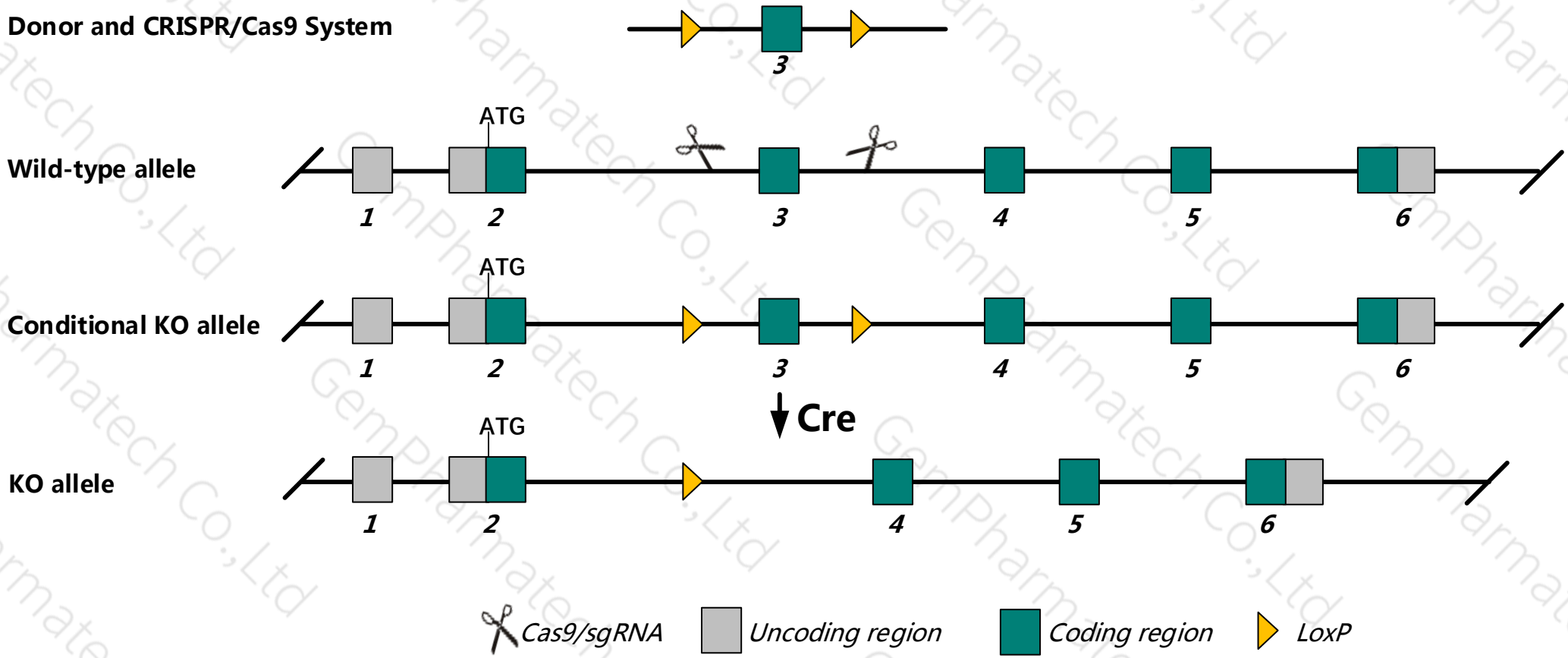
Project Overview

Project Name	***Grm2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Grm2* gene. The schematic diagram is as follows:



Technical routes

- The *Grm2* gene has 4 transcripts.According to the structure of *Grm2* gene, exon3 of *Grm2*-201 (ENSMUST00000023959.12) transcript is recommended as the knockout region.The region contains 838bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Grm2* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.

Notice

- According to the existing MGI data，mice homozygous for disruptions in this gene display subtile behavioral modifications and moderate abnormalities in long term depression and EPSP in the hippocampus.
- Transcript *Grm2-202* may not be affected.
- The *Grm2* 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

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Grm2 glutamate receptor, metabotropic 2 [*Mus musculus* (house mouse)]

Gene ID: 108068, updated on 12-Aug-2019

Summary

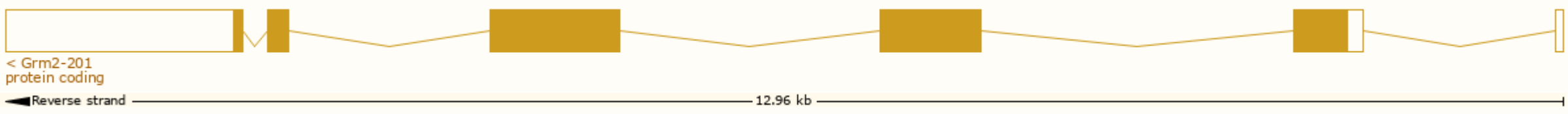
Official Symbol	Grm2 provided by MGI
Official Full Name	glutamate receptor, metabotropic 2 provided by MGI
Primary source	MGI:MGI:1351339
See related	Ensembl:ENSMUSG00000023192
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gprc1b; mGluR2; mGluR7; 4930441L02Rik
Expression	Biased expression in frontal lobe adult (RPKM 23.0), cortex adult (RPKM 16.8) and 6 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

