Lifr Cas9-CKO Strategy

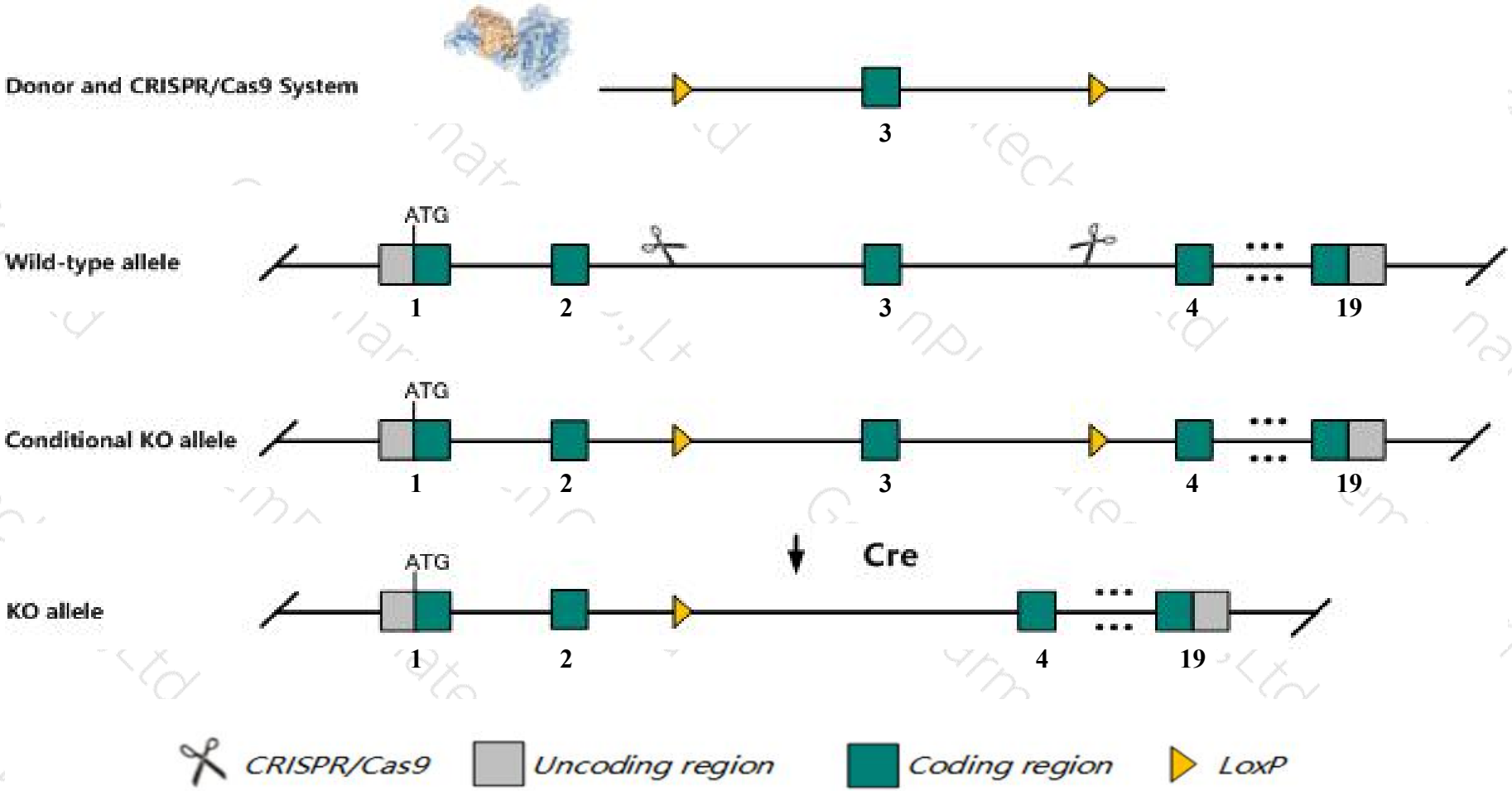
Designer: Xiaojing Li
Design Date: 2019-9-11
Reviewer: JiaYu

Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Lifr* gene has 8 transcripts. According to the structure of *Lifr* gene, exon3 of *Lifr-203* (ENSMUST00000171588.1) transcript is recommended as the knockout region.The region contains 131bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Lifr* 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 null mutations die as neonates with reduced numbers of facial and spinal motor neurons, neurons of the nucleus ambiguus, and astrocytes. Mutants also show impaired placentation, severe osteopenia, and low hepatic glycogen stores.

