# *Clcn3* Cas9-KO Strategy

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# Project Overview

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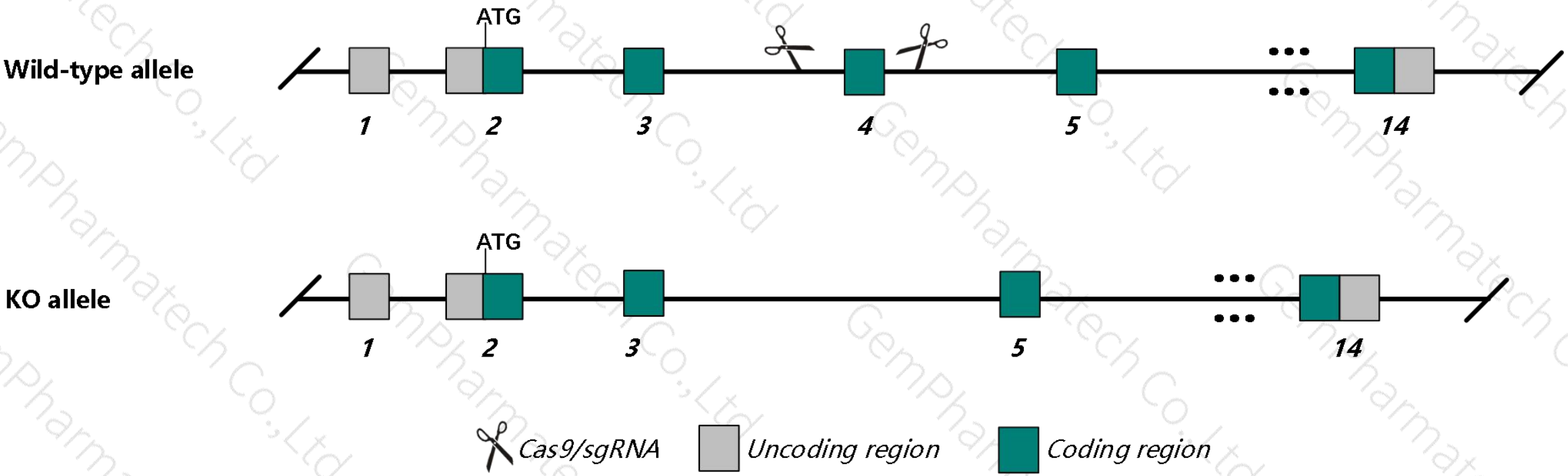
|---|---|
| **Project Name** | ***Clcn3*** |
| **Project type** | **Cas9-KO** |
| **Strain background** | **C57BL/6JGpt** |

# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Clcn3* gene. The schematic diagram is as follows:



# Technical routes

➢ The *Clcn3* gene has 10 transcripts. According to the structure of *Clcn3* gene, exon4 of *Clcn3-201* (ENSMUST00000004430.13) transcript is recommended as the knockout region.The region contains 100bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Clcn3* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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.

# Notice



- According to the existing MGI data,Nullizygous mutations cause degeneration of hippocampal neurons and retinal photoreceptors, reduced body weight, behavioral deficits, gliosis, kyphosis and premature death, and may alter male fertility, ileum morphology, liver physiology, seizure susceptibility, and behavioral response to drugs.

- The *Clcn3* gene is located on the Chr8. 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 gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

