Ndrg1 Cas9-CKO Strategy

Designer: Yang Zeng
Reviewer: Ruirui Zhang

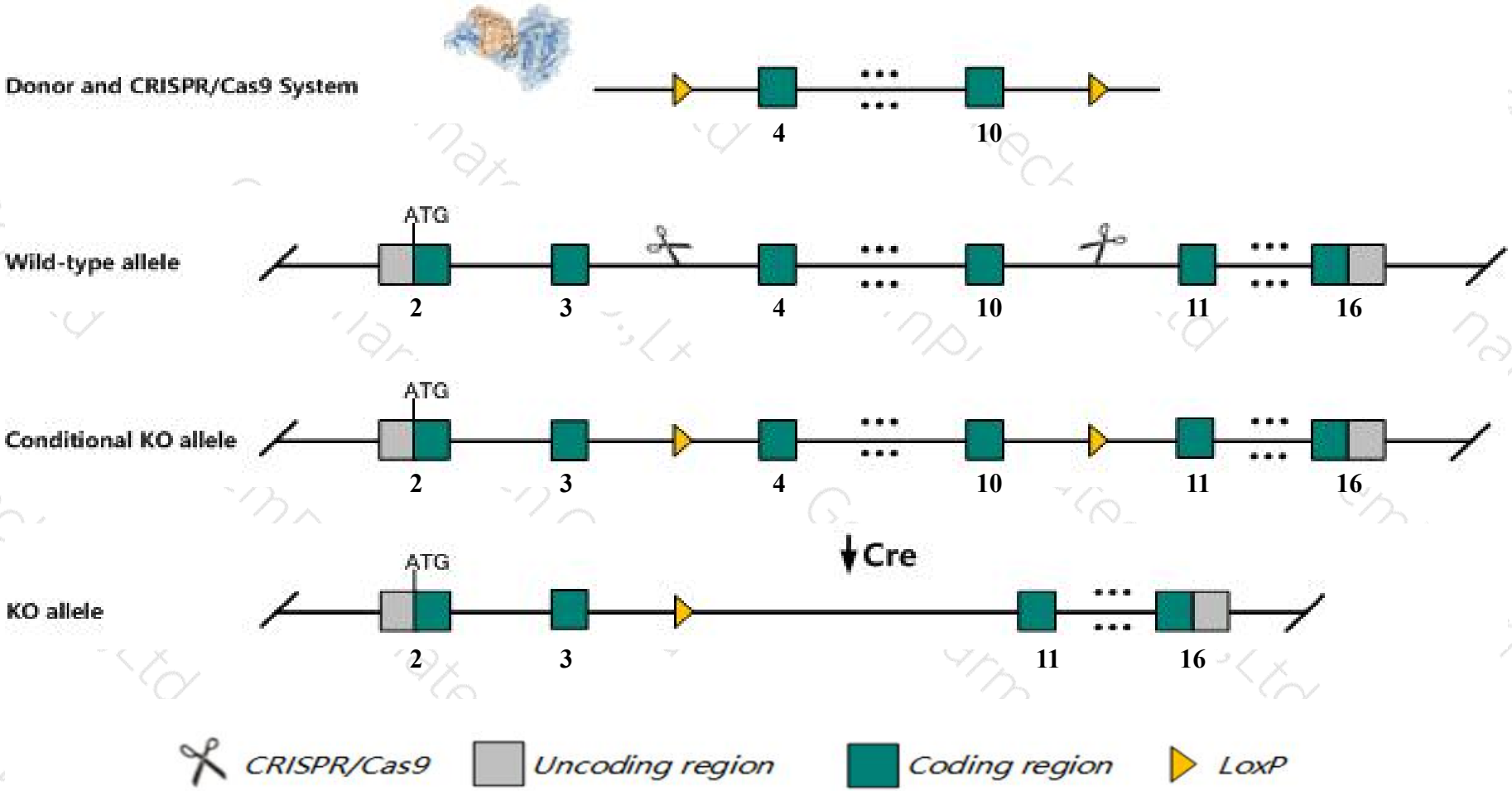
Design Date: 2019-12-23

Project Overview

Project Name	***Ndrg1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ndrg1* gene. The schematic diagram is as follows:



Technical routes

- The *Ndrg1* gene has 13 transcripts. According to the structure of *Ndrg1* gene, exon4-exon10 of *Ndrg1-201* (ENSMUST00000005256.13) transcript is recommended as the knockout region.The region contains 599bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Ndrg1* 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 exhibit a progressive demyelinating disorder of the peripheral nerves with hindlimb weakness, some mice die between 1 to 10 months.

