Scn5a Cas9-KO Strategy

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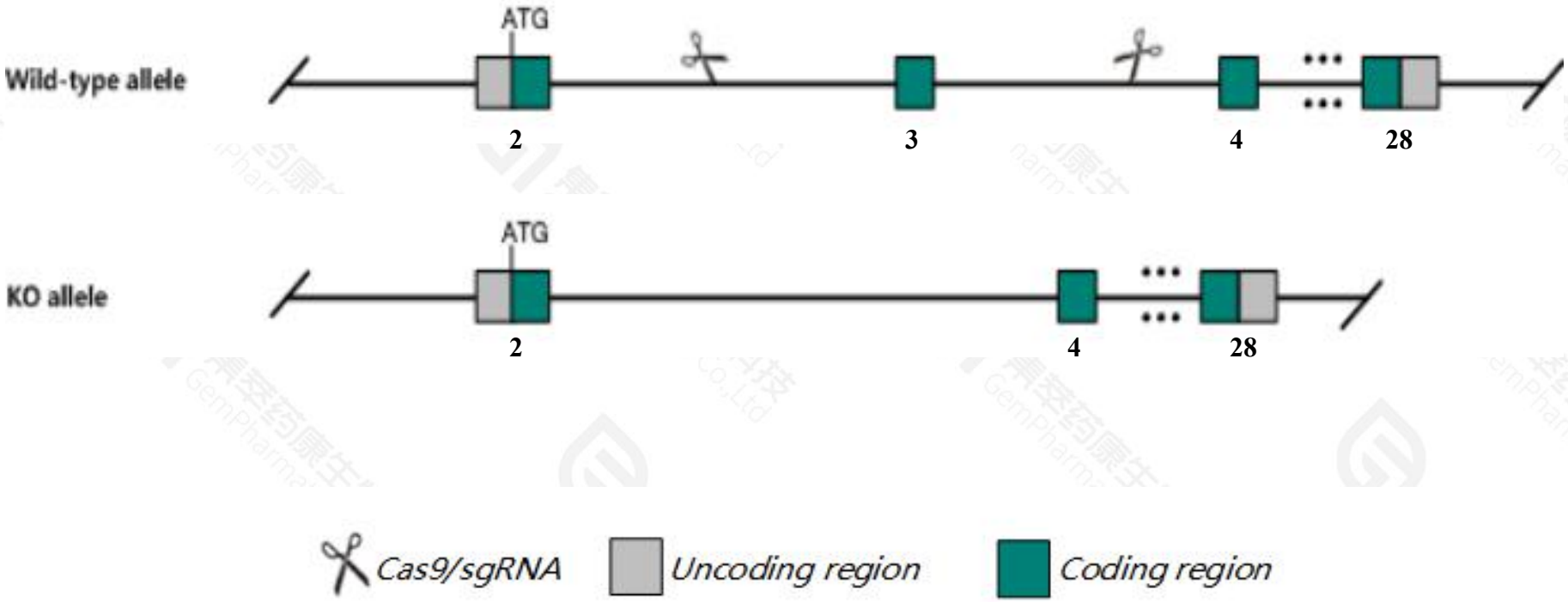
Design Date: 2021-2-24

Project Overview

Project Name	***Scn5a***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Scn5a* gene. The schematic diagram is as follows:



Technical routes

➢ The *Scn5a* gene has 4 transcripts. According to the structure of *Scn5a* gene, exon3 of *Scn5a-202*(ENSMUST00000117911.7) transcript is recommended as the knockout region.The region contains 119bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Scn5a* 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,mice homozygous for mutations in this gene die prenatally usually during organogenesis and may display decreased embryo size and abnormal cardiovascular system physiology. Heterozygous mice typically display abnormal heartbeats and defects in the function of the impulse conduction system.

