# *Cldn1* Cas9-CKO Strategy

**Designer:** Huan Wang

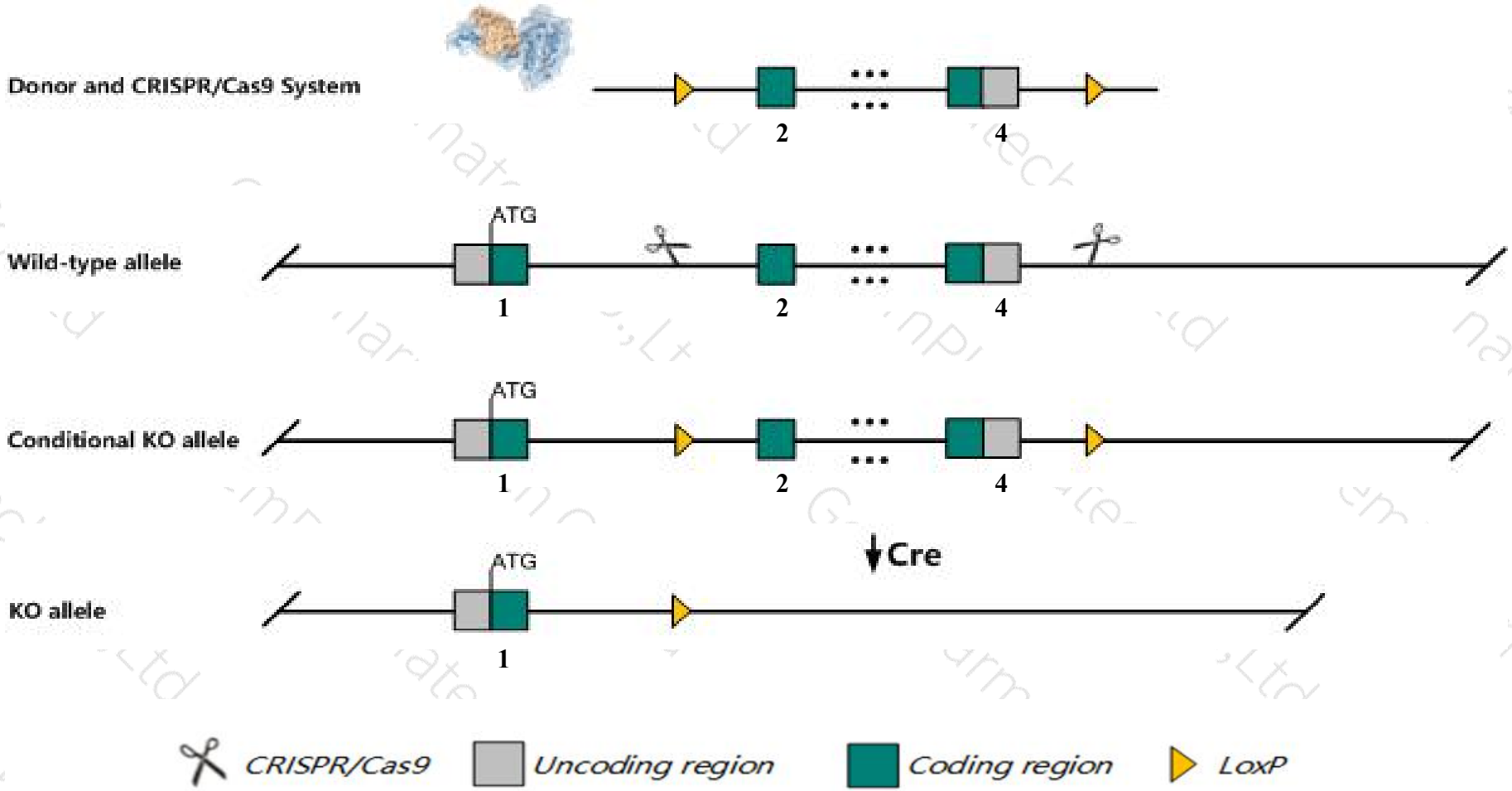
**Design Date:** 2019-7-24

# Project Overview

| | |
|---|---|
| **Project Name** | ***Cldn1*** |
| **Project type** | **Cas9-CKO** |
| **Strain background** | **C57BL/6JGpt** |

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cldn1* gene. The schematic diagram is as follows:



# Technical routes

- The *Cldn1* gene has 2 transcripts. According to the structure of *Cldn1* gene, exon2-exon4 of *Cldn1-201* (ENSMUST00000023154.2) transcript is recommended as the knockout region.The region contains most of coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Cldn1* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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,Animals homozygous for a mutation in this gene have wrinkled skin and die within 1 day after birth.