# Gene information（NCBI）

集萃药康 GemPharmatech

## Clcn3 chloride channel, voltage-sensitive 3 [Mus musculus (house mouse)]

Gene ID: 12725, updated on 28-Mar-2019

### Summary

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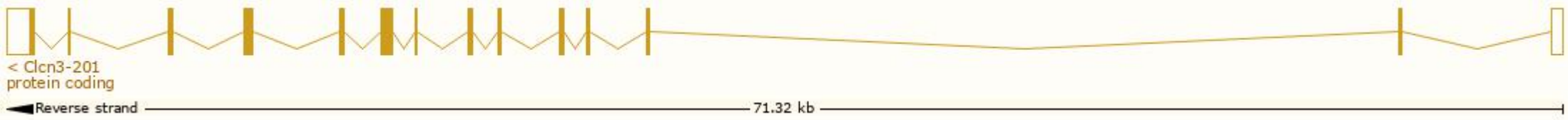
|---|---|
| **Official Symbol** | Clcn3 provided by MGI |
| **Official Full Name** | chloride channel, voltage-sensitive 3 provided by MGI |
| **Primary source** | MGI:MGI:103555 |
| **See related** | Ensembl:ENSMUSG00000004319 |
| **Gene type** | protein coding |
| **RefSeq status** | VALIDATED |
| **Organism** | Mus musculus |
| **Lineage** | Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus |
| **Also known as** | Clc3 |
| **Expression** | Ubiquitous expression in cerebellum adult (RPKM 11.4), frontal lobe adult (RPKM 10.6) and 28 other tissues See more |
| **Orthologs** | human all |

# Transcript information（Ensembl）

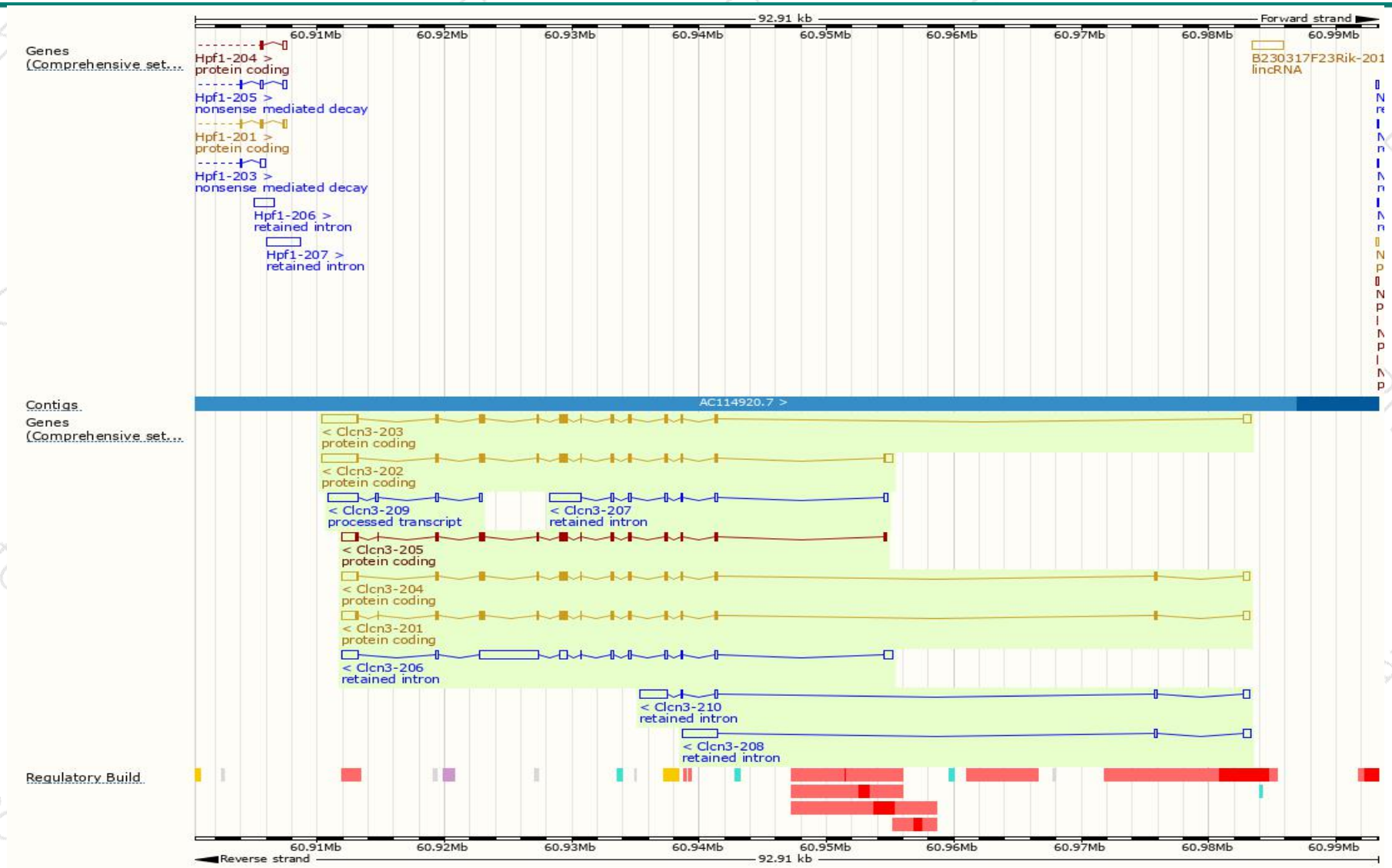
The gene has 10 transcripts,all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|
| Clcn3-202 | ENSMUST00000056508.11 | 5684 | 791aa | Protein coding | CCDS22322 | Q8K4W8 | TSL:1 GENCODE basic APPRIS P3 |
| Clcn3-203 | ENSMUST00000093490.8 | 5518 | 760aa | Protein coding | CCDS22323 | P51791 Q790S0 | TSL:1 GENCODE basic |
| Clcn3-201 | ENSMUST00000004430.13 | 4175 | 866aa | Protein coding | CCDS52555 | Q3TF45 | TSL:1 GENCODE basic APPRIS ALT1 |
| Clcn3-204 | ENSMUST00000110301.1 | 4099 | 818aa | Protein coding | CCDS52554 | P51791 | TSL:1 GENCODE basic APPRIS ALT1 |
| Clcn3-205 | ENSMUST00000110302.7 | 3768 | 839aa | Protein coding | - | E9Q2I1 | TSL:5 GENCODE basic APPRIS ALT1 |
| Clcn3-209 | ENSMUST00000145741.7 | 2954 | No protein | Processed transcript | - | - | TSL:3 |
| Clcn3-206 | ENSMUST00000129672.7 | 8193 | No protein | Retained intron | - | - | TSL:1 |
| Clcn3-207 | ENSMUST00000132234.7 | 3480 | No protein | Retained intron | - | - | TSL:1 |
| Clcn3-208 | ENSMUST00000145493.1 | 3462 | No protein | Retained intron | - | - | TSL:1 |
| Clcn3-210 | ENSMUST00000147824.7 | 3034 | No protein | Retained intron | - | - | TSL:1 |