➢ The *Lifr* gene is located on the Chr15. 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

Lifr LIF receptor alpha [Mus musculus (house mouse)]

Gene ID: 16880, updated on 31-Jan-2019

Summary

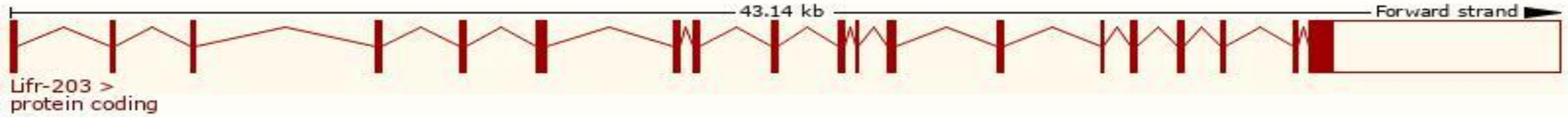
Official Symbol	Lifr provided by MGI
Official Full Name	LIF receptor alpha provided by MGI
Primary source	MGI:MGI:96788
See related	Ensembl:ENSMUSG00000054263
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	A230075M04Rik, AW061234, LIF
Expression	Broad expression in placenta adult (RPKM 14.4), liver adult (RPKM 13.6) and 23 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

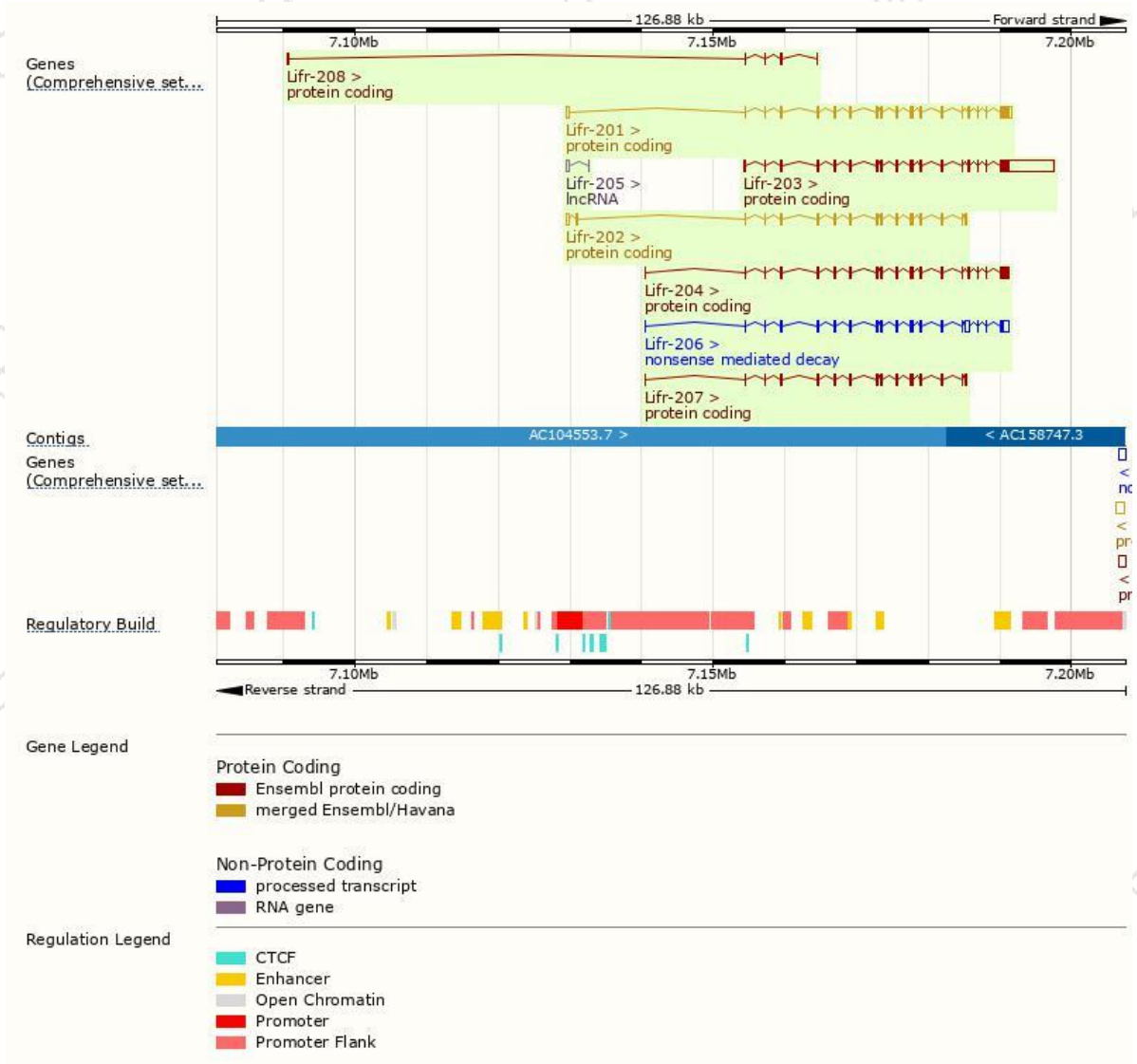
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Lifr-203	ENSMUST00000171588.1	9655	1092aa	Protein coding	CCDS27369	P42703	TSL:5 GENCODE basic APPRIS P3
Lifr-201	ENSMUST00000067190.11	4143	1092aa	Protein coding	CCDS27369	P42703	TSL:1 GENCODE basic APPRIS P3
Lifr-204	ENSMUST00000226471.1	3392	1092aa	Protein coding	CCDS27369	P42703	GENCODE basic APPRIS P3
Lifr-202	ENSMUST00000164529.8	2844	719aa	Protein coding	CCDS49577	P42703	TSL:1 GENCODE basic APPRIS ALT2
Lifr-207	ENSMUST00000227727.1	2489	719aa	Protein coding	CCDS49577	P42703	GENCODE basic APPRIS ALT2
Lifr-208	ENSMUST00000228723.1	577	140aa	Protein coding	-	A0A2I3BRT7	CDS 3' incomplete
Lifr-206	ENSMUST00000226934.1	3891	719aa	Nonsense mediated decay	CCDS49577	P42703	
Lifr-205	ENSMUST00000226826.1	299	No protein	lncRNA	-	-	