➢ The *Cldn1* gene is located on the Chr16. 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

## Cldn1 claudin 1 [Mus musculus (house mouse)]

Gene ID: 12737, updated on 5-Feb-2019

### Summary

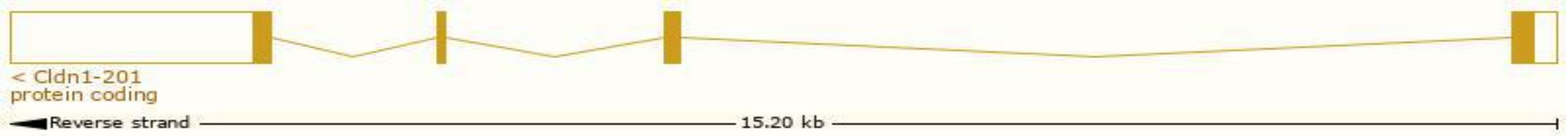
| | |
|---|---|
| **Official Symbol** | Cldn1 provided by MGI |
| **Official Full Name** | claudin 1 provided by MGI |
| **Primary source** | MGI:MGI:1276109 |
| **See related** | Ensembl:ENSMUSG00000022512 |
| **Gene type** | protein coding |
| **RefSeq status** | REVIEWED |
| **Organism** | Mus musculus |
| **Lineage** | Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus |
| **Also known as** | AI596271 |
| **Summary** | This gene encodes a member of the claudin family. Claudins are integral membrane proteins and components of tight junction strands. Tight junction strands serve as a physical barrier to prevent solutes and water from passing freely through the paracellular space between epithelial or endothelial cell sheets, and also play critical roles in maintaining cell polarity and signal transductions. The knockout mice lacking this gene die soon after birth as a consequence of dehydration from trandermal water loss, indicating that this gene is indispensable for creating and maintaining the epidermal barrier. The protein encoded by this gene also has gastric tumor suppressive activity, and is a key factor for hepatitis C virus (HCV) entry. [provided by RefSeq, Aug 2010] |
| **Expression** | Biased expression in liver E18 (RPKM 82.6), liver adult (RPKM 15.9) and 6 other tissuesSee more |
| **Orthologs** | human all |

