# *Usp14* Cas9-KO Strategy

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**Reviewer:** Yang Zeng

**Design Date:** 2018-6-8

# Project Overview

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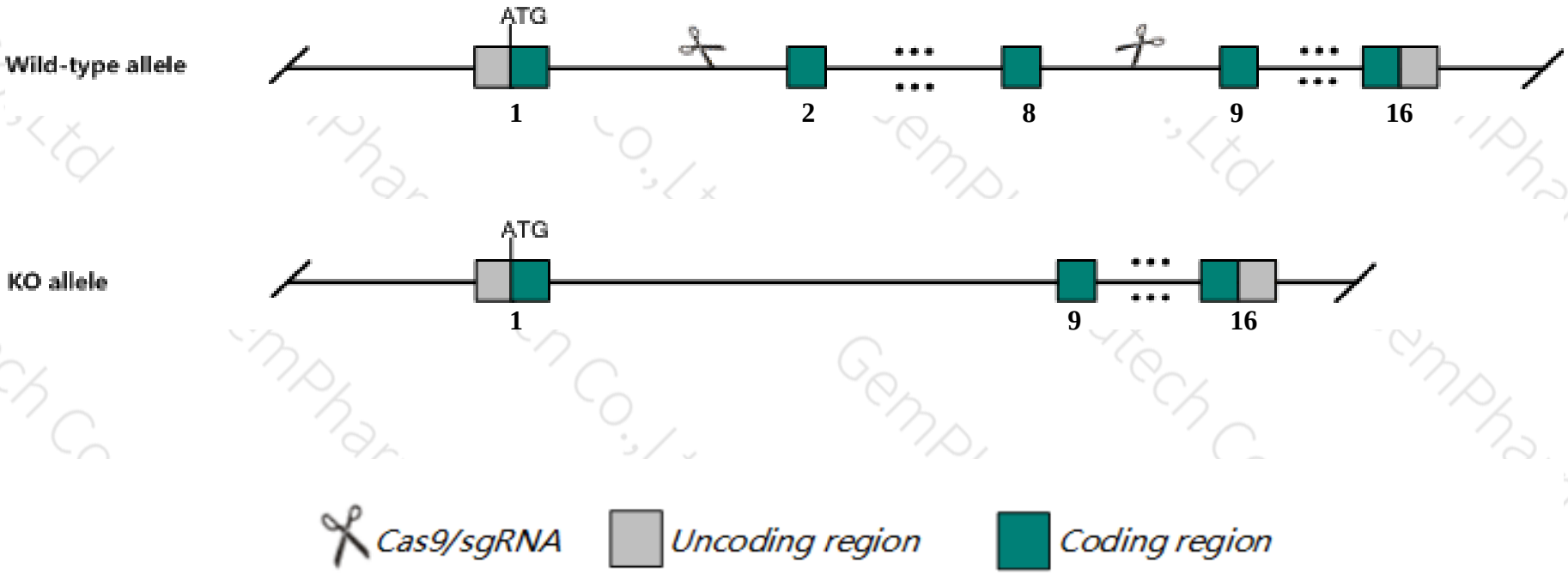
|---|---|
| **Project Name** | ***Usp14*** |
| **Project type** | **Cas9-KO** |
| **Strain background** | **C57BL/6JGpt** |

# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Usp14* gene. The schematic diagram is as follows:



# Technical routes

➢ The *Usp14* gene has 10 transcripts. According to the structure of *Usp14* gene, exon2-exon8 of *Usp14-201* (ENSMUST00000092096.13) transcript is recommended as the knockout region.The region contains 659bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Usp14* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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,Homozygotes for a hypomorphic mutation develop severe tremors by 3 weeks of age, followed by hindlimb paralysis and premature death. An underdeveloped corpus callosum, hippocampus, dentate gyrus and forebrain structures, and notable defects in synaptic transmission in both the CNS and PNS are seen.

- Transcript *Usp14-204* may not be affected.