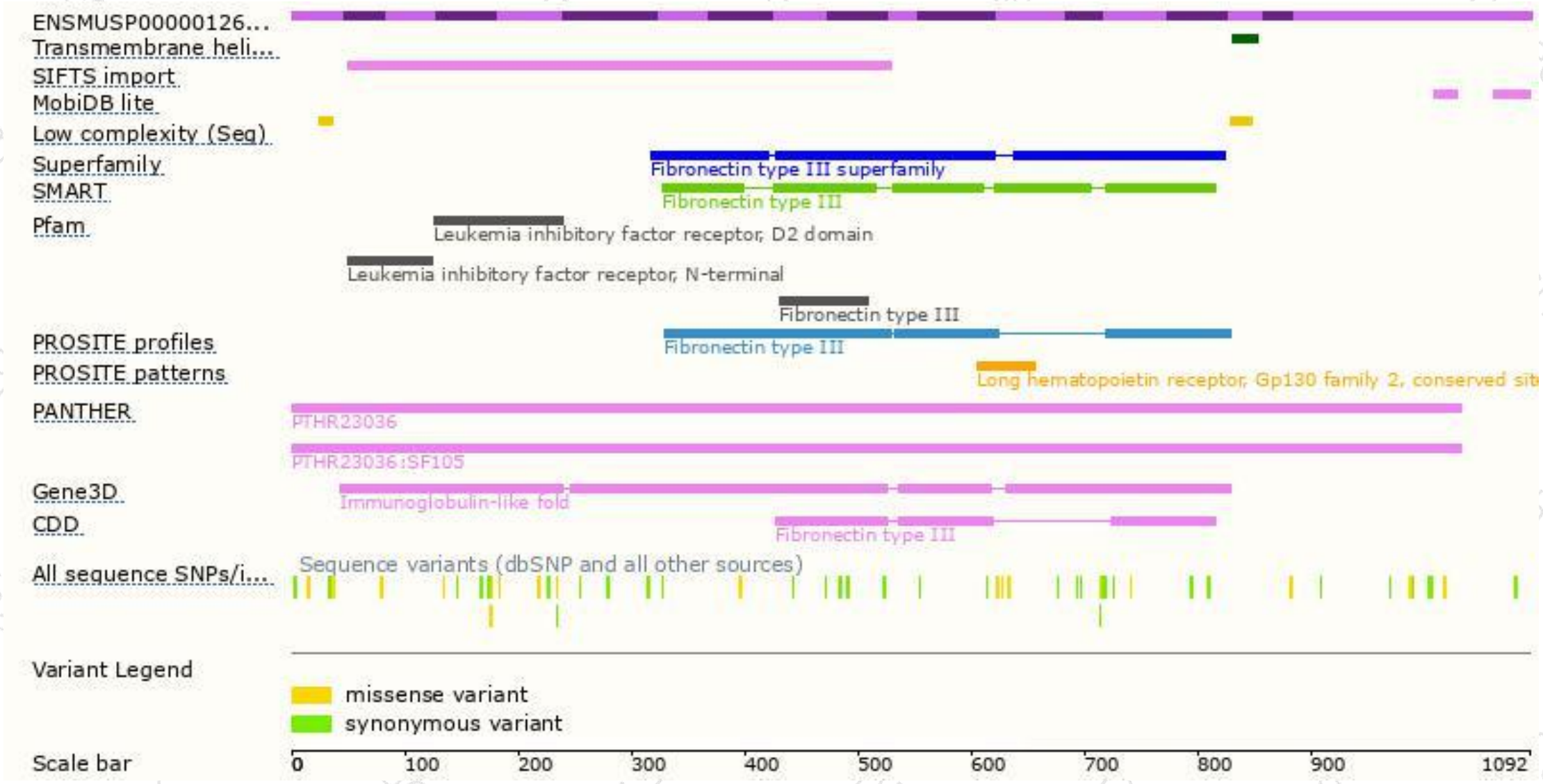
The strategy is based on the design of *Lifr-203* transcript,The transcription is shown below