- The *Ndrg1* 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

Ndrg1 N-myc downstream regulated gene 1 [*Mus musculus* (house mouse)]

Gene ID: 17988, updated on 21-Oct-2019

Summary

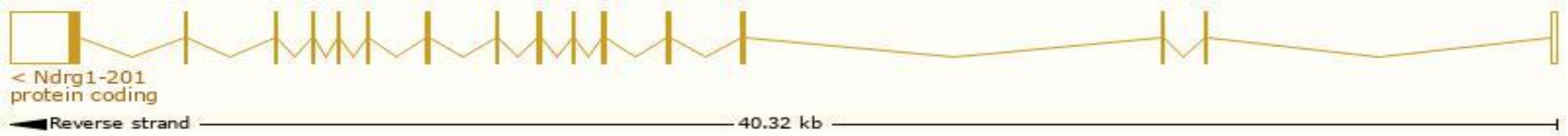
Official Symbol	Ndrg1 provided by MGI
Official Full Name	N-myc downstream regulated gene 1 provided by MGI
Primary source	MGI:MGI:1341799
See related	Ensembl:ENSMUSG00000005125
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	RTP; DRG1; NMSL; Ndr1; Ndrl; TDD5; CAP43; CMT4D; HMSNL; PROXY1
Expression	Biased expression in kidney adult (RPKM 707.1), liver E18 (RPKM 98.7) and 4 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

