Adar Cas9-CKO Strategy

Designer: Xiaojing Li

Reviewer: JiaYu

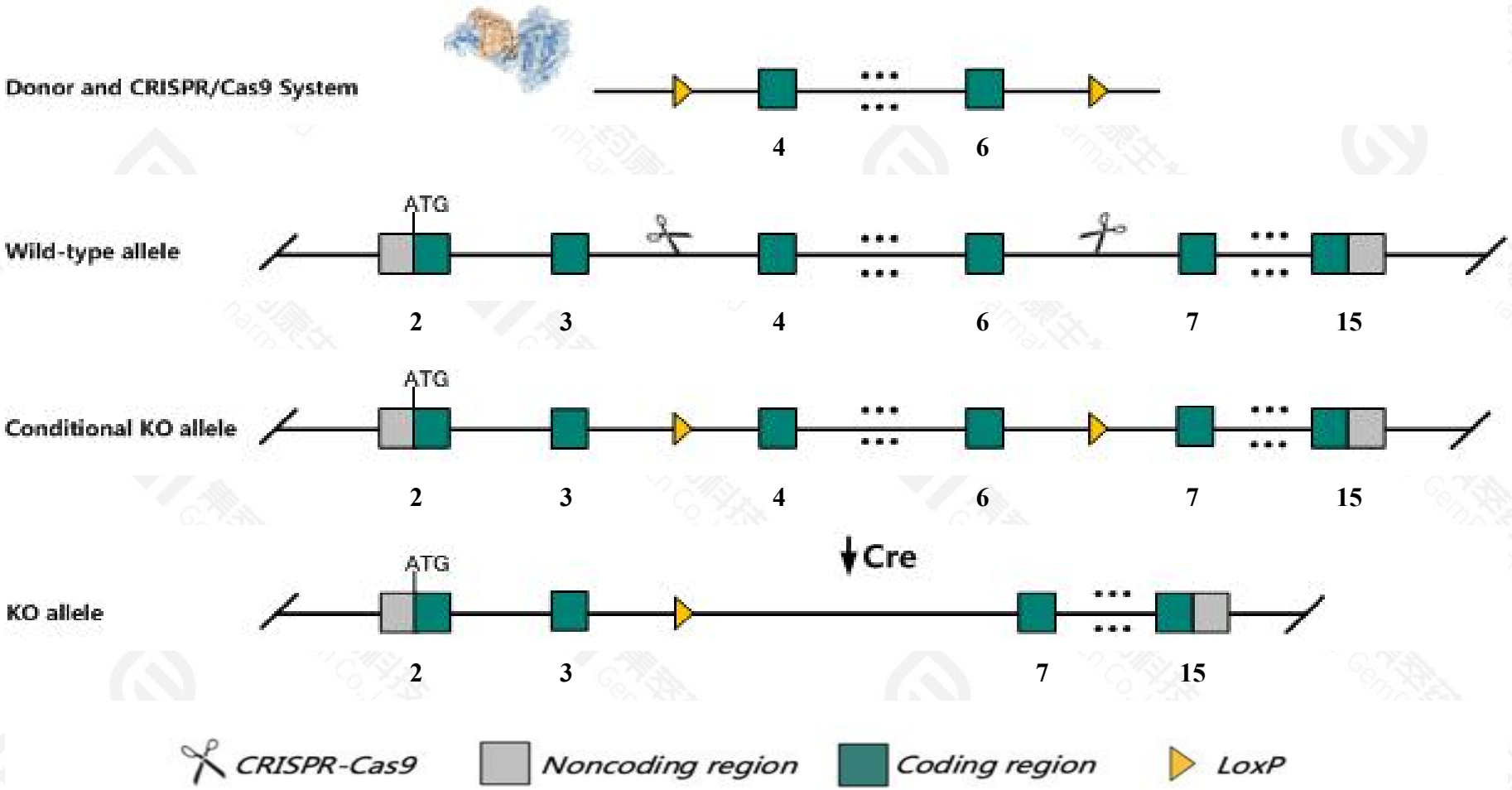
Design Date: 2023-1-19

Project Overview

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

Conditional Knockout strategy

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



Technical routes

➢ The *Adar* gene has 9 transcripts. According to the structure of *Adar* gene, exon4-exon6 of *Adar-202*(ENSMUST00000098924.9) transcript is recommended as the knockout region.The region contains 473bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR-Cas9 technology to modify *Adar* 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,homozygous null mice die during gestation. Inactivation of this locus has been associated with increased apoptosis and, in some lines, defects in both primitive and definitive hematopoiesis.

