Trpm2 Cas9-CKO Strategy

Designer: Ruirui Zhang
Reviewer: Huimin Su

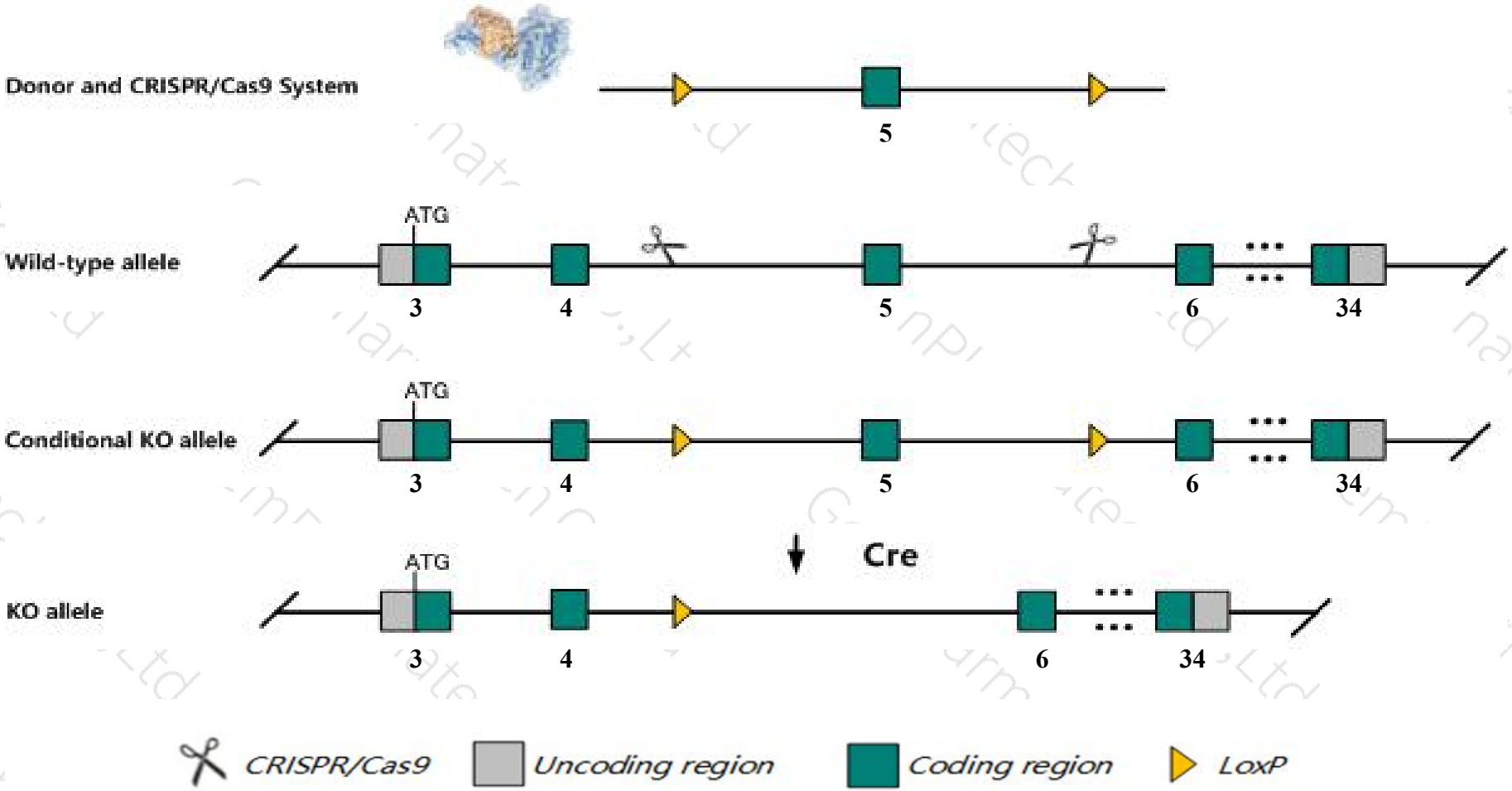
Design Date: 2019-9-21

Project Overview

Project Name	***Trpm2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Trpm2* gene. The schematic diagram is as follows:



Technical routes

- The *Trpm2* gene has 10 transcripts. According to the structure of *Trpm2* gene, exon5 of *Trpm2-203* (ENSMUST00000105401.8) transcript is recommended as the knockout region.The region contains 169bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Trpm2* 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 knock-out allele display impaired reactive oxygen species (ROS)-induced chemokine production in monocytes, and reduced neutrophil infiltration and ulceration in a dextran sulfate sodium-induced colitis inflammation model.

