Cntn2 Cas9-CKO Strategy

Designer: Daohua Xu

Reviewer: Huimin Su

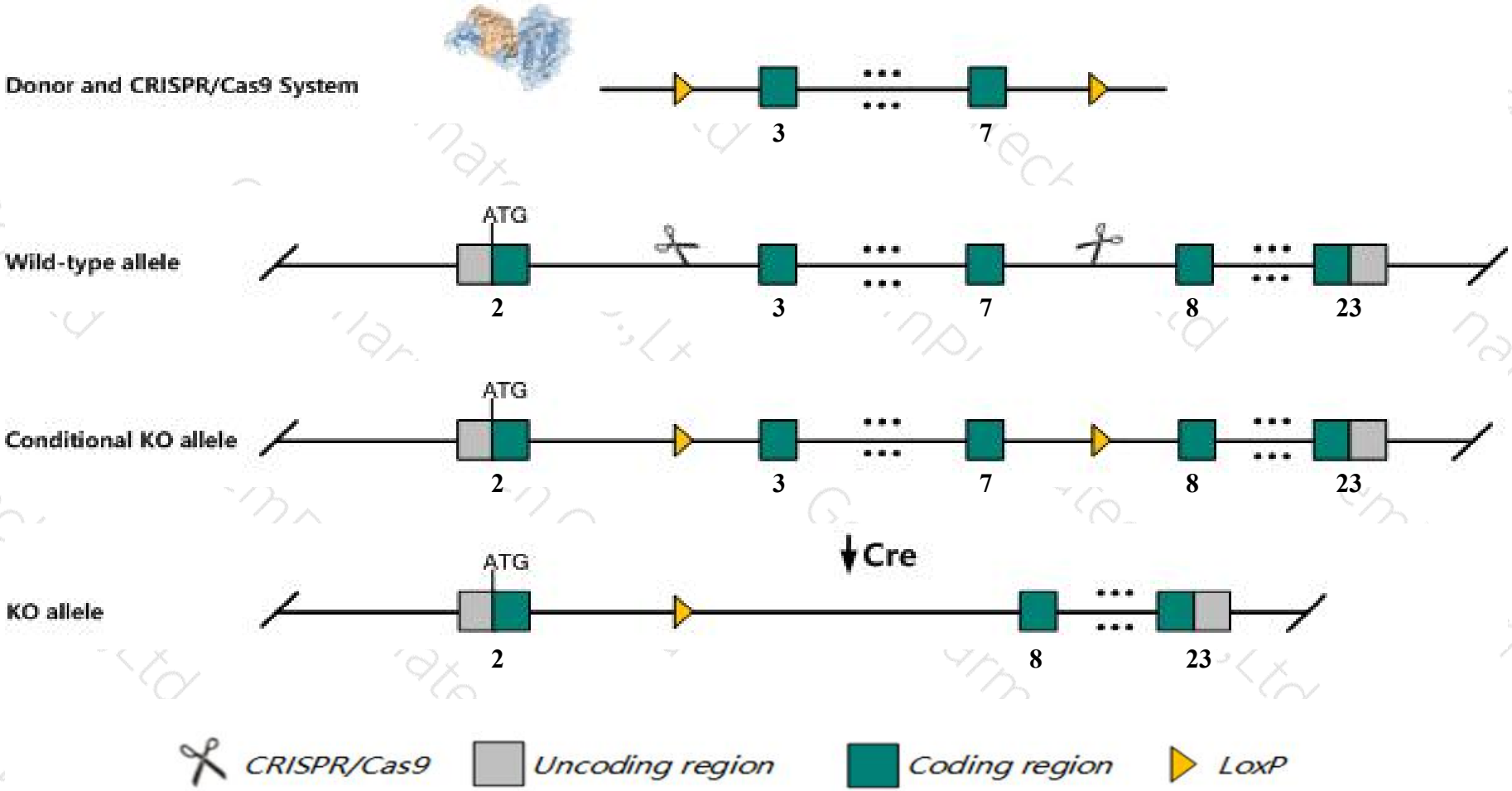
Design Date: 2019-9-25

Project Overview

Project Name	***Cntn2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cntn2* gene. The schematic diagram is as follows:



Technical routes

- The *Cntn2* gene has 8 transcripts. According to the structure of *Cntn2* gene, exon3-exon7 of *Cntn2-201* (ENSMUST00000086521.10) transcript is recommended as the knockout region.The region contains 727bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Cntn2* 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,Targeted mutation of this locus results in molecular abnormalities in the central nervous system.