➢ The *Adar* gene is located on the Chr3. 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

Adar adenosine deaminase, RNA-specific [Mus musculus (house mouse)]

Gene ID: 56417, updated on 28-Mar-2019

Summary

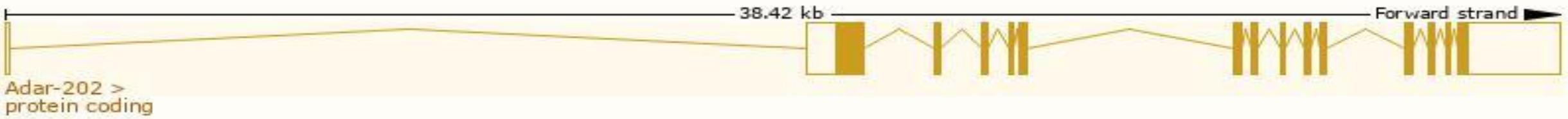
Official Symbol	Adar provided by MGI
Official Full Name	adenosine deaminase, RNA-specific provided by MGI
Primary source	MGI:MGI:1889575
See related	Ensembl:ENSMUSG00000027951
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AV242451, Adar1, Adar1p110, Adar1p150, DRADA, mZaADAR
Expression	Ubiquitous expression in cortex adult (RPKM 21.4), cerebellum adult (RPKM 21.1) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

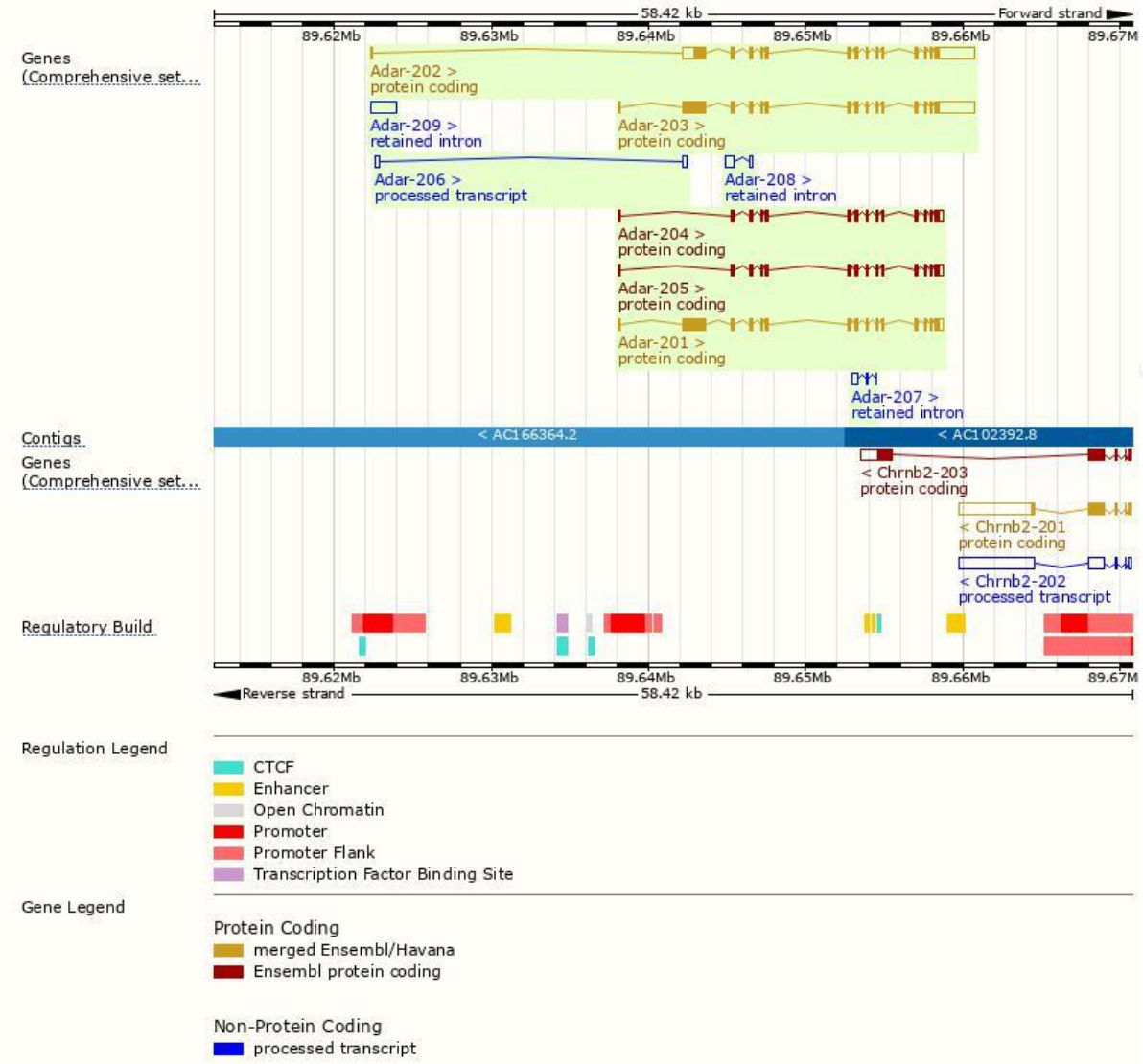
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Adar-202	ENSMUST00000098924.8	5909	930aa	Protein coding	CCDS17514	Q99MU3	TSL:1 GENCODE basic APPRIS ALT2
Adar-203	ENSMUST00000107405.5	5871	1178aa	Protein coding	CCDS50963	Q99MU3	TSL:1 GENCODE basic APPRIS ALT2
Adar-201	ENSMUST00000029563.13	3753	1152aa	Protein coding	CCDS17513	Q99MU3	TSL:1 GENCODE basic APPRIS P4
Adar-204	ENSMUST00000118341.5	2399	660aa	Protein coding	-	Q99MU3	TSL:1 GENCODE basic
Adar-205	ENSMUST00000121094.7	2321	634aa	Protein coding	-	Q99MU3	TSL:1 GENCODE basic
Adar-206	ENSMUST00000123691.1	519	No protein	Processed transcript	-	-	TSL:2
Adar-209	ENSMUST00000197253.1	1668	No protein	Retained intron	-	-	TSL:NA
Adar-208	ENSMUST00000150637.1	708	No protein	Retained intron	-	-	TSL:2
Adar-207	ENSMUST00000131030.1	481	No protein	Retained intron	-	-	TSL:2