➢ The *Trpm2* gene is located on the Chr10. 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

Trpm2 transient receptor potential cation channel, subfamily M, member 2 [*Mus musculus* (house mouse)]

Gene ID: 28240, updated on 21-Aug-2019

Summary

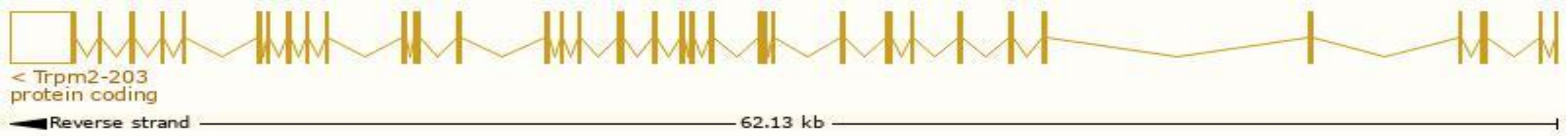
Official Symbol	Trpm2 provided by MGI
Official Full Name	transient receptor potential cation channel, subfamily M, member 2 provided by MGI
Primary source	MGI:MGI:1351901
See related	Ensembl:ENSMUSG00000009292
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Trp7; TRPC7; Trrp7; C79133; LTRPC2; 9830168K16Rik
Expression	Biased expression in spleen adult (RPKM 10.5), cerebellum adult (RPKM 4.0) and 12 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

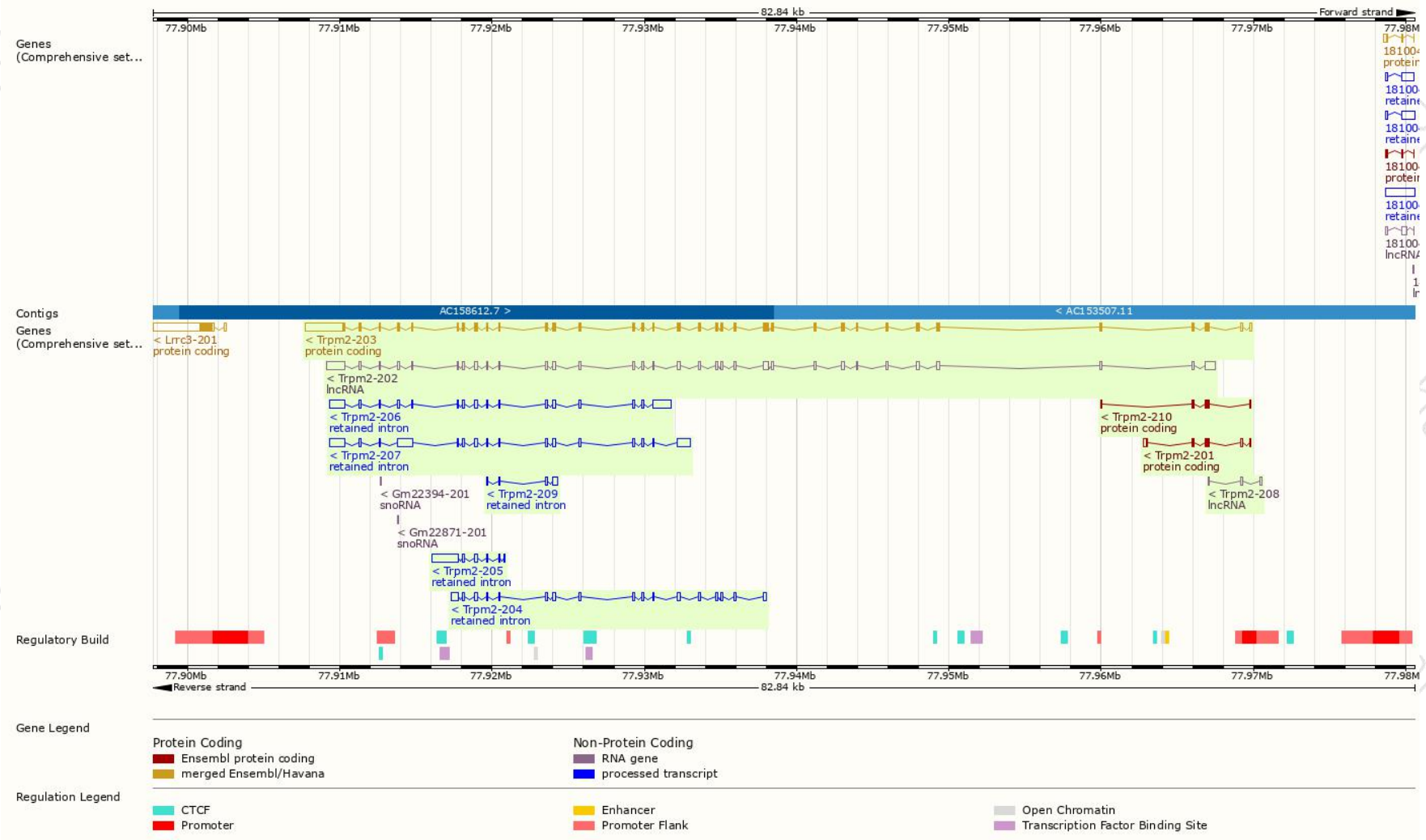
The gene has 10 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Trpm2-203	ENSMUST00000105401.8	7274	1506aa	Protein coding	CCDS48611	Q91YD4	TSL:1 GENCODE basic APPRIS P1
Trpm2-201	ENSMUST00000105399.1	731	109aa	Protein coding	-	D3Z7Q7	TSL:1 GENCODE basic
Trpm2-210	ENSMUST00000219997.1	436	104aa	Protein coding	-	A0A1W2P6F5	CDS 3' incomplete TSL:2
Trpm2-207	ENSMUST00000153842.8	4508	No protein	Retained intron	-	-	TSL:2
Trpm2-206	ENSMUST00000140471.7	3990	No protein	Retained intron	-	-	TSL:1
Trpm2-204	ENSMUST00000126206.2	2838	No protein	Retained intron	-	-	TSL:1
Trpm2-205	ENSMUST00000138238.7	2196	No protein	Retained intron	-	-	TSL:1
Trpm2-209	ENSMUST00000217806.1	627	No protein	Retained intron	-	-	TSL:3
Trpm2-202	ENSMUST00000105400.8	5980	No protein	lncRNA	-	-	TSL:1
Trpm2-208	ENSMUST00000154996.1	352	No protein	lncRNA	-	-	TSL:3

