Sez6 Cas9-CKO Strategy

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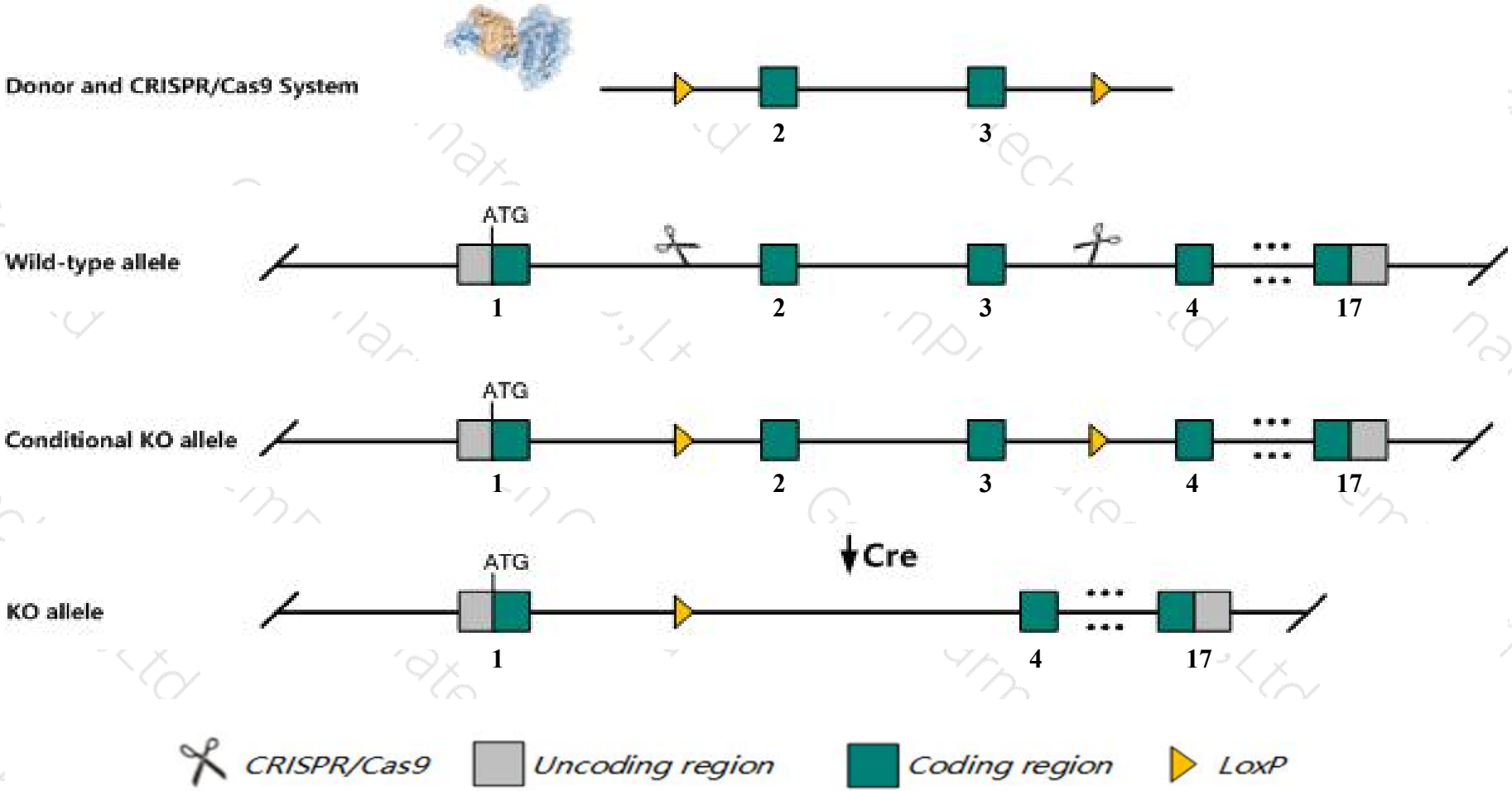
Date:2020-02-06

Project Overview

Project Name	***Sez6***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sez6* gene. The schematic diagram is as follows:



Technical routes

- The *Sez6* gene has 9 transcripts. According to the structure of *Sez6* gene, exon2-exon3 of *Sez6-202* (ENSMUST00000093995.9) transcript is recommended as the knockout region.The region contains 794bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Sez6* 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.