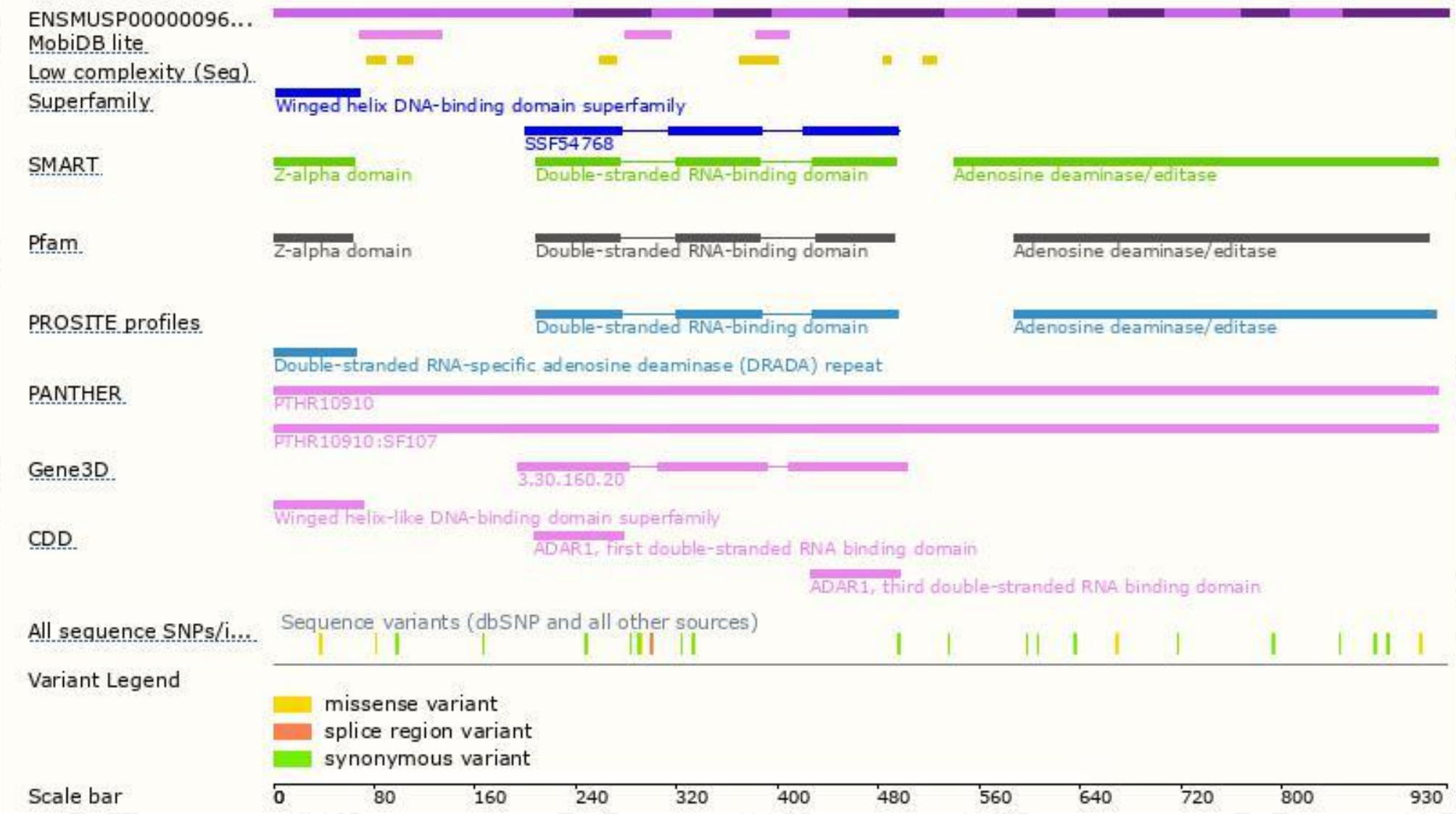
The strategy is based on the design of *Adar-202* transcript,the transcription is shown below:



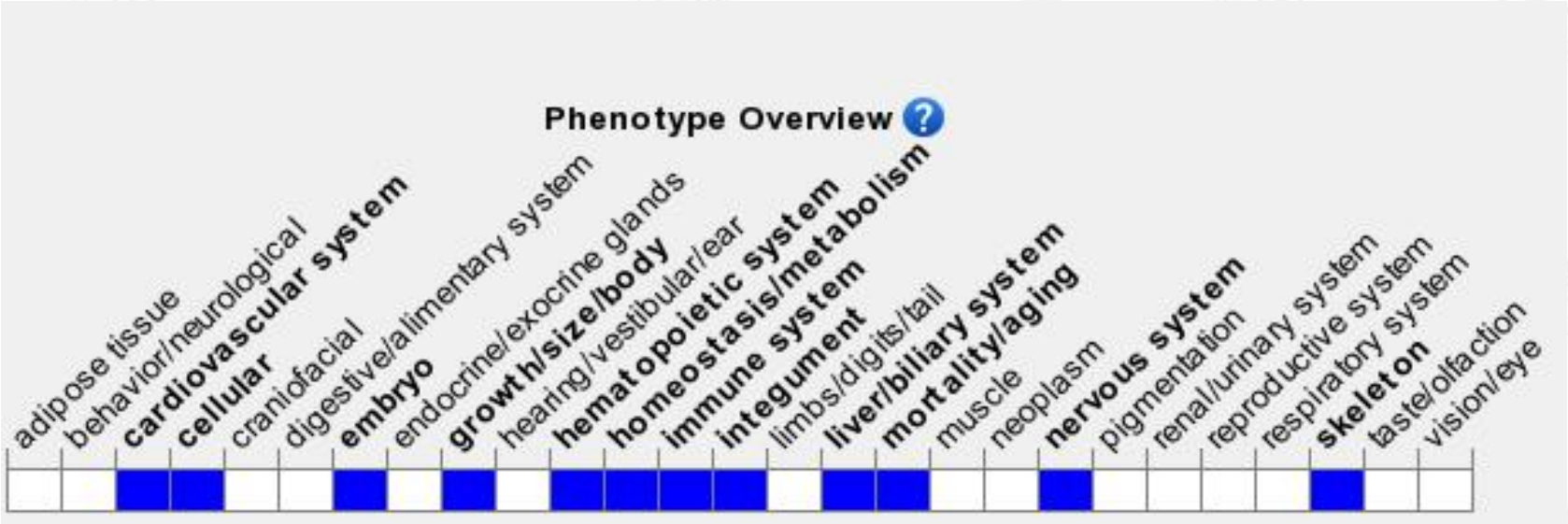
Genomic location distribution



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,homozygous null mice die during gestation. Inactivation of this locus has been associated with increased apoptosis and, in some lines, defects in both primitive and definitive hematopoiesis.

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