Lmna Cas9-CKO Strategy

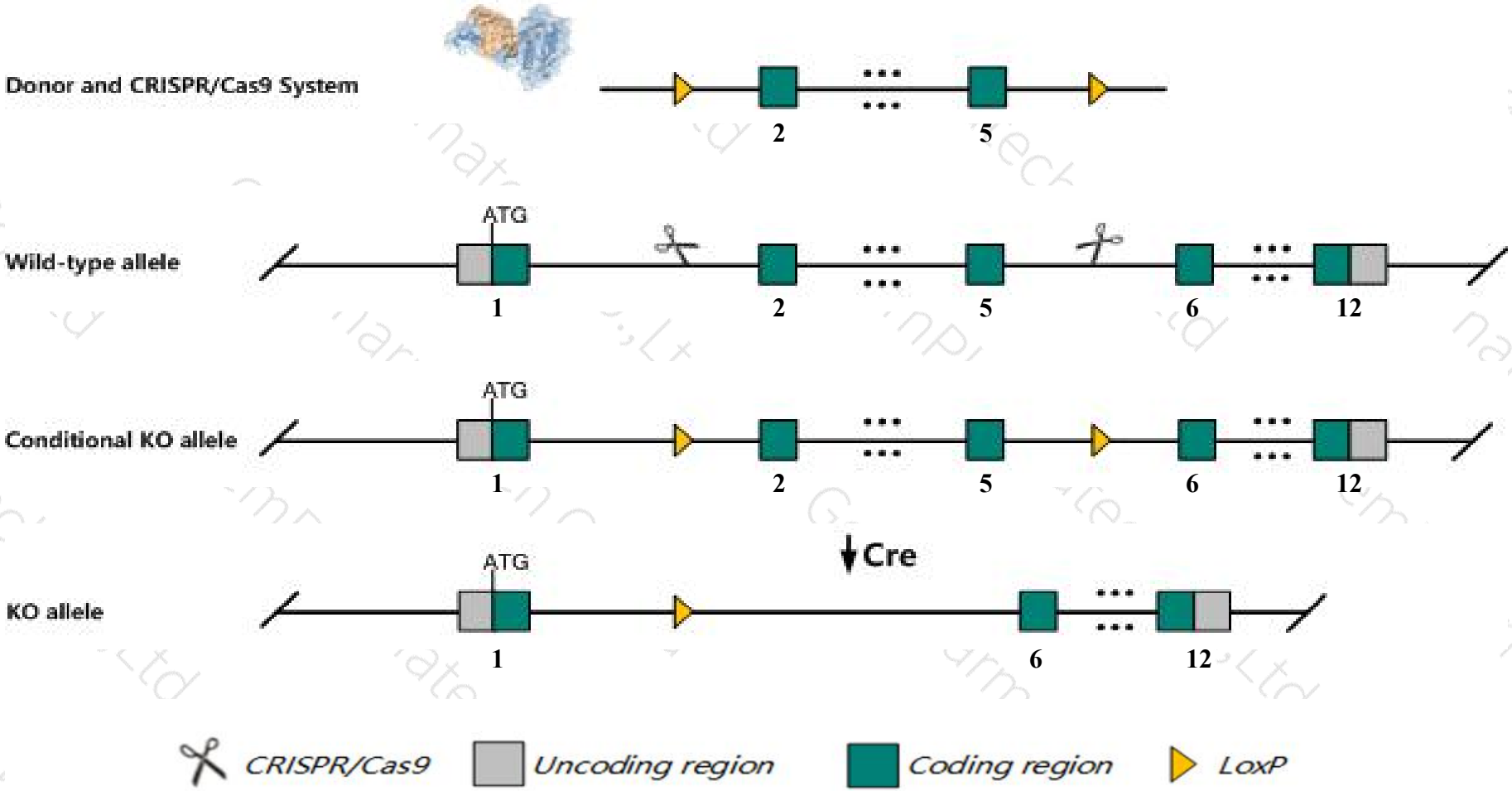
Designer: Xiaojing Li
Design Date: 2019-9-11
Reviewer: JiaYu

Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Lmna* gene has 7 transcripts. According to the structure of *Lmna* gene, exon2-exon5 of *Lmna-201* (ENSMUST00000029699.12) transcript is recommended as the knockout region.The region contains 580bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Lmna* 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,Homozygotes for targeted mutations exhibit retarded postnatal growth, muscular dystrophy, reduced fat stores, micrognathy, abnormal dentition, impaired gonadal development, malformed scapulae, hyperkeratosis, and die by 8 weeks of age.