- The *Usp14* gene is located on the Chr18. 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

## Usp14 ubiquitin specific peptidase 14 [ *Mus musculus* (house mouse) ]

Gene ID: 59025, updated on 5-Mar-2019

### Summary

| | |
|---|---|
| **Official Symbol** | Usp14 provided by MGI |
| **Official Full Name** | ubiquitin specific peptidase 14 provided by MGI |
| **Primary source** | MGI:MGI:1928898 |
| **See related** | Ensembl:ENSMUSG00000047879 |
| **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** | ax; C78769; nmf375; AW107924; 2610005K12Rik; 2610037B11Rik |
| **Expression** | Broad expression in CNS E18 (RPKM 14.6), cortex adult (RPKM 14.0) and 27 other tissues See more |
| **Orthologs** | human all |

# Transcript information（Ensembl）

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

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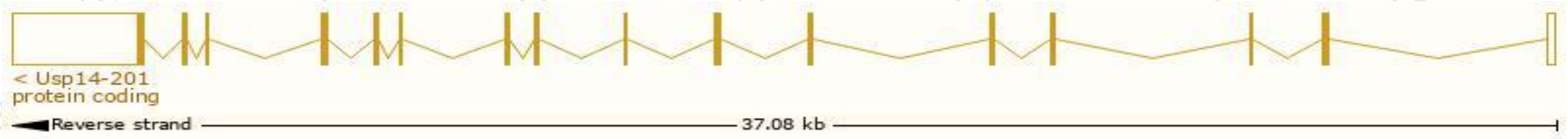
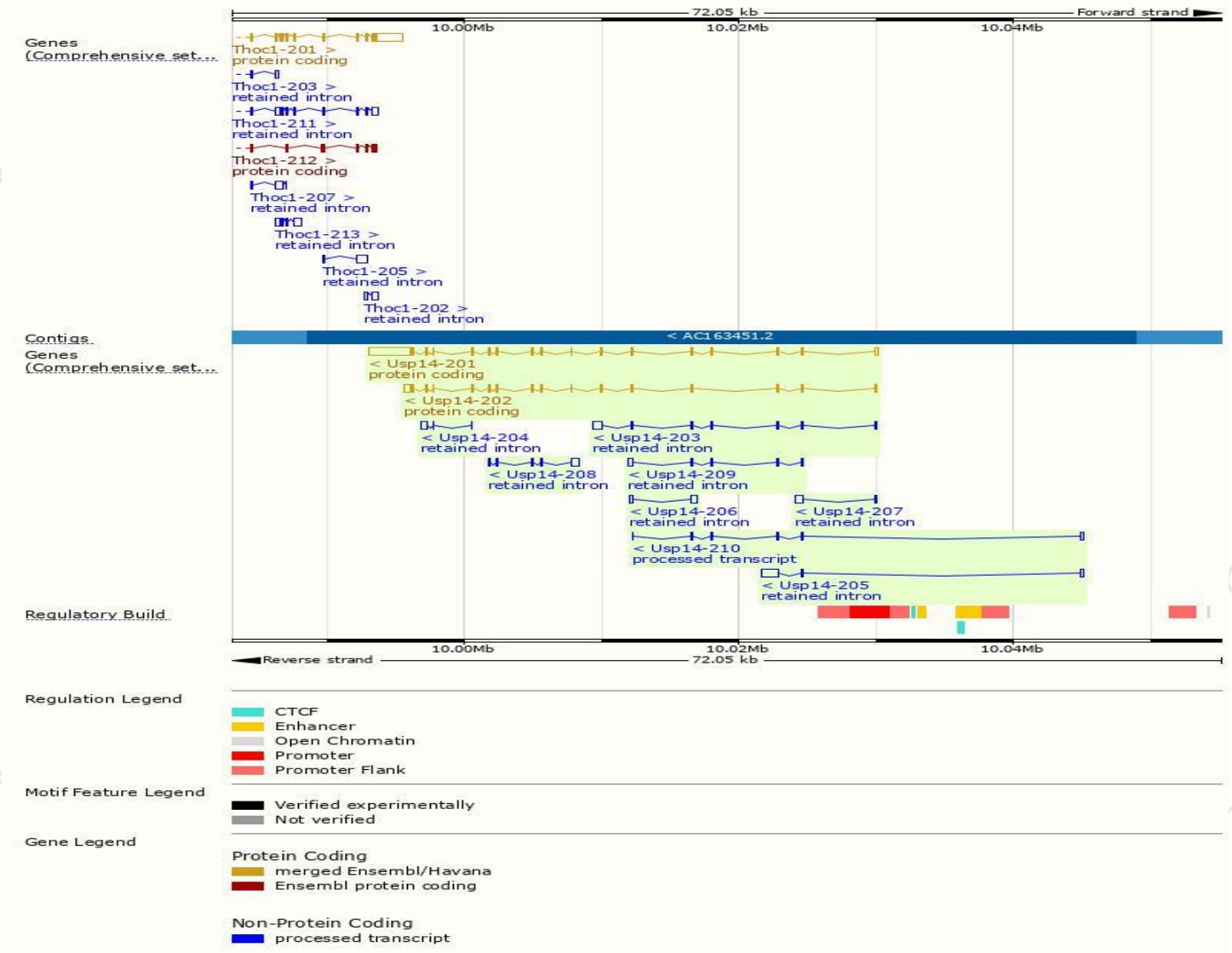
| Name | Transcript ID | bp | Protein | Translation ID | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|---|
| Usp14-201 | ENSMUST00000092096.13 | 4726 | 493aa | ENSMUSP00000089728.6 | Protein coding | CCDS37735 | Q9JMA1 | TSL:1 GENCODE basic APPRIS P1 |
| Usp14-202 | ENSMUST00000116669.1 | 2019 | 458aa | ENSMUSP00000112368.1 | Protein coding | CCDS37734 | E9PYI8 | TSL:1 GENCODE basic |
| Usp14-203 | ENSMUST00000128334.7 | 1346 | No protein | - | Retained intron | - | - | TSL:1 |
| Usp14-204 | ENSMUST00000133594.1 | 477 | No protein | - | Retained intron | - | - | TSL:5 |
| Usp14-205 | ENSMUST00000142013.1 | 1484 | No protein | - | Retained intron | - | - | TSL:1 |
| Usp14-206 | ENSMUST00000145929.1 | 656 | No protein | - | Retained intron | - | - | TSL:1 |
| Usp14-207 | ENSMUST00000150321.1 | 639 | No protein | - | Retained intron | - | - | TSL:2 |
| Usp14-208 | ENSMUST00000154088.1 | 989 | No protein | - | Retained intron | - | - | TSL:3 |
| Usp14-209 | ENSMUST00000234165.1 | 664 | No protein | - | Retained intron | - | - | - |
| Usp14-210 | ENSMUST00000234243.1 | 612 | No protein | - | lncRNA | - | - | - |