Notice

- According to the existing MGI data,Mice homozygous for a null allele exhibit increased short dendrites, decreased excitatory synaptic signaling, resistance to pharmacologically induces seizures, decreased activity and impaired learning and coordination.
- The effect on transcript *Sez6*-206&208 is unknown.
- Transcript *Sez6*-203 may not be affected.
- The *Sez6* gene is located on the Chr11. 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）

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Sez6 seizure related gene 6 [*Mus musculus* (house mouse)]

Gene ID: 20370, updated on 12-Aug-2019

Summary

Official Symbol	Sez6 provided by MGI
Official Full Name	seizure related gene 6 provided by MGI
Primary source	MGI:MGI:104745
See related	Ensembl:ENSMUSG00000000632
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	sez-6; BSRP-C; D11Bhm177e
Expression	Biased expression in whole brain E14.5 (RPKM 47.2), CNS E14 (RPKM 47.1) and 5 other tissues See more
Orthologs	human all

Genomic context

Location: 11 B5; 11 46.74 cM

See Sez6 in Genome Data Viewer

Exon count: 19

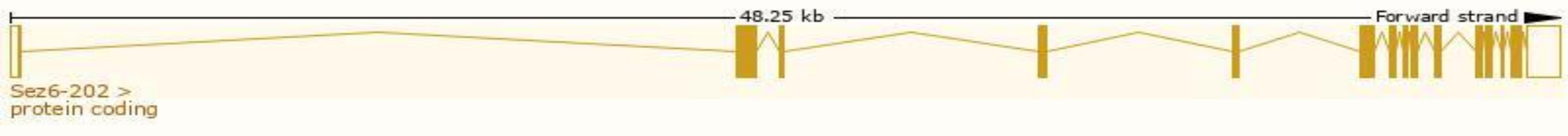
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	11	NC_000077.6 (77930587..77979052)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	11	NC_000077.5 (77744445..77792550)