- The *Lmna* 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

Lmna lamin A [Mus musculus (house mouse)]

Gene ID: 16905, updated on 7-Apr-2019

Summary

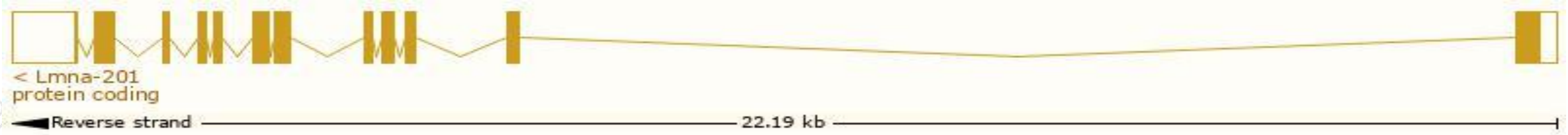
Official Symbol	Lmna provided by MGI
Official Full Name	lamin A provided by MGI
Primary source	MGI:MGI:96794
See related	Ensembl:ENSMUSG00000028063
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Dhe
Summary	This gene encodes a protein that is a member of the lamin family. Nuclear lamins, intermediate filament-like proteins, are the major components of the nuclear lamina, a protein meshwork associated with the inner nuclear membrane. This meshwork is thought to maintain the integrity of the nuclear envelope, participate in chromatin organization, and regulate gene transcription. Vertebrate lamins consist of two types, A and B. This protein is an A-type and is proposed to be developmentally regulated. In mouse deficiency of this gene is associated with muscular dystrophy. Mouse lines with different mutations in this gene serve as pathophysiological models for several human laminopathies. In humans, mutations in this gene lead to several diseases: Emery-Dreifuss muscular dystrophy, familial partial lipodystrophy, limb girdle muscular dystrophy, dilated cardiomyopathy, Charcot-Marie-Tooth disease, and Hutchinson-Gilford progeria syndrome. Alternative splicing results in multiple transcript variants that encode different protein isoforms. [provided by RefSeq, May 2013]
Expression	Broad expression in colon adult (RPKM 180.7), stomach adult (RPKM 89.6) and 19 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

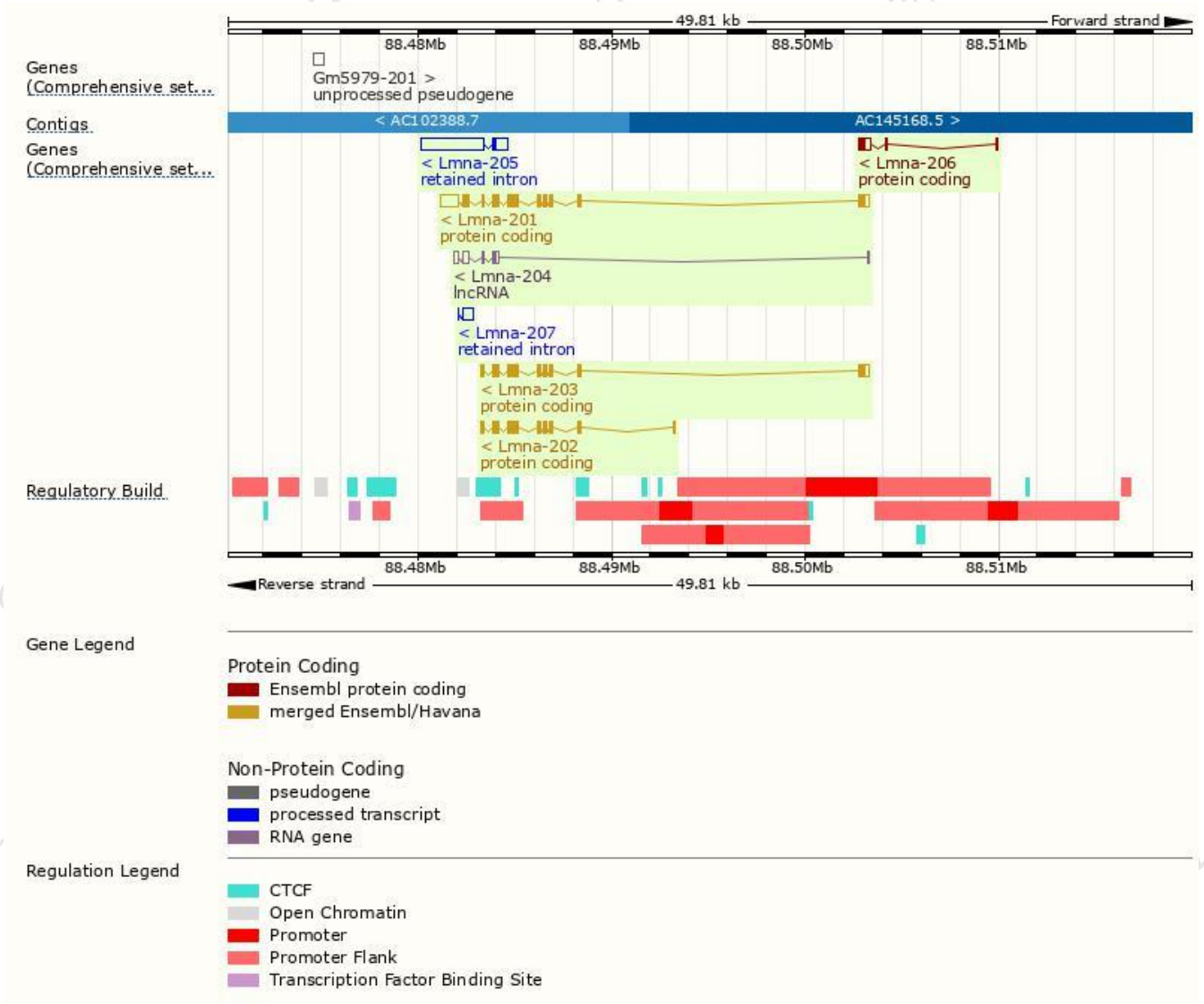
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Lmna-201	ENSMUST00000029699.12	3156	665aa	Protein coding	CCDS38482	P48678	TSL:1 GENCODE basic APPRIS P1
Lmna-203	ENSMUST00000120377.7	2021	574aa	Protein coding	CCDS50951	P48678	TSL:1 GENCODE basic
Lmna-202	ENSMUST00000036252.6	1536	462aa	Protein coding	CCDS38483	P48678	TSL:1 GENCODE basic
Lmna-206	ENSMUST00000149068.1	785	117aa	Protein coding	-	D3YUF7	CDS 3' incomplete TSL:2
Lmna-204	ENSMUST00000135494.3	901	No protein	Processed transcript	-	-	TSL:5
Lmna-205	ENSMUST00000147537.5	3948	No protein	Retained intron	-	-	TSL:1
Lmna-207	ENSMUST00000150496.1	543	No protein	Retained intron	-	-	TSL:1