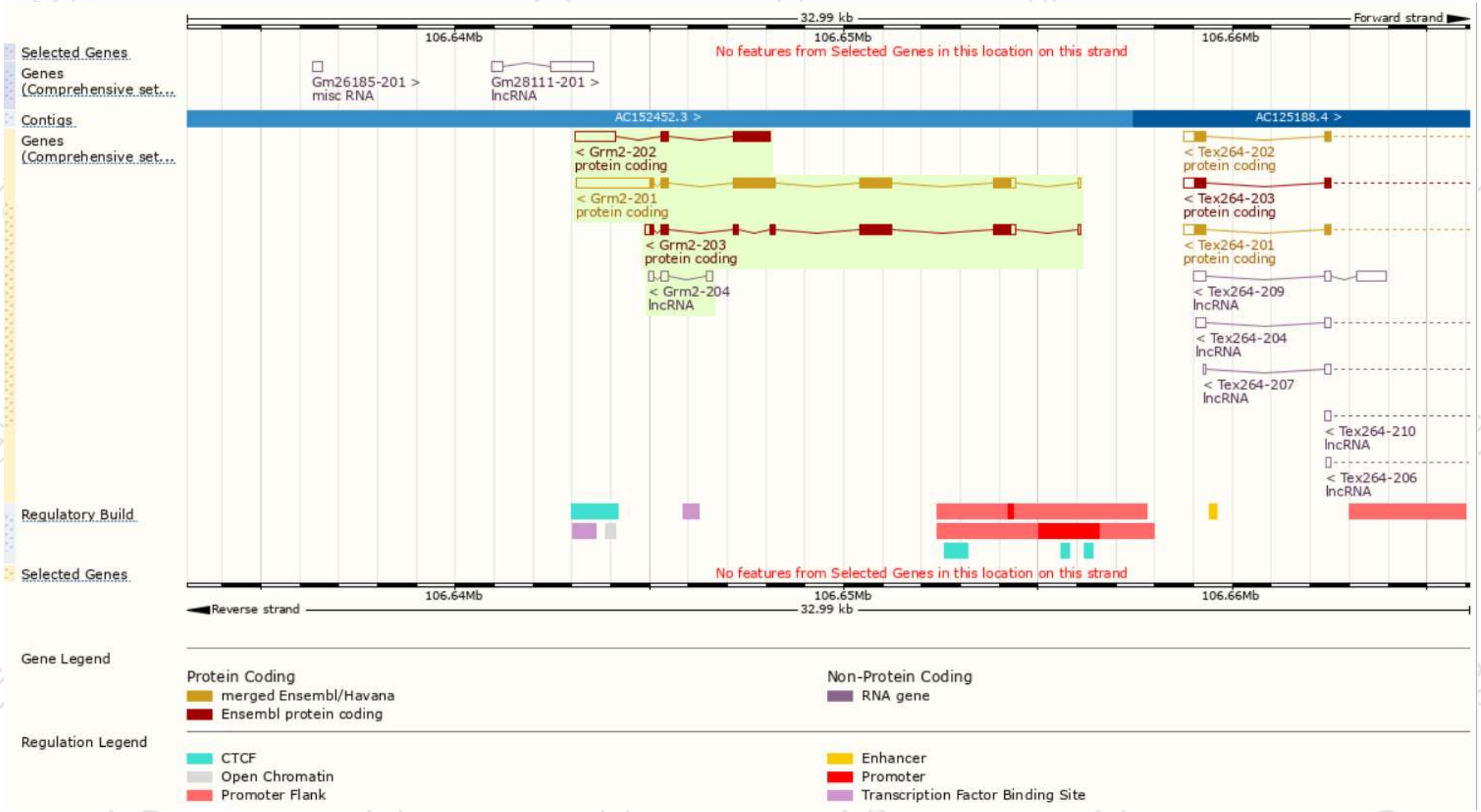
The gene has 4 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Grm2-201	ENSMUST00000023959.12	4711	872aa	Protein coding	CCDS52914	Q14BI2	TSL:5 GENCODE basic APPRIS P1
Grm2-203	ENSMUST00000201681.1	2111	594aa	Protein coding	-	A0A0J9YVF0	TSL:1 GENCODE basic
Grm2-202	ENSMUST00000200826.3	2181	381aa	Protein coding	-	A0A0J9YU95	CDS 5' incomplete TSL:1
Grm2-204	ENSMUST00000201955.1	448	No protein	lncRNA	-	-	TSL:3

