Kcnh2 Cas9-CKO Strategy

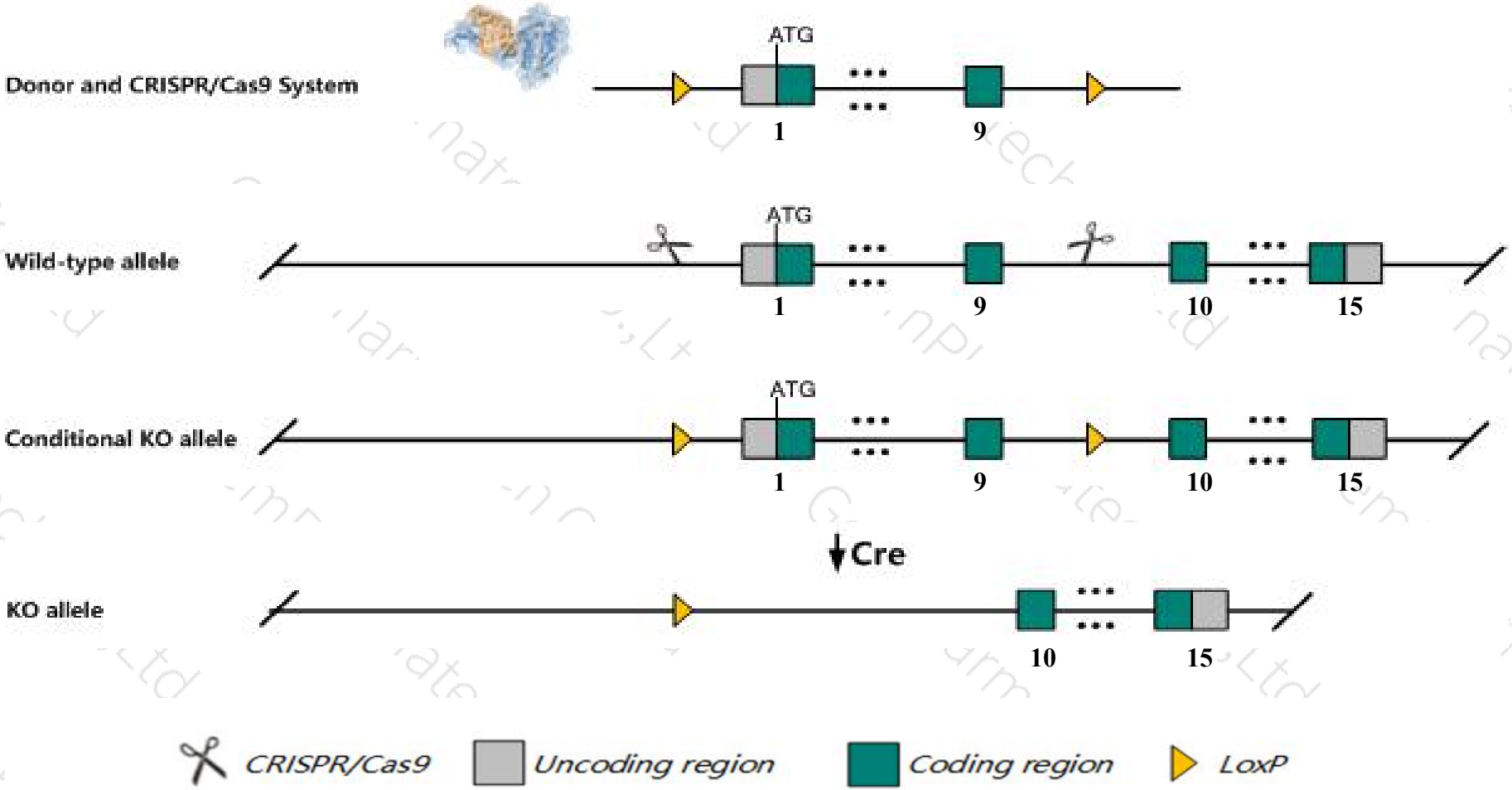
Designer: Huan Wang
Reviewer: Huan Fan
Design Date: 2020-2-18

Project Overview

Project Name	***Kcnh2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Kcnh2* gene. The schematic diagram is as follows:



Technical routes

- The *Kcnh2* gene has 5 transcripts. According to the structure of *Kcnh2* gene, exon1-exon9 of *Kcnh2-201* (ENSMUST00000036092.9) transcript is recommended as the knockout region.The region contains start codon ATG. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Kcnh2* 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,Mutant mice which maintain expression of the A isoform and lack expression of the B isoform are predisposed to episodic sinus bradycardia. Mice with mutations causing defects in both isoforms are embryonic lethal with defects in cardiac development and function.