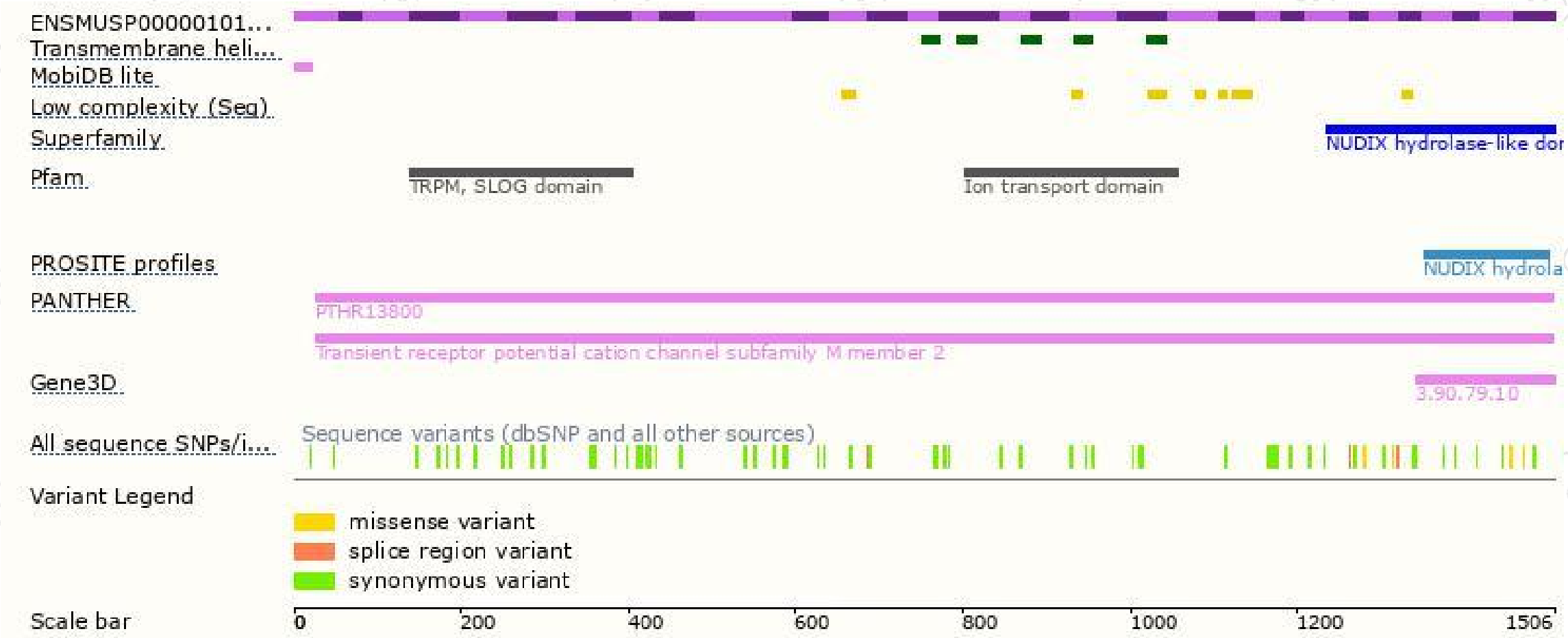
The strategy is based on the design of *Trpm2-203* transcript,The transcription is shown below