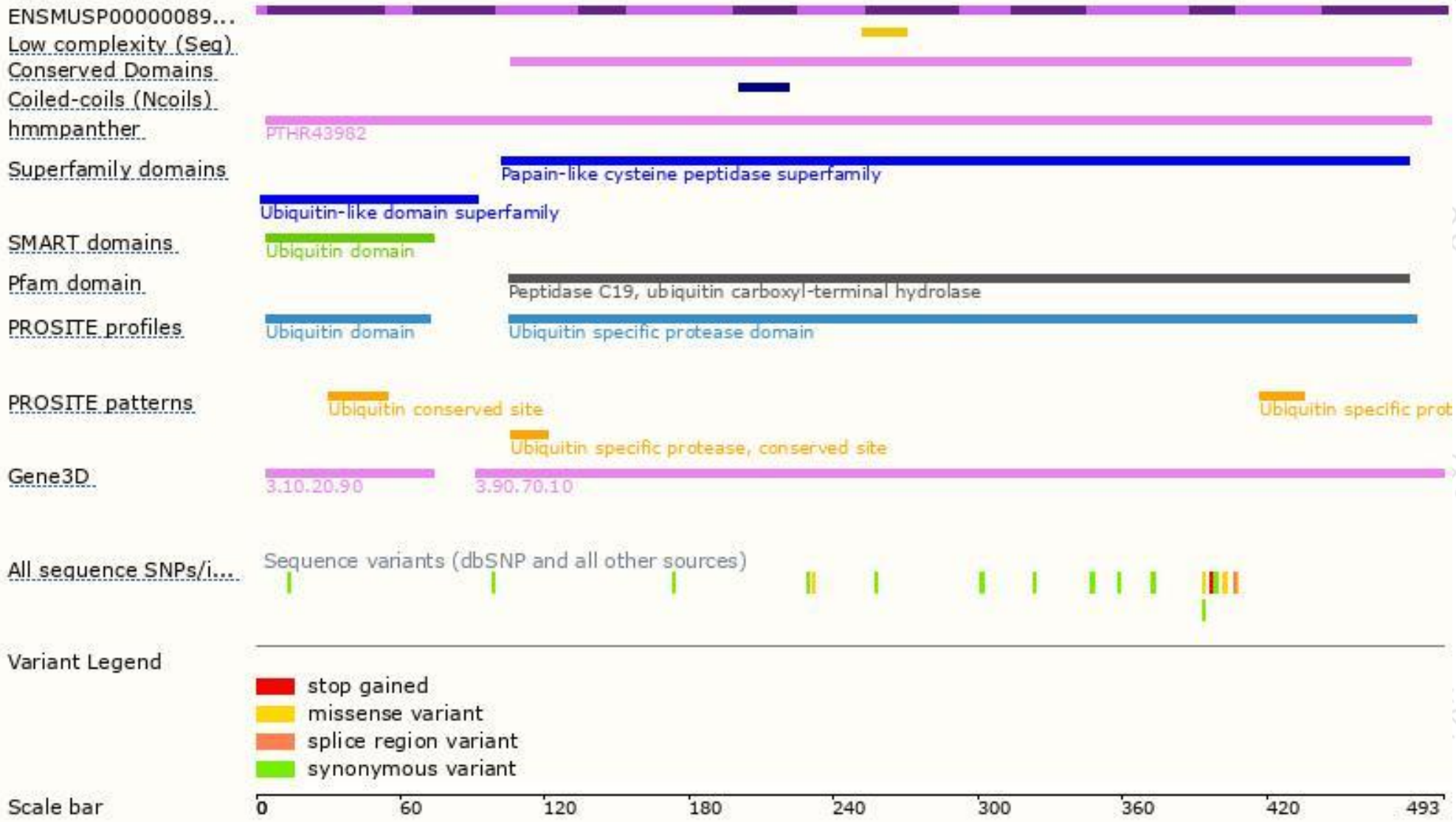
The strategy is based on the design of *Usp14-201* transcript,The transcription is shown below