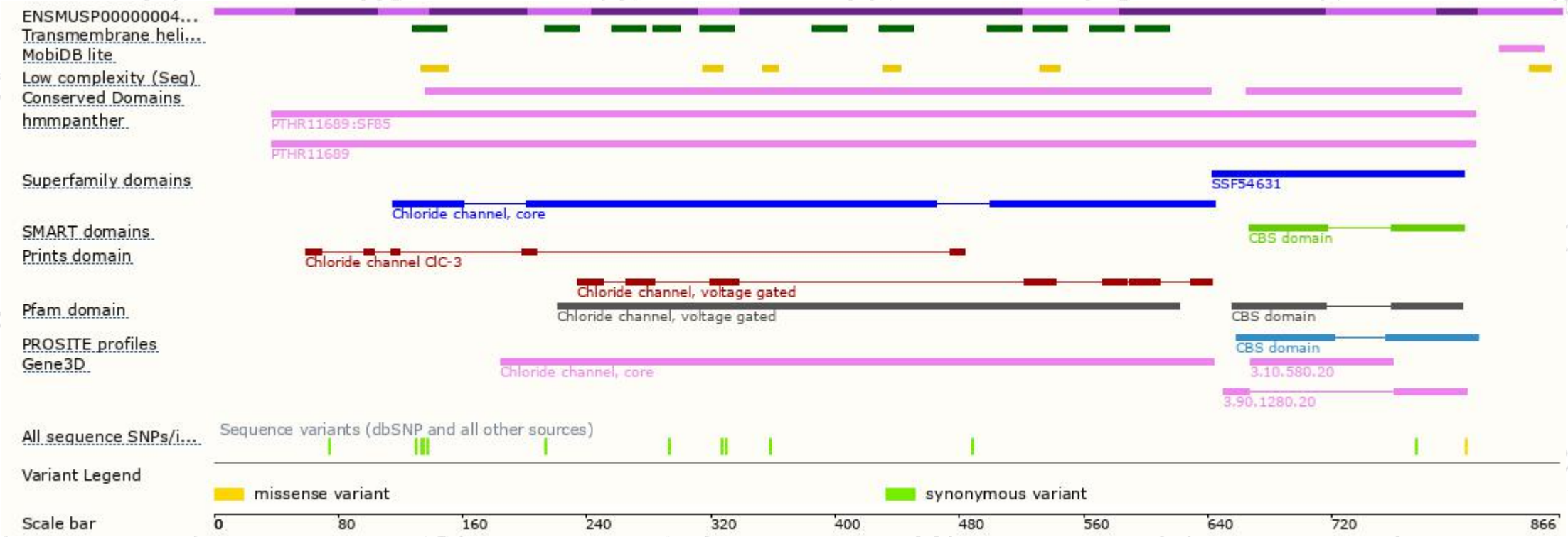
The strategy is based on the design of *Clcn3-201* transcript,The transcription is shown below