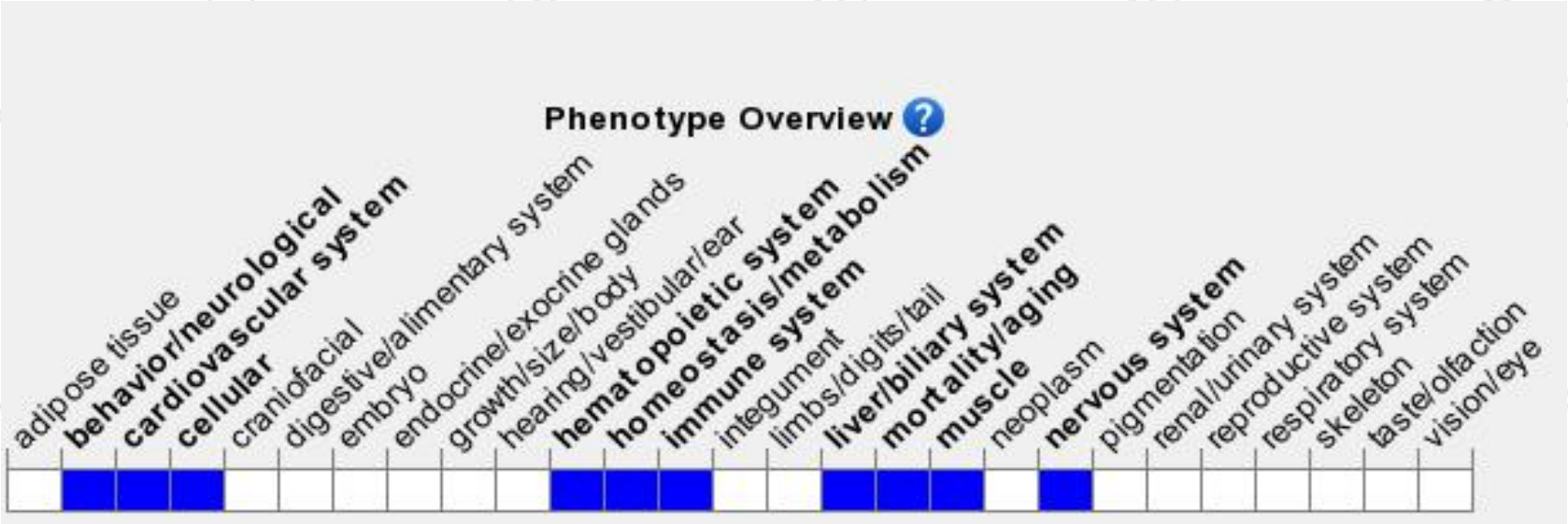
Genomic location distribution



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 a knock-out allele display impaired reactive oxygen species (ROS)-induced chemokine production in monocytes, and reduced neutrophil infiltration and ulceration in a dextran sulfate sodium-induced colitis inflammation model.

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