- The KO region contains functional region of the *Gm15589* gene. Knockout the region will affect the function of *Gm15589* gene

- The *Kcnh2* gene is located on the Chr5. 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

Kcnh2 potassium voltage-gated channel, subfamily H (eag-related), member 2 [Mus musculus (house mouse)]

Gene ID: 16511, updated on 31-Jan-2019

Summary

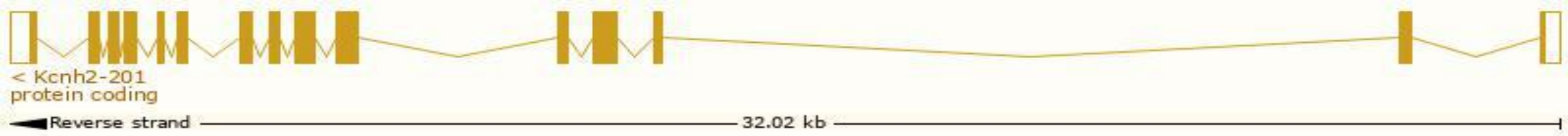
Official Symbol	Kcnh2 provided by MGI
Official Full Name	potassium voltage-gated channel, subfamily H (eag-related), member 2 provided by MGI
Primary source	MGI:MGI:1341722
See related	Ensembl:ENSMUSG00000038319
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI326795, ERG1, LQT, Lqt2, M-erg, Merg1, merg1a, merg1b
Expression	Broad expression in ovary adult (RPKM 33.4), thymus adult (RPKM 29.0) and 22 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

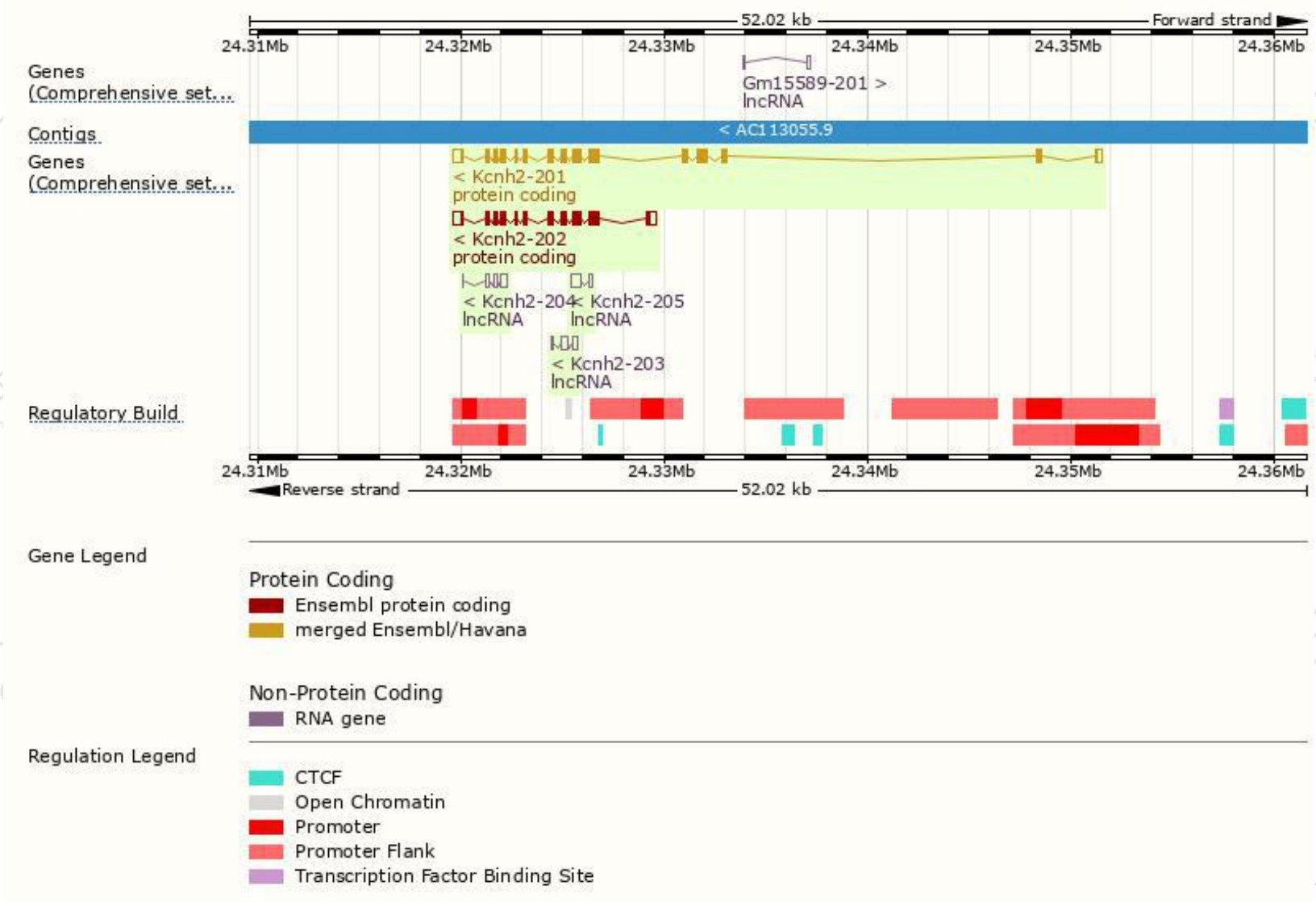
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kcnh2-201	ENSMUST00000036092.9	4221	1162aa	Protein coding	CCDS19116	Q53Z09	TSL:1 GENCODE basic APPRIS P1
Kcnh2-202	ENSMUST00000115098.6	3193	820aa	Protein coding	CCDS80224	A0A0R4J1K0	TSL:1 GENCODE basic
Kcnh2-204	ENSMUST00000129246.1	741	No protein	lncRNA	-	-	TSL:2
Kcnh2-203	ENSMUST00000126791.1	710	No protein	lncRNA	-	-	TSL:3
Kcnh2-205	ENSMUST00000142197.1	605	No protein	lncRNA	-	-	TSL:2