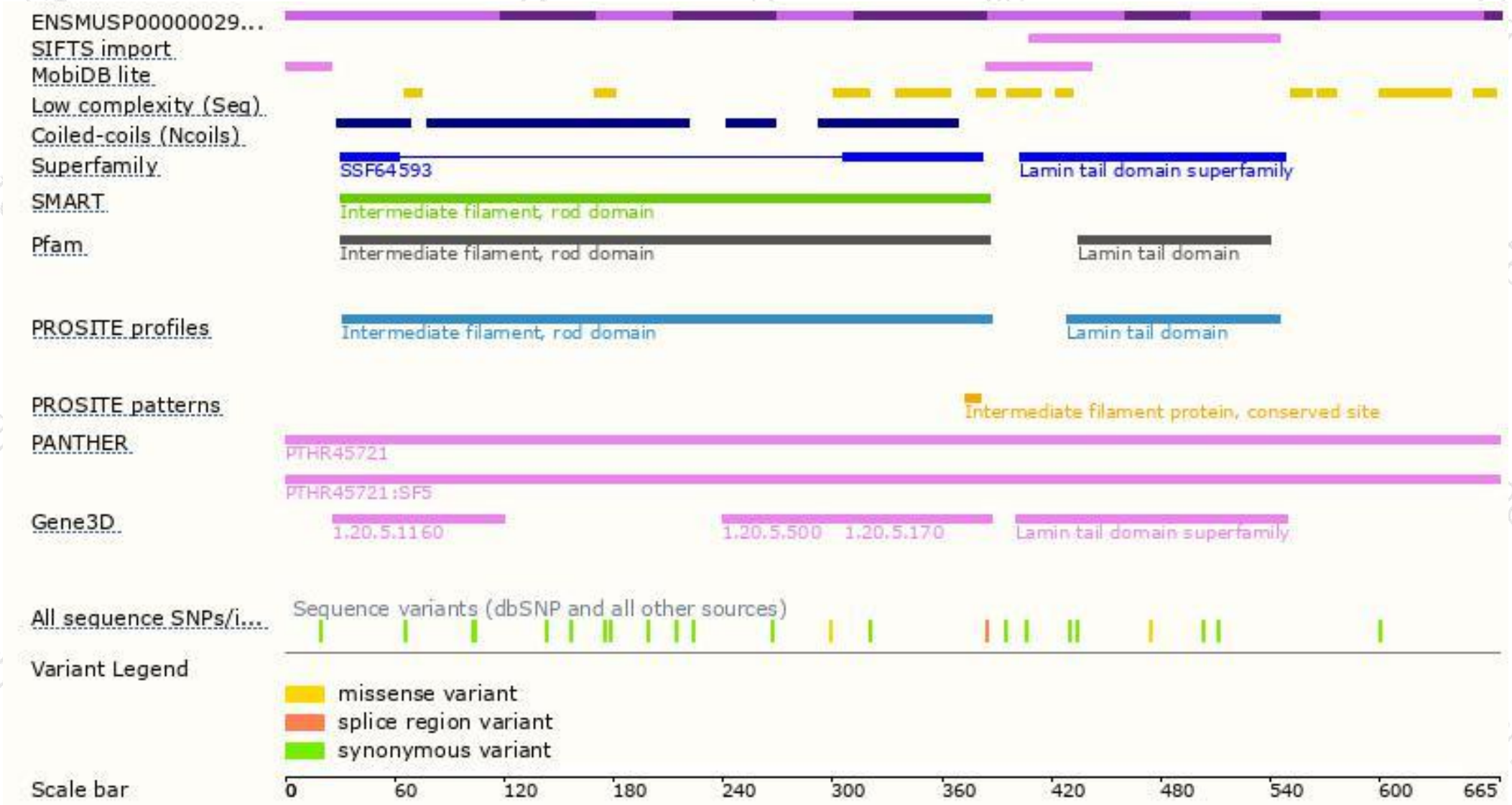
The strategy is based on the design of *Lmna-201* transcript,The transcription is shown below