# Transcript information（Ensembl）

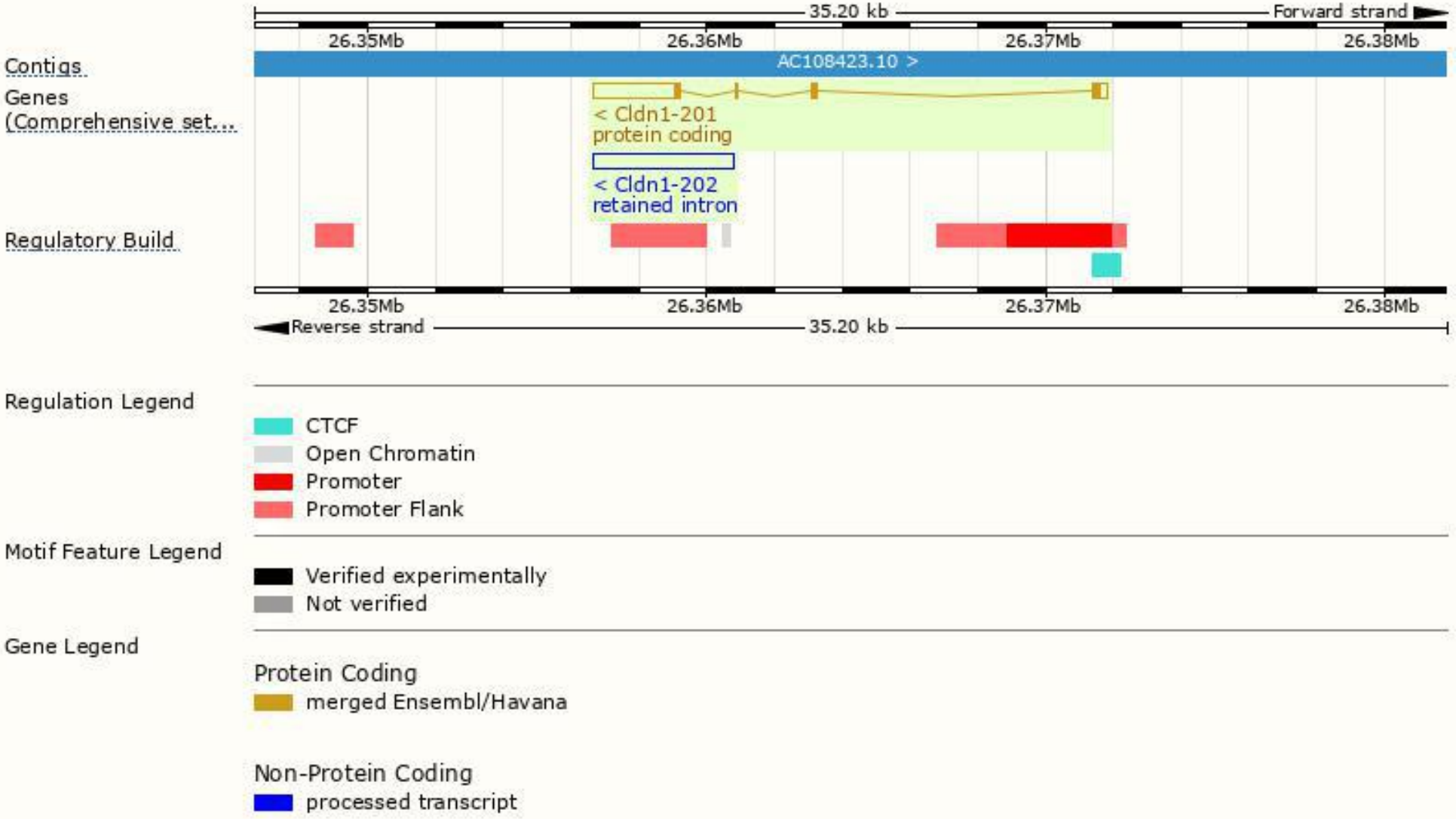
The gene has 2 transcripts,all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|
| Cldn1-201 | ENSMUST00000023154.2 | 3247 | 211aa | Protein coding | CCDS28087 | O88551 Q4FJV3 | TSL:1 GENCODE basic APPRIS P1 |
| Cldn1-202 | ENSMUST00000232215.1 | 4145 | No protein | Retained intron | - | - | |