➢ The *Scn5a* 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 gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

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Scn5a sodium channel, voltage-gated, type V, alpha [Mus musculus (house mouse)]

Gene ID: 20271, updated on 13-Mar-2020

Summary

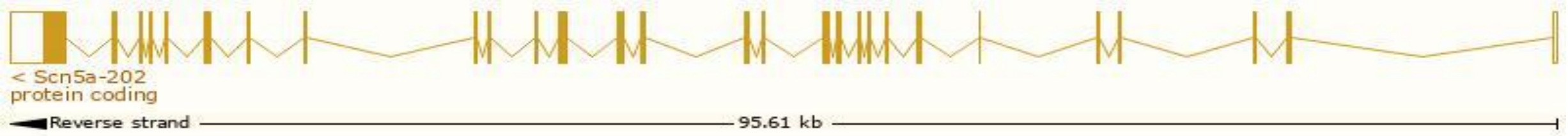
Official Symbol	Scn5a provided by MGI
Official Full Name	sodium channel, voltage-gated, type V, alpha provided by MGI
Primary source	MGI:MGI:98251
See related	Ensembl:ENSMUSG00000032511
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Nav1.5, Nav1.5c, SkM1, SkM2, mH1
Expression	Biased expression in heart adult (RPKM 37.1) and lung adult (RPKM 3.7)See more
Orthologs	human all

Transcript information（Ensembl）

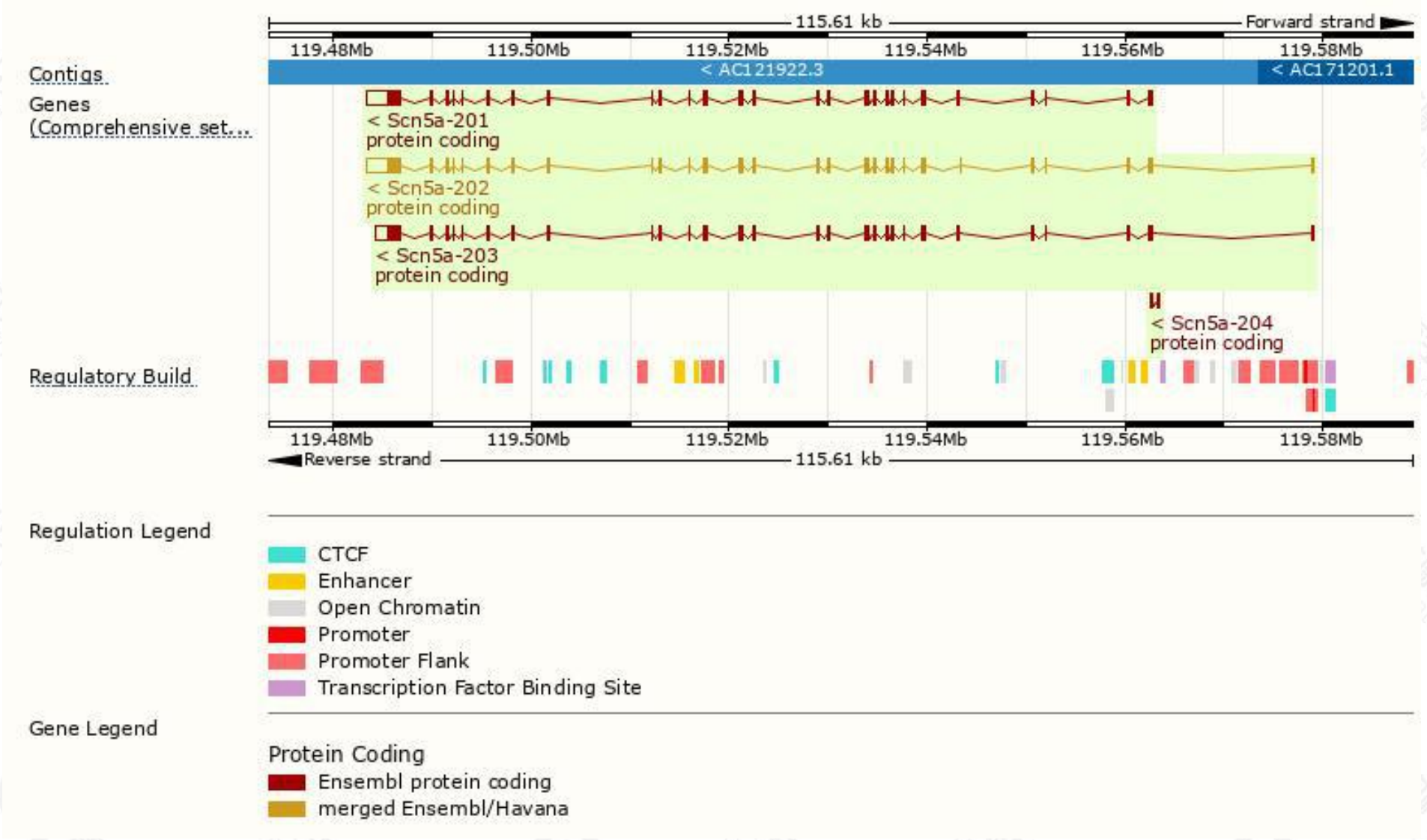
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Scn5a-202	ENSMUST00000117911.7	8453	2020aa	Protein coding	CCDS52965	A0A0R4J1M7	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Scn5a-201	ENSMUST00000065196.12	8287	2020aa	Protein coding	CCDS57715	K3W4N7	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT1
Scn5a-203	ENSMUST00000120420.1	7536	2019aa	Protein coding	-	Q9JJV9	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT1
Scn5a-204	ENSMUST00000138934.1	359	29aa	Protein coding	-	D3YXW2	CDS 3' incomplete TSL:3

