

# ***Wasf2 Cas9-CKO Strategy***

**Designer:**

**Daohua Xu**



# Project Overview

**Project Name**

***Wasf2***

**Project type**

**Cas9-CKO**

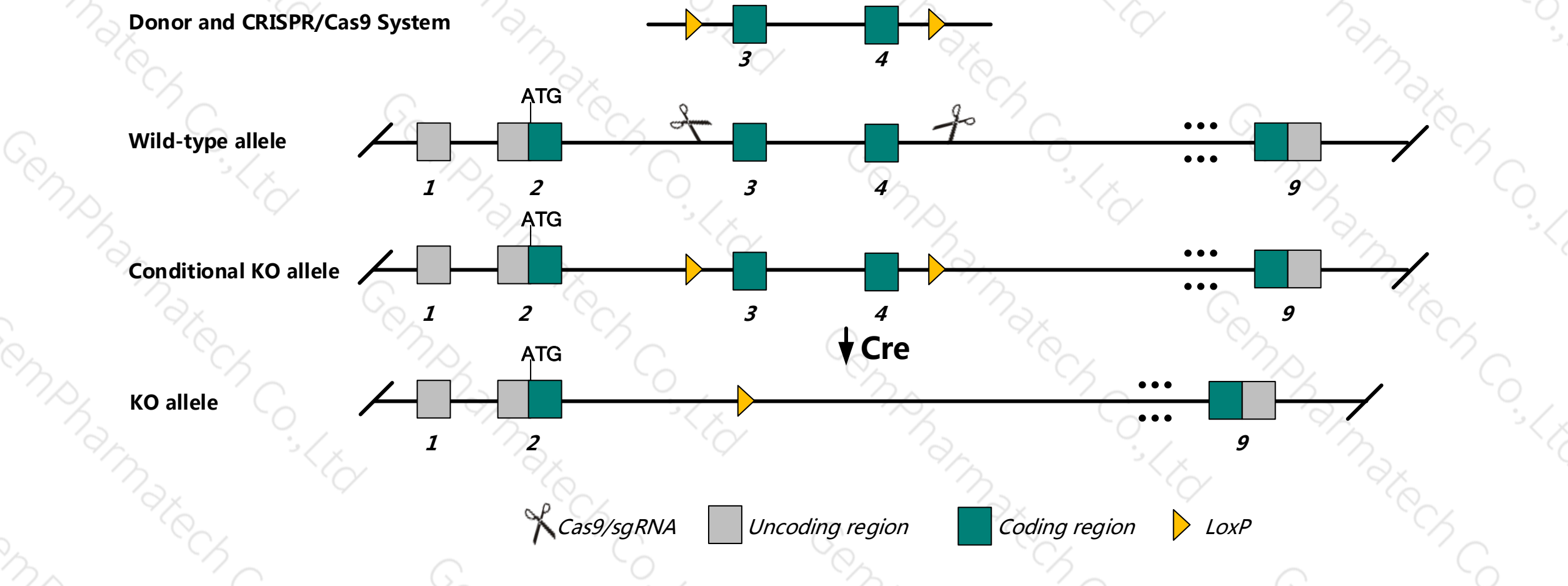
**Strain background**

**C57BL/6JGpt**



# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Wasf2* gene. The schematic diagram is as follows:





# Technical routes

- The *Wasf2* gene has 6 transcripts. According to the structure of *Wasf2* gene, exon3-exon4 of *Wasf2*-201 (ENSMUST00000084241.11) transcript is recommended as the knockout region. The region contains 289bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Wasf2* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.



- According to the existing MGI data , Homozygous mutants show impaired embryonic development and do not survive to term. In addition to reduced embryo size, observed defects include hemorrhaging, abnormal somite development, perturbed angiogenesis, and shrunken cerebral ventricles.
- The *Wasf2* gene is located on the Chr4. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.



# Gene information ( NCBI )



## Wsf2 WAS protein family, member 2 [ *Mus musculus* (house mouse) ]

Gene ID: 242687, updated on 9-Sep-2018

Summary

Official Symbol	Wsf2 provided by <a href="#">MGI</a>
Official Full Name	WAS protein family, member 2 provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:1098641</a>
See related	<a href="#">Ensembl:ENSMUSG00000028868</a> <a href="#">Vega:OTTMUSG00000010477</a>
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	WAVE2; AW742646; D4Ert13e
Expression	Ubiquitous expression in thymus adult (RPKM 47.2), lung adult (RPKM 40.4) and 28 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

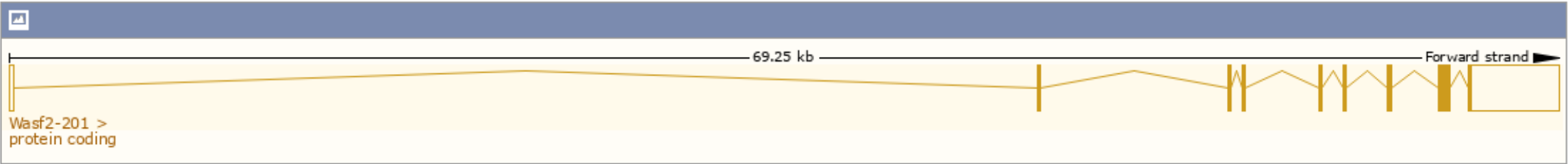


# Transcript information ( Ensembl )

The gene has 6 transcripts, and all transcripts are shown below:

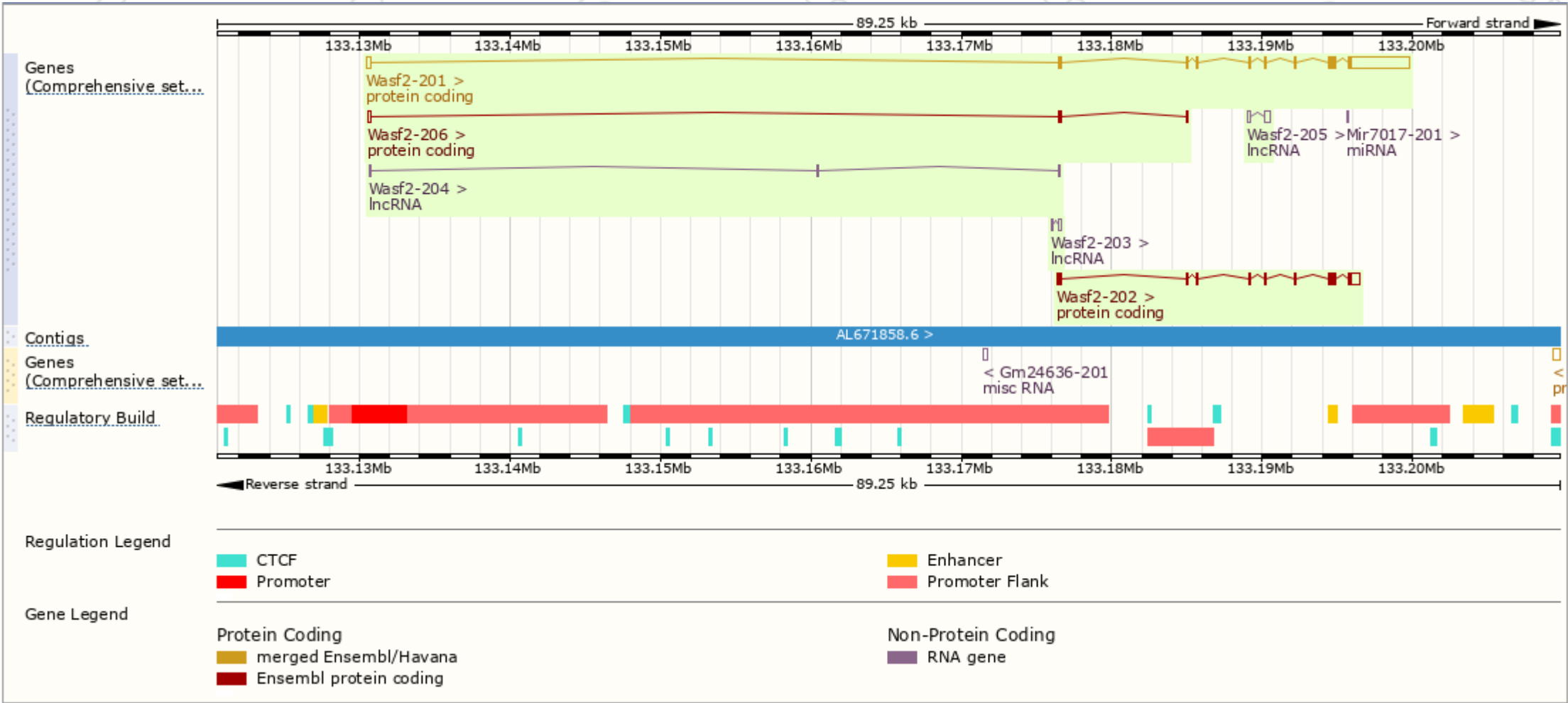
Show/hide columns (1 hidden)							Filter	
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
Wasf2-201	<a href="#">ENSMUST00000084241.11</a>	5642	<a href="#">497aa</a>	Protein coding	<a href="#">CCDS18742</a>	<a href="#">Q8BH43</a>	TSL:1	GENCODE basic APPRIS P1
Wasf2-202	<a href="#">ENSMUST00000105912.1</a>	2236	<a href="#">497aa</a>	Protein coding	<a href="#">CCDS18742</a>	<a href="#">Q8BH43</a>	TSL:1	GENCODE basic APPRIS P1
Wasf2-206	<a href="#">ENSMUST00000138831.1</a>	483	<a href="#">85aa</a>	Protein coding	-	<a href="#">B1AUN0</a>	CDS 3' incomplete	TSL:2
Wasf2-205	<a href="#">ENSMUST00000136637.1</a>	599	No protein	lncRNA	-	-	TSL:3	
Wasf2-204	<a href="#">ENSMUST00000136132.1</a>	365	No protein	lncRNA	-	-	TSL:5	
Wasf2-203	<a href="#">ENSMUST00000123578.1</a>	223	No protein	lncRNA	-	-	TSL:5	