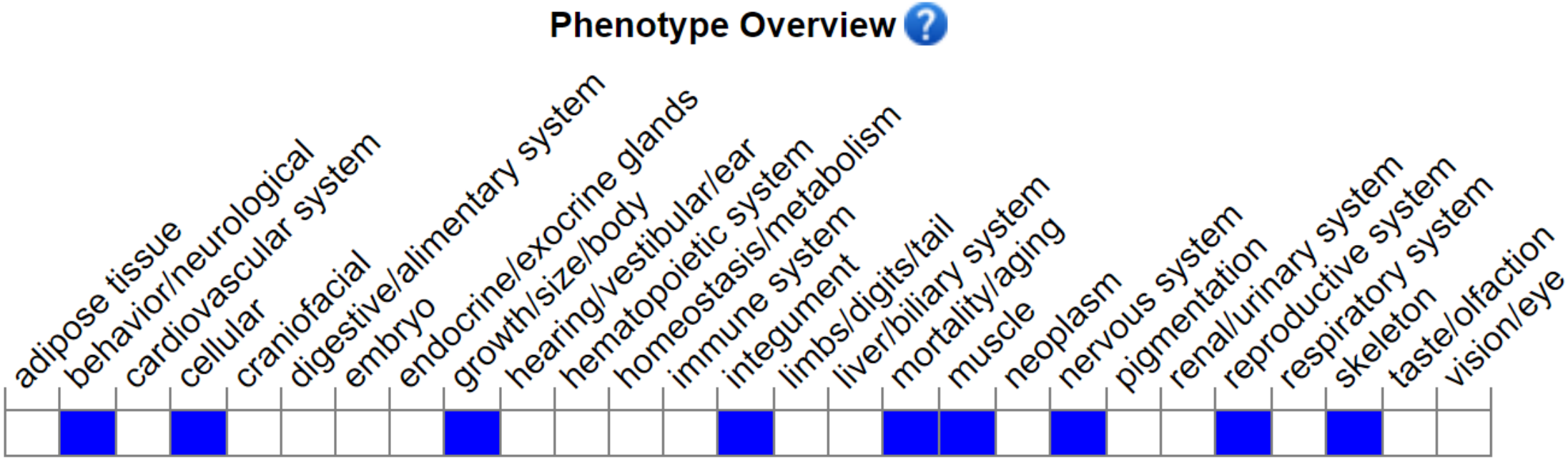
# Genomic location distribution



# 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,Homozygotes for a hypomorphic mutation develop severe tremors by 3 weeks of age, followed by hindlimb paralysis and premature death. An underdeveloped corpus callosum, hippocampus, dentate gyrus and forebrain structures, and notable defects in synaptic transmission in both the CNS and PNS are seen.

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

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