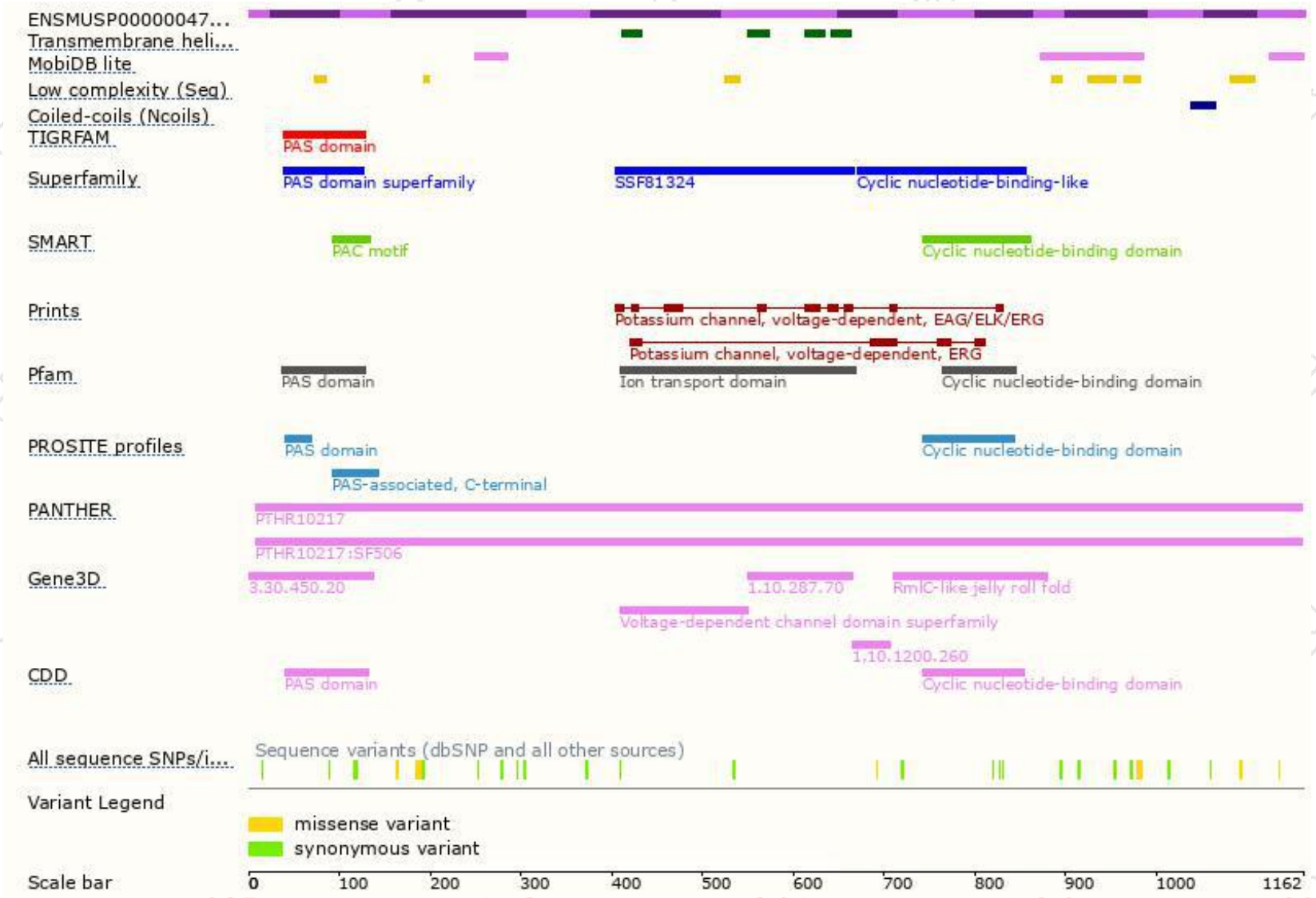
The strategy is based on the design of *Kcnh2-201* transcript,The transcription is shown below