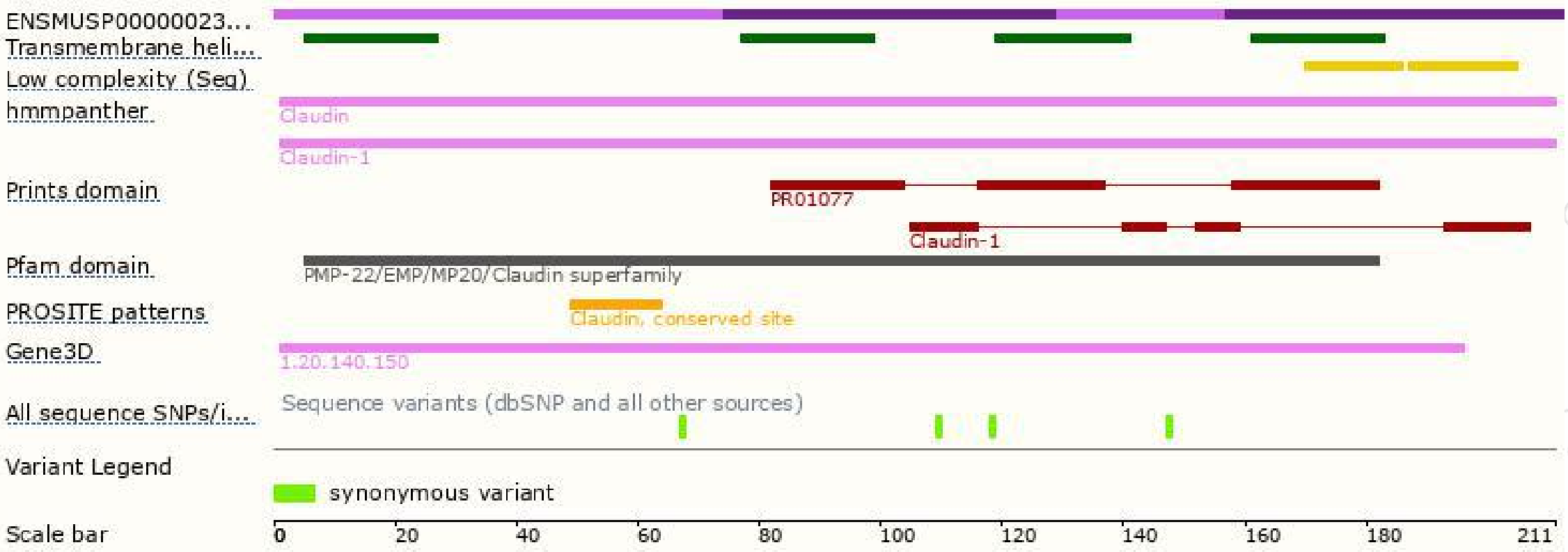
The strategy is based on the design of *Cldn1-201* transcript,The transcription is shown below



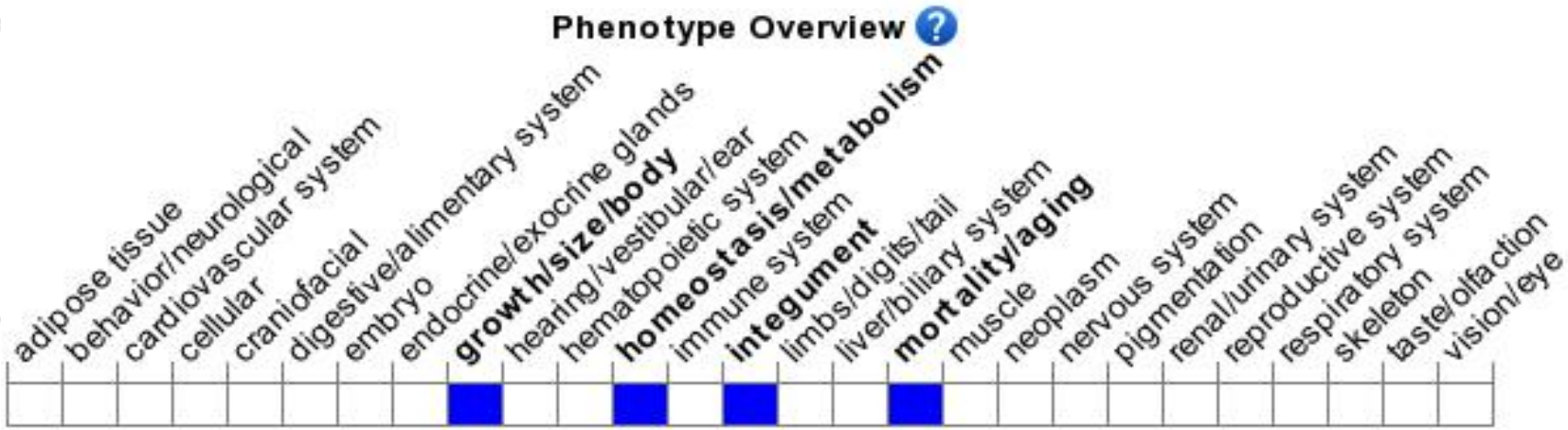
# Genomic location distribution



# 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,Animals homozygous for a mutation in this gene have wrinkled skin and die within 1 day after birth.

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