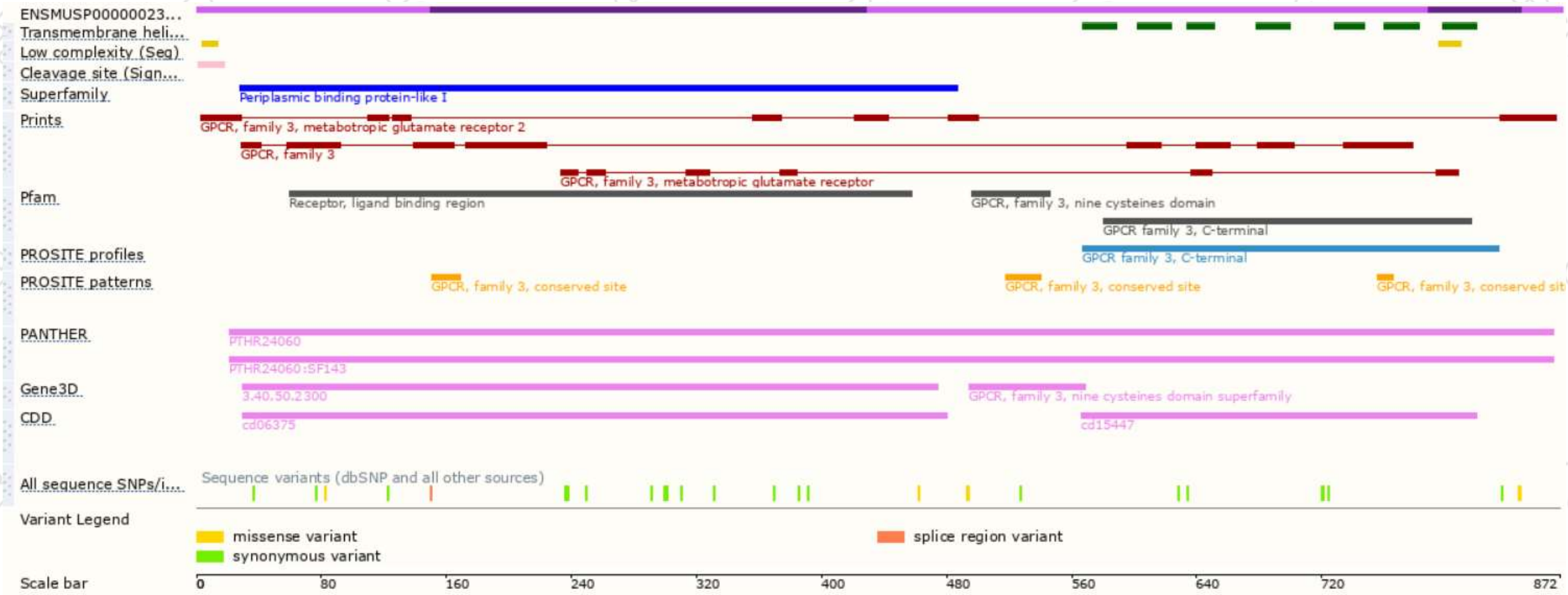
The strategy is based on the design of *Grm2*-201 transcript,The transcription is shown below



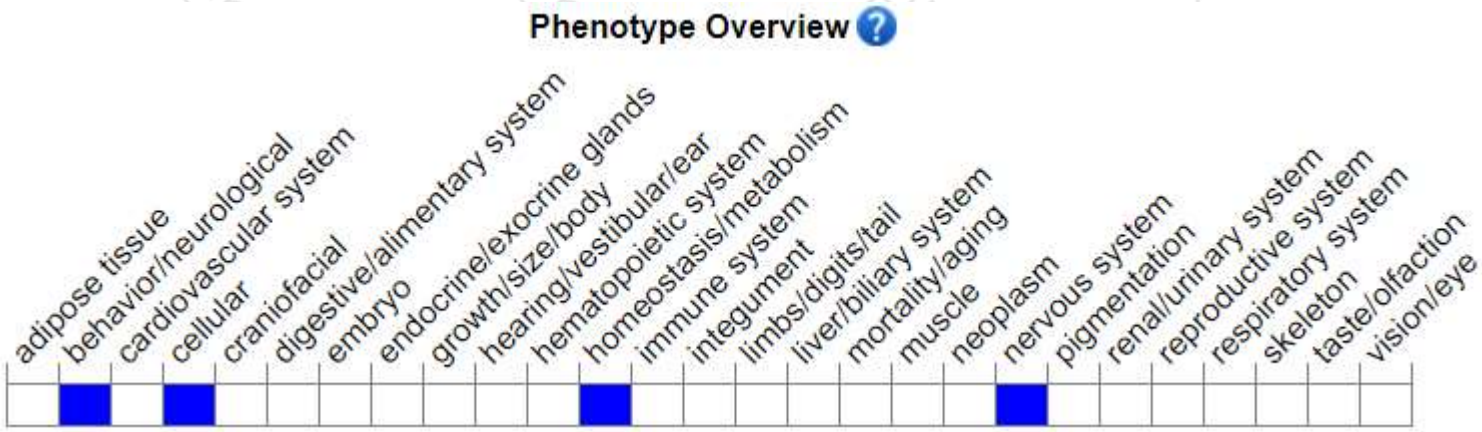
Genomic location distribution



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/) .

Mice homozygous for disruptions in this gene display subtile behavioral modifications and moderate abnormalities in long term depression and EPSP in the hippocampus.

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

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