- The *Cntn2* gene is located on the Chr1. 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）

集萃药康 GemPharmatech

Cntn2 contactin 2 [Mus musculus (house mouse)]

Gene ID: 21367, updated on 17-Feb-2019

Summary

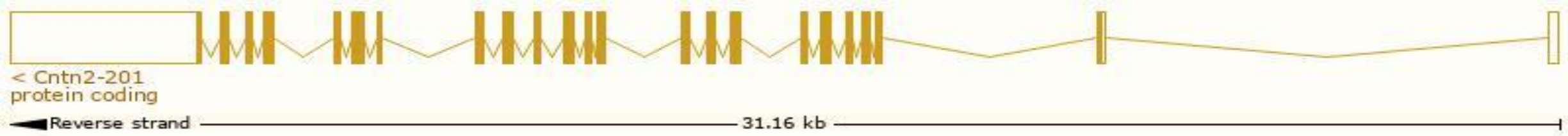
Official Symbol	Cntn2 provided by MGI
Official Full Name	contactin 2 provided by MGI
Primary source	MGI:MGI:104518
See related	Ensembl:ENSMUSG00000053024
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	D130012K04Rik, TAG-1, TAG1, Tax
Summary	This gene encodes a member of the contactin family of proteins, part of the immunoglobulin superfamily of cell adhesion molecules. The encoded glycosylphosphatidylinositol (GPI)-anchored neuronal membrane protein plays a role in the proliferation, migration, and axon guidance of neurons of the developing cerebellum. Mice lacking a functional copy of this gene exhibit epileptic seizures and elevated expression of A1 adenosine receptors. [provided by RefSeq, Sep 2016]
Expression	Biased expression in cerebellum adult (RPKM 27.9), CNS E14 (RPKM 19.9) and 5 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

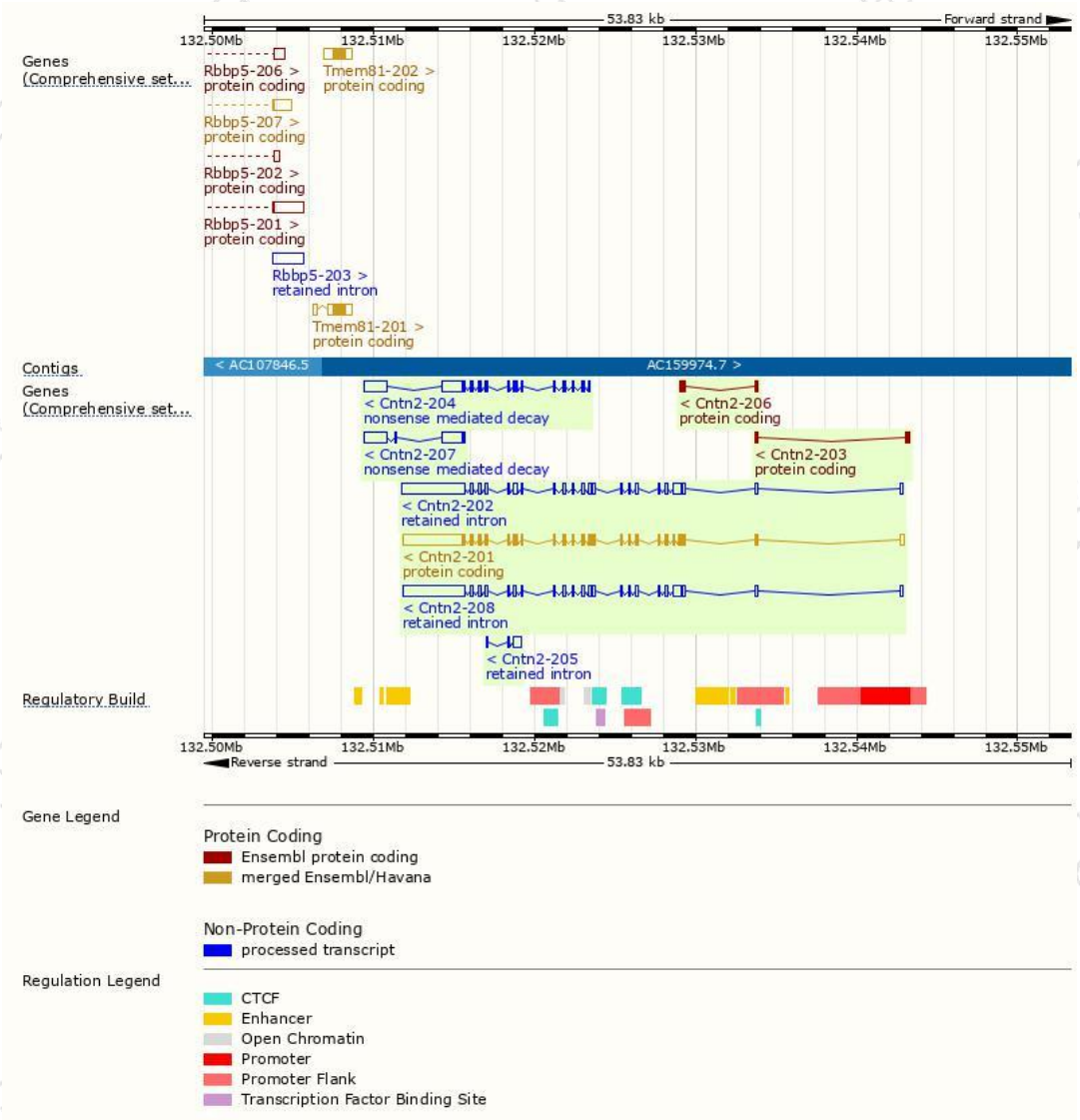
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cntn2-201	ENSMUST00000086521.10	7177	1040aa	Protein coding	CCDS15288	Q61330	TSL:1 GENCODE basic APPRIS P1
Cntn2-206	ENSMUST00000188943.1	460	102aa	Protein coding	-	A0A087WPI9	CDS 3' incomplete TSL:2
Cntn2-203	ENSMUST00000186530.2	308	25aa	Protein coding	-	A0A087WPS6	CDS 3' incomplete TSL:3
Cntn2-204	ENSMUST00000188065.6	4287	544aa	Nonsense mediated decay	-	A0A087WQQ9	CDS 5' incomplete TSL:1
Cntn2-207	ENSMUST00000189528.6	2836	34aa	Nonsense mediated decay	-	A0A087WST5	CDS 5' incomplete TSL:1
Cntn2-202	ENSMUST00000186487.6	7334	No protein	Retained intron	-	-	TSL:1
Cntn2-208	ENSMUST00000190601.6	7236	No protein	Retained intron	-	-	TSL:1
Cntn2-205	ENSMUST00000188143.1	696	No protein	Retained intron	-	-	TSL:3