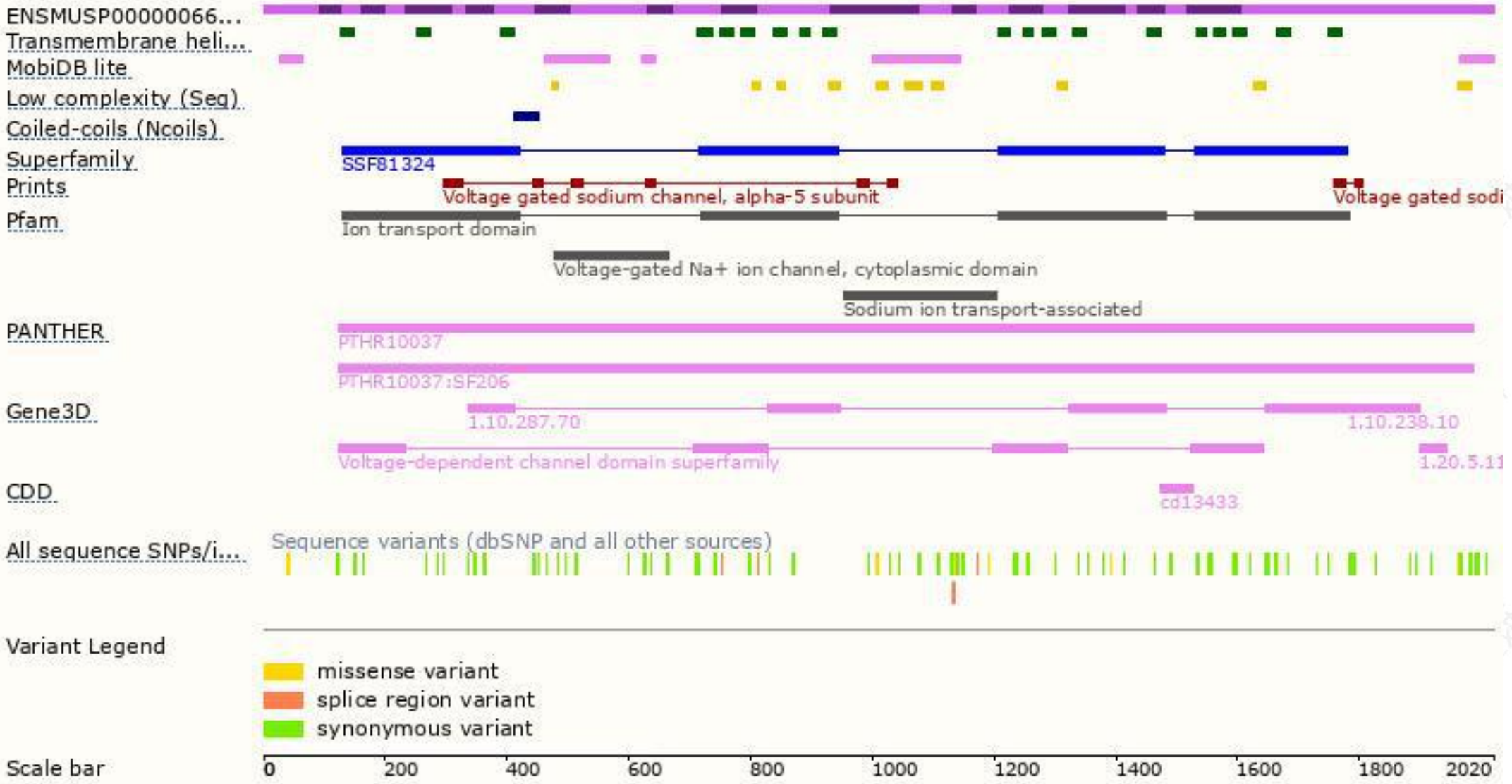
The strategy is based on the design of *Scn5a-202* transcript,the transcription is shown below:



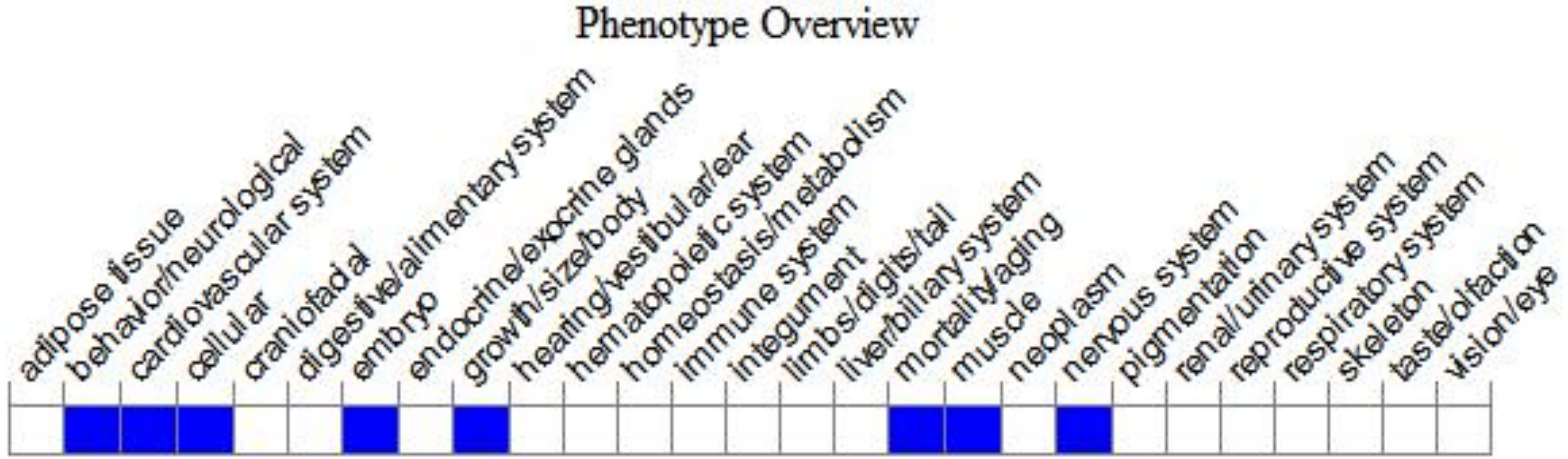
Genomic location distribution



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/).

According to the existing MGI data,mice homozygous for mutations in this gene die prenatally usually during organogenesis and may display decreased embryo size and abnormal cardiovascular system physiology. Heterozygous mice typically display abnormal heartbeats and defects in the function of the impulse conduction system.

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

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