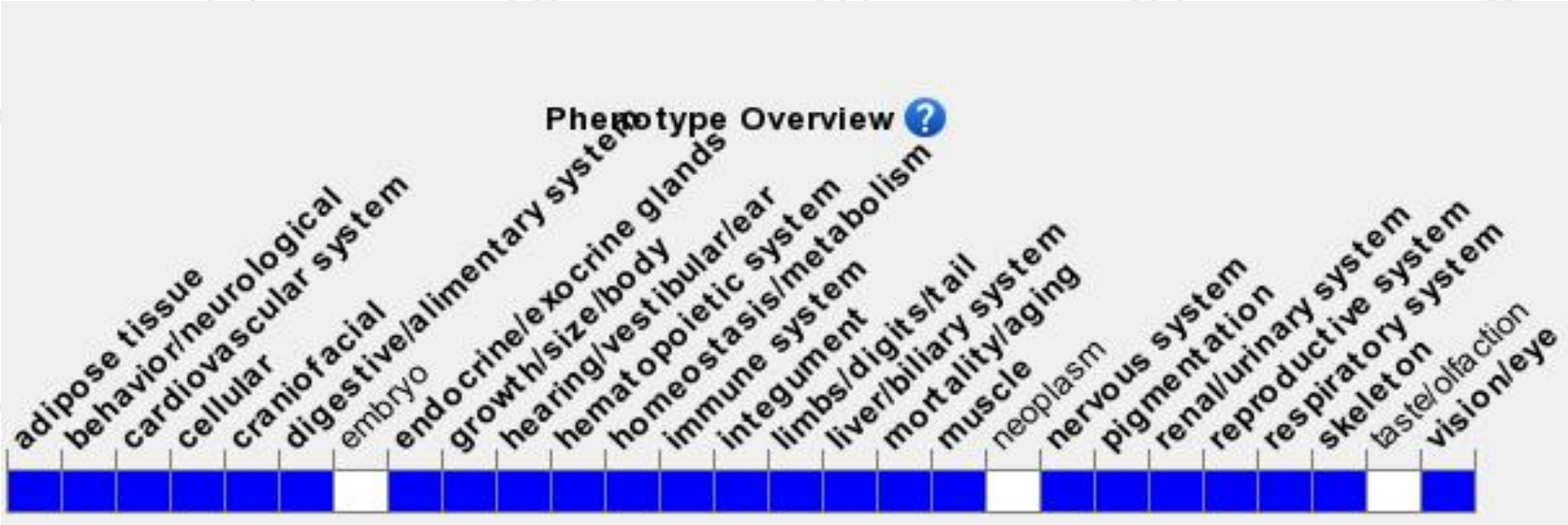
Genomic location distribution



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,Homozygotes for targeted mutations exhibit retarded postnatal growth, muscular dystrophy, reduced fat stores, micrognathy, abnormal dentition, impaired gonadal development, malformed scapulae, hyperkeratosis, and die by 8 weeks of age.

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