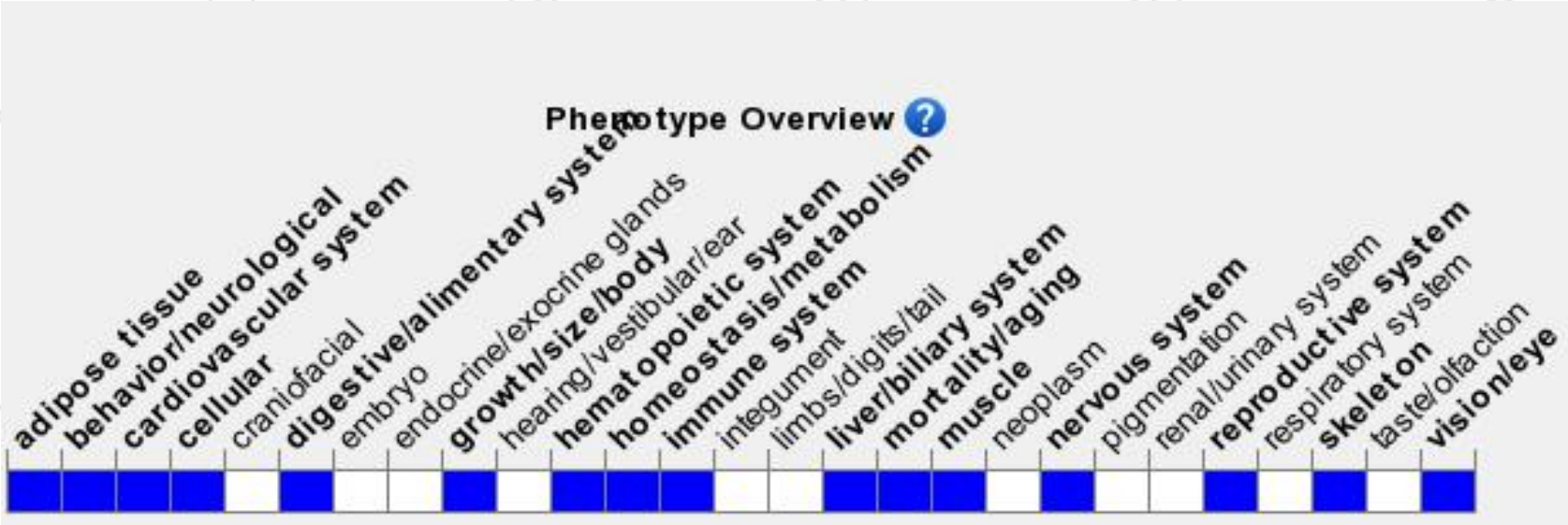
# Genomic location distribution



# 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,Nullizygous mutations cause degeneration of hippocampal neurons and retinal photoreceptors, reduced body weight, behavioral deficits, gliosis, kyphosis and premature death, and may alter male fertility, ileum morphology, liver physiology, seizure susceptibility, and behavioral response to drugs.

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

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