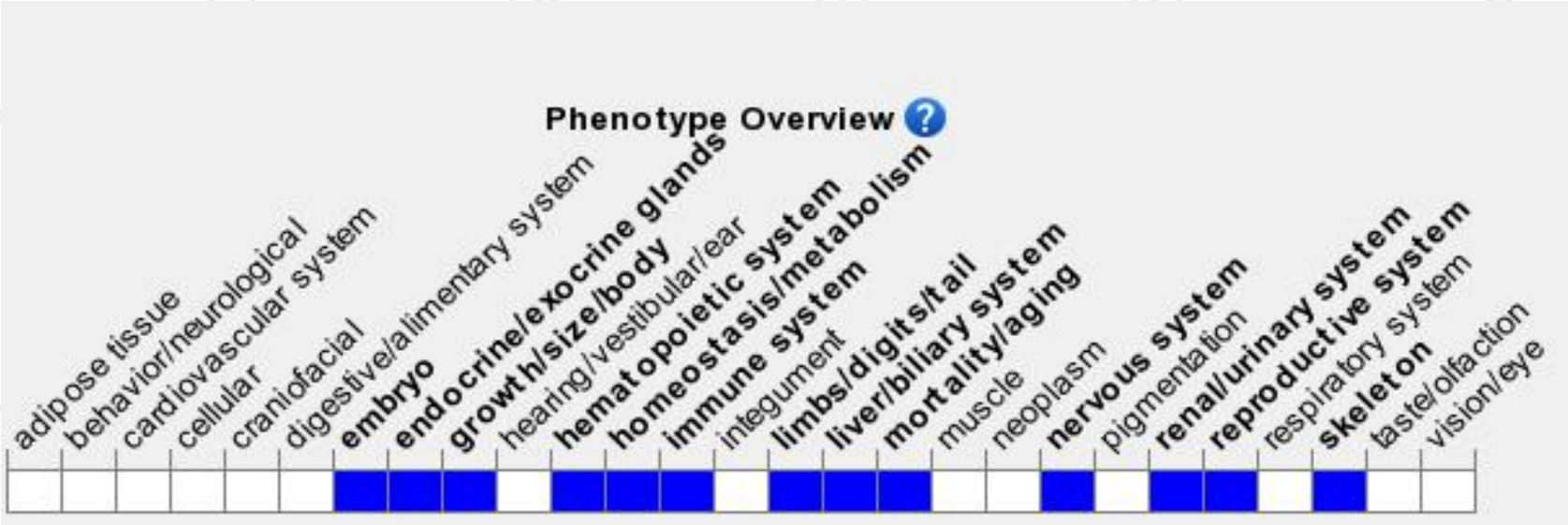
Genomic location distribution



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,Homozygotes for targeted null mutations die as neonates with reduced numbers of facial and spinal motor neurons, neurons of the nucleus ambiguus, and astrocytes. Mutants also show impaired placentation, severe osteopenia, and low hepatic glycogen stores.

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