Transcript information（Ensembl）

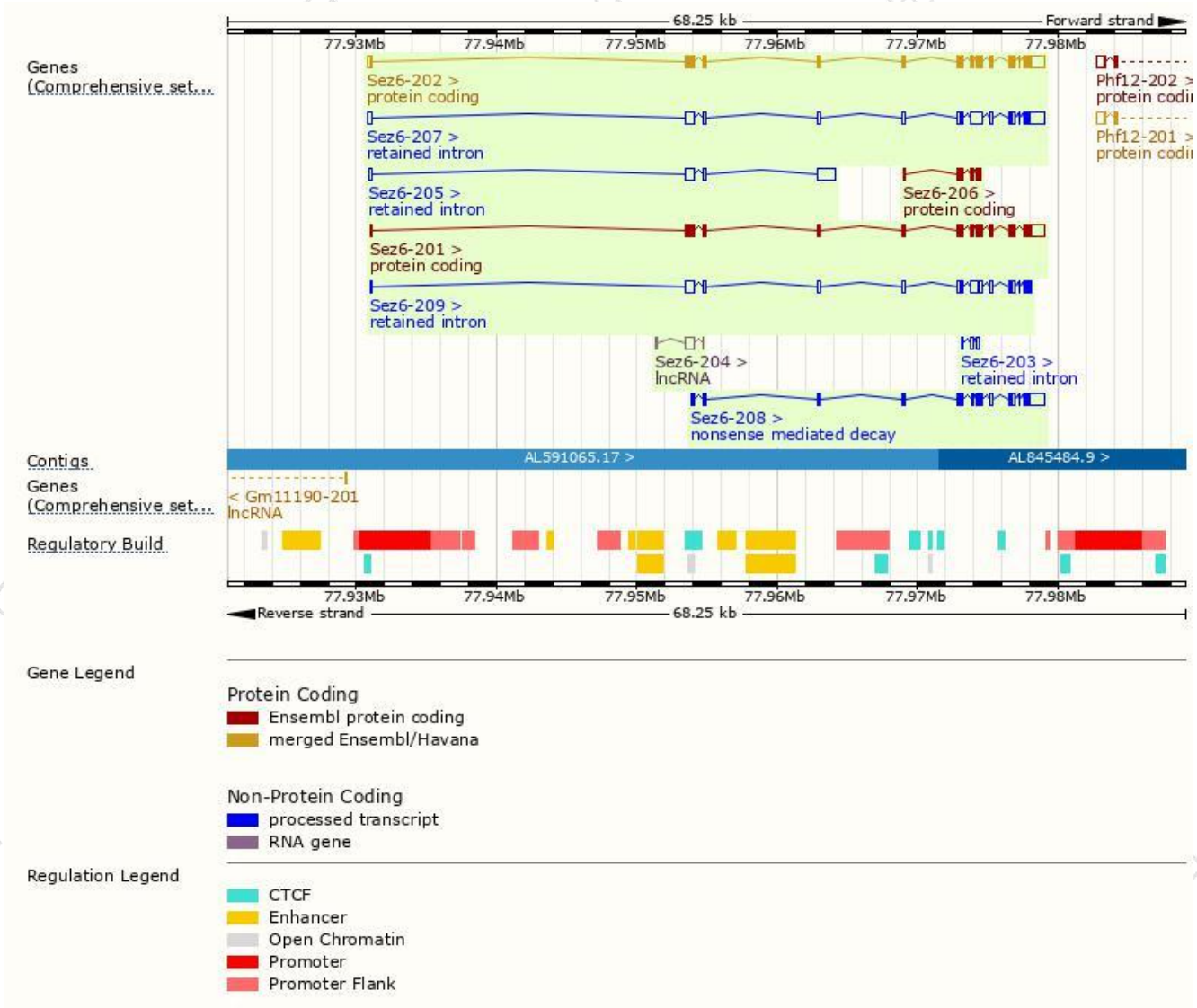
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sez6-202	ENSMUST00000093995.9	4239	991aa	Protein coding	CCDS25087	Q7TSK2	TSL:1 GENCODE basic APPRIS P3
Sez6-201	ENSMUST00000000646.13	3964	977aa	Protein coding	CCDS70255	Q7TSK2	TSL:1 GENCODE basic APPRIS ALT2
Sez6-206	ENSMUST00000140630.1	832	278aa	Protein coding	-	F6SXT0	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Sez6-208	ENSMUST00000151982.7	3424	452aa	Nonsense mediated decay	-	F6XNR8	CDS 5' incomplete TSL:5
Sez6-207	ENSMUST00000142542.7	4523	No protein	Retained intron	-	-	TSL:2
Sez6-209	ENSMUST00000155087.7	3226	No protein	Retained intron	-	-	TSL:2
Sez6-205	ENSMUST00000138749.7	2229	No protein	Retained intron	-	-	TSL:1
Sez6-203	ENSMUST00000126866.1	467	No protein	Retained intron	-	-	TSL:3
Sez6-204	ENSMUST00000138346.1	813	No protein	lncRNA	-	-	TSL:5