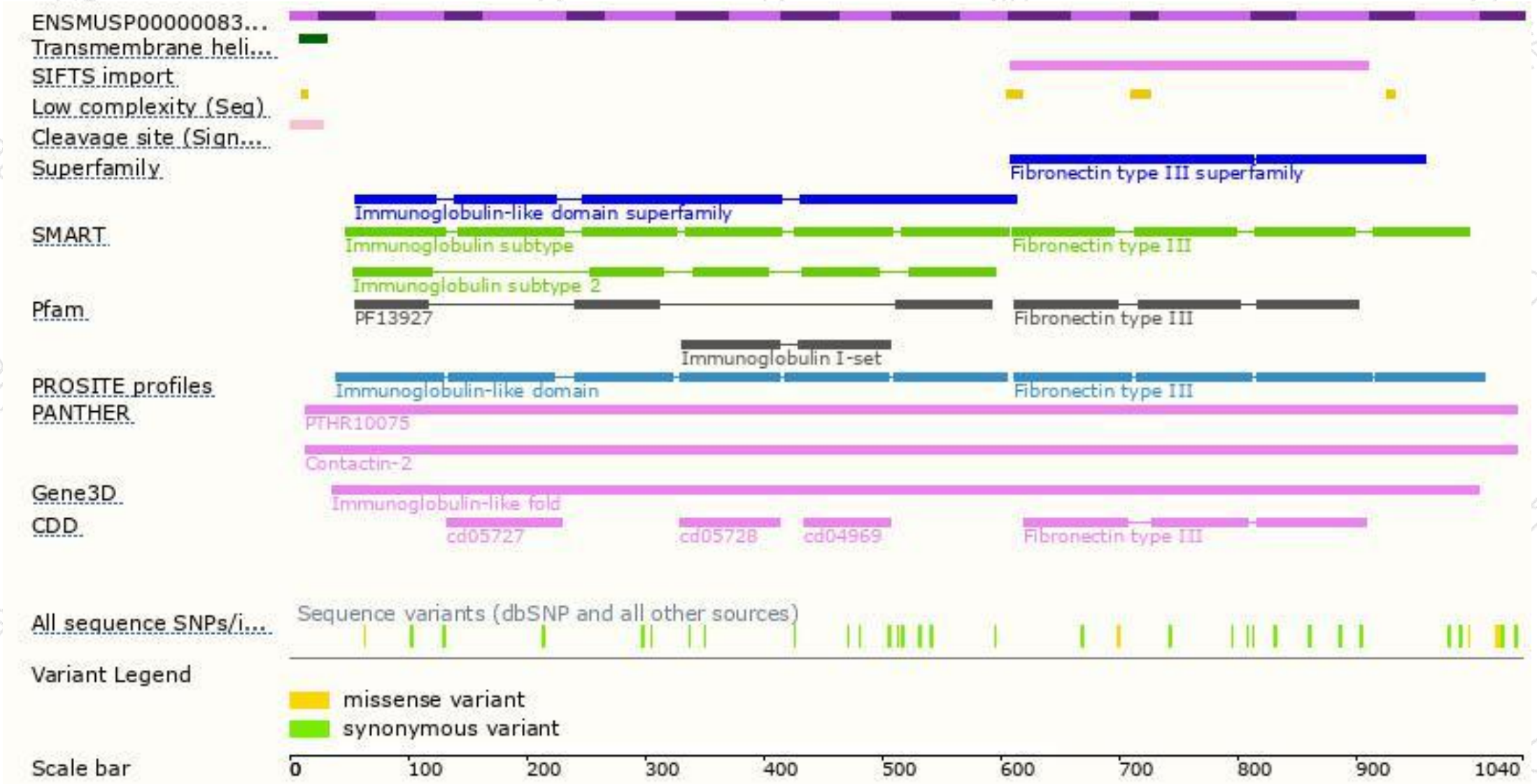
The strategy is based on the design of *Cntn2-201* transcript,The transcription is shown below