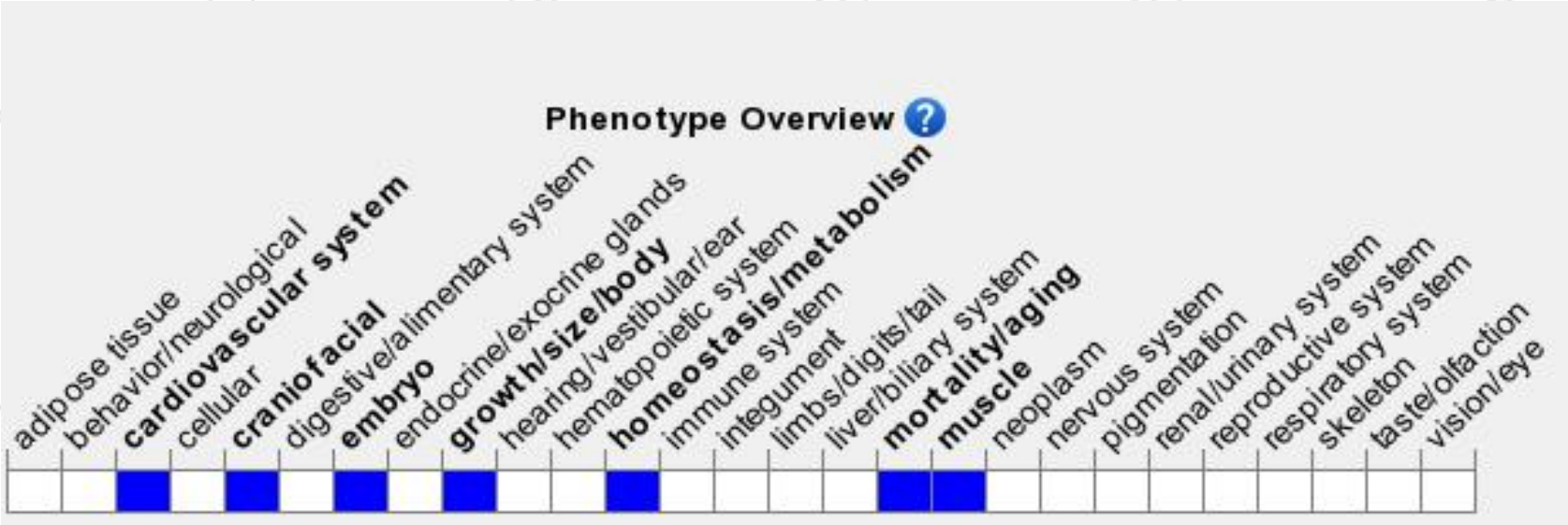
Genomic location distribution



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,Mutant mice which maintain expression of the A isoform and lack expression of the B isoform are predisposed to episodic sinus bradycardia. Mice with mutations causing defects in both isoforms are embryonic lethal with defects in cardiac development and function.

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