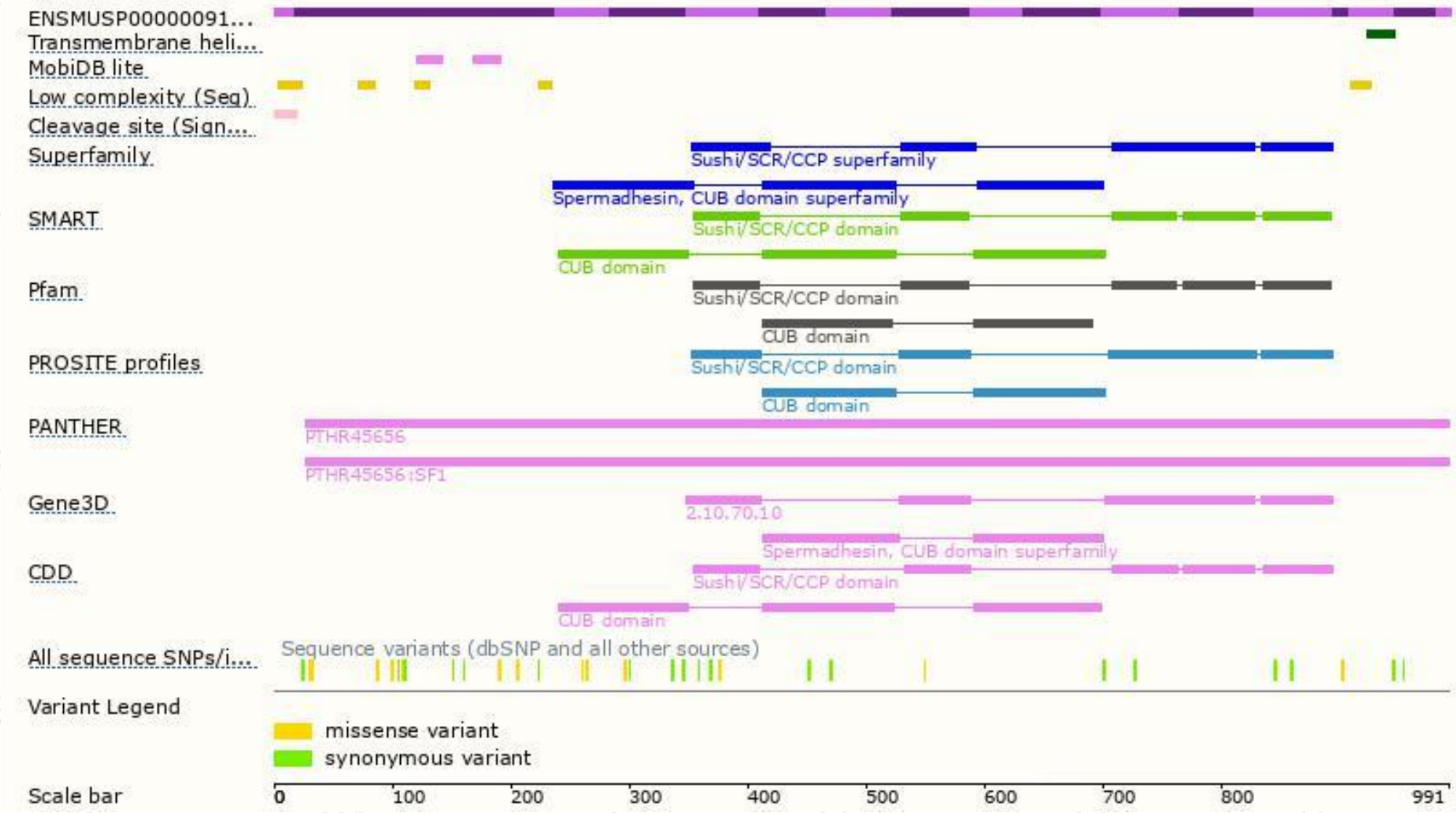
The strategy is based on the design of *Sez6-202* transcript,The transcription is shown below



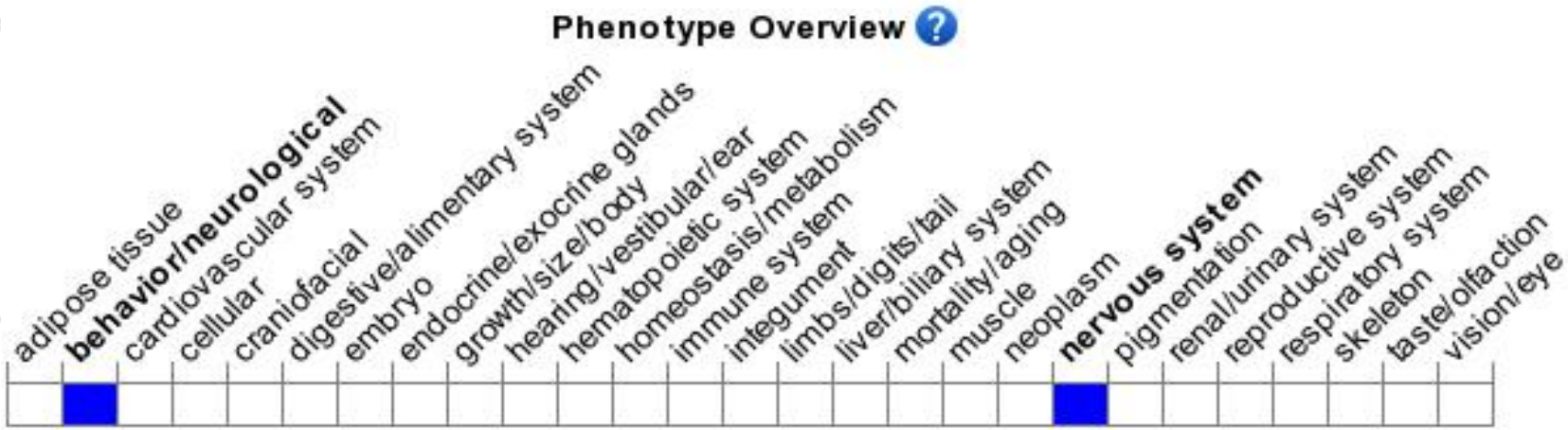
Genomic location distribution



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/).

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

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