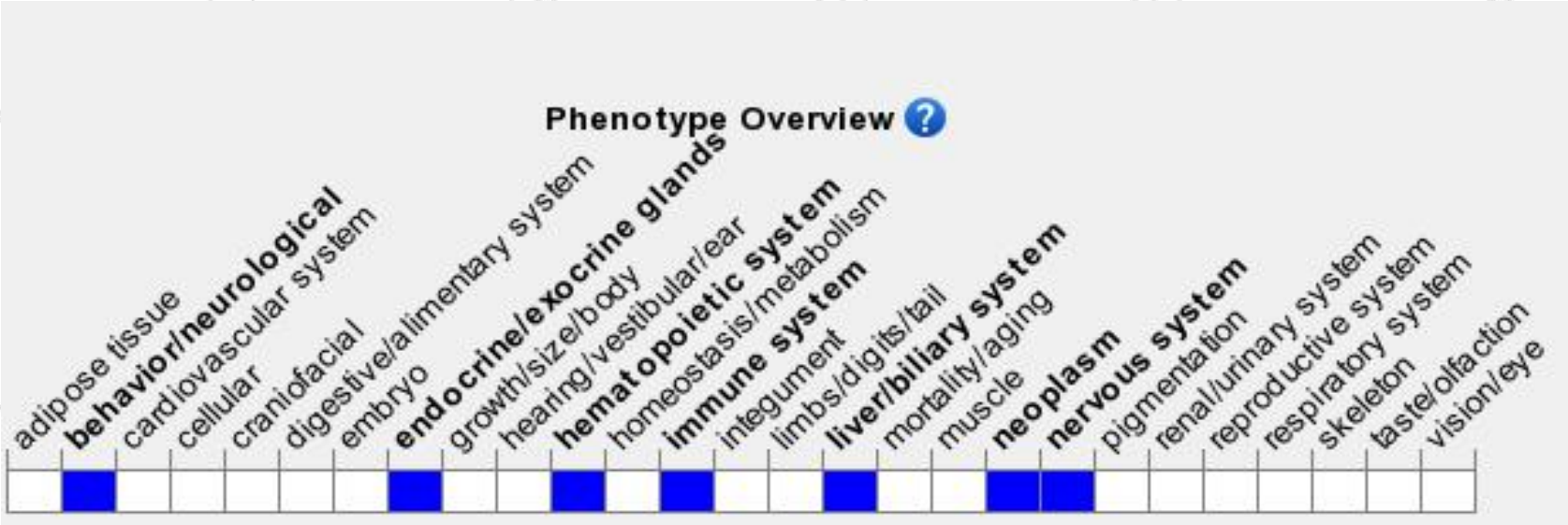
Genomic location distribution



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,Targeted mutation of this locus results in molecular abnormalities in the central nervous system.

集萃药康
GemPharmatech

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

Tel: 400-9660890

集萃药康生物科技
GemPharmatech Co.,Ltd