The strategy is based on the design of *Wasf2-201* transcript, The transcription is shown below



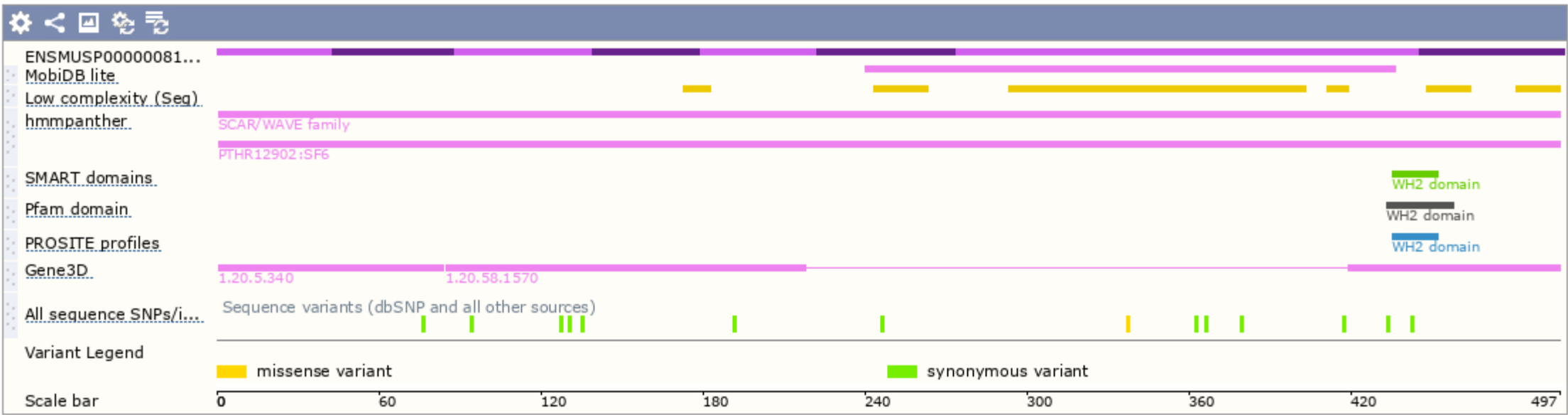


# Genomic location distribution



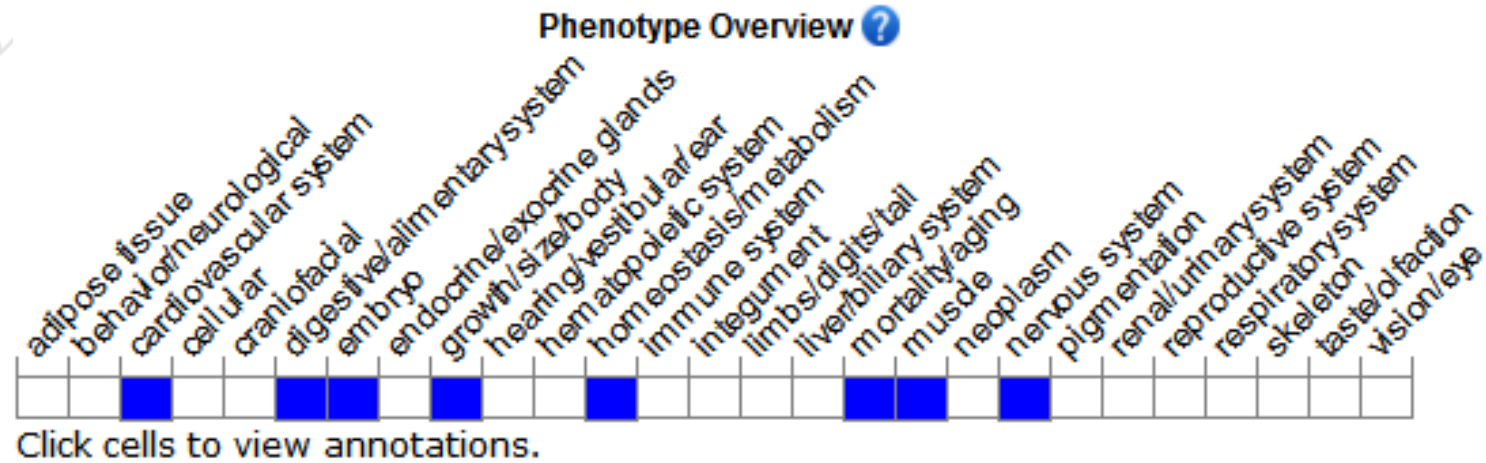


# Protein domain





# 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, Homozygous mutants show impaired embryonic development and do not survive to term. In addition to reduced embryo size, observed defects include hemorrhaging, abnormal somite development, perturbed angiogenesis, and shrunken cerebral ventricles.



If you have any questions, you are welcome to inquire.  
Tel: 025-5864 1534



集萃药康生物科技  
GemPharmatech Co.,Ltd