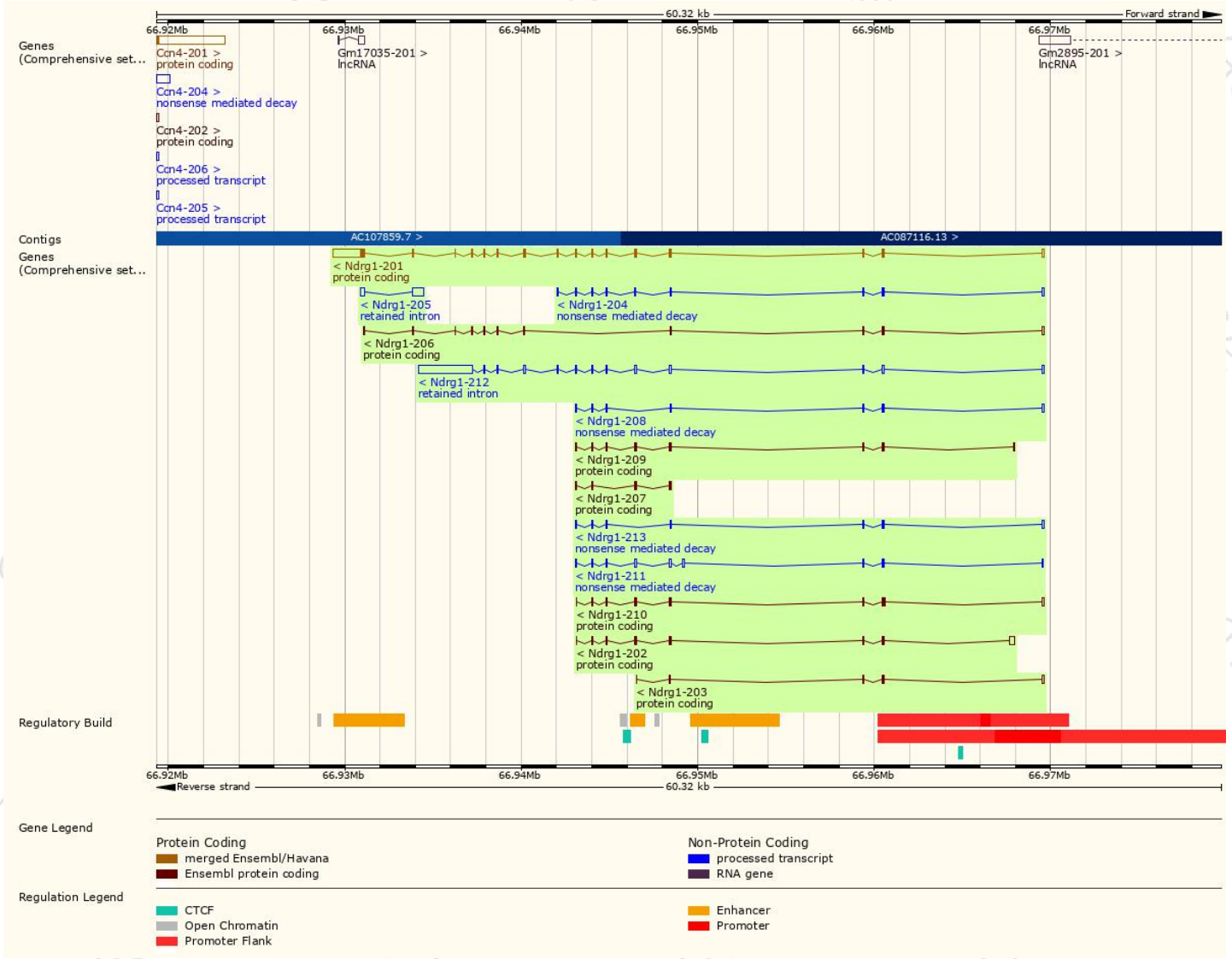
The gene has 13 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Translation ID	Biotype	CCDS	UniProt	Flags
Ndrg1-201	ENSMUST00000005256.13	2889	394aa	ENSMUSP00000005256.6	Protein coding	CCDS37092	Q545R3 Q62433	TSL:1 GENCODE basic APPRIS P1
Ndrg1-202	ENSMUST00000163496.7	738	155aa	ENSMUSP00000130584.1	Protein coding	-	E9Q5I8	CDS 3' incomplete TSL:5
Ndrg1-210	ENSMUST00000170903.7	667	157aa	ENSMUSP00000127302.1	Protein coding	-	E9Q3F9	CDS 3' incomplete TSL:5
Ndrg1-209	ENSMUST00000168979.7	583	174aa	ENSMUSP00000126985.1	Protein coding	-	E9PVF3	CDS 3' incomplete TSL:3
Ndrg1-206	ENSMUST00000166420.7	577	141aa	ENSMUSP00000127099.1	Protein coding	-	E9Q514	CDS 3' incomplete TSL:3
Ndrg1-207	ENSMUST00000167817.7	356	119aa	ENSMUSP00000127075.1	Protein coding	-	F6VLR8	CDS 5' and 3' incomplete TSL:3
Ndrg1-203	ENSMUST00000164070.1	316	53aa	ENSMUSP00000126091.1	Protein coding	-	E9Q147	CDS 3' incomplete TSL:5
Ndrg1-211	ENSMUST00000171266.7	759	39aa	ENSMUSP00000129093.1	Nonsense mediated decay	-	E9Q0J8	TSL:5
Ndrg1-204	ENSMUST00000164675.7	591	93aa	ENSMUSP00000130150.1	Nonsense mediated decay	-	E9Q7V2	TSL:3
Ndrg1-208	ENSMUST00000168542.7	473	49aa	ENSMUSP00000127940.1	Nonsense mediated decay	-	E9PZC7	TSL:5
Ndrg1-213	ENSMUST00000172447.7	444	53aa	ENSMUSP00000130281.1	Nonsense mediated decay	-	E9Q7G8	TSL:5
Ndrg1-212	ENSMUST00000171569.7	4049	No protein	-	Retained intron	-	-	TSL:1
Ndrg1-205	ENSMUST00000165966.1	849	No protein	-	Retained intron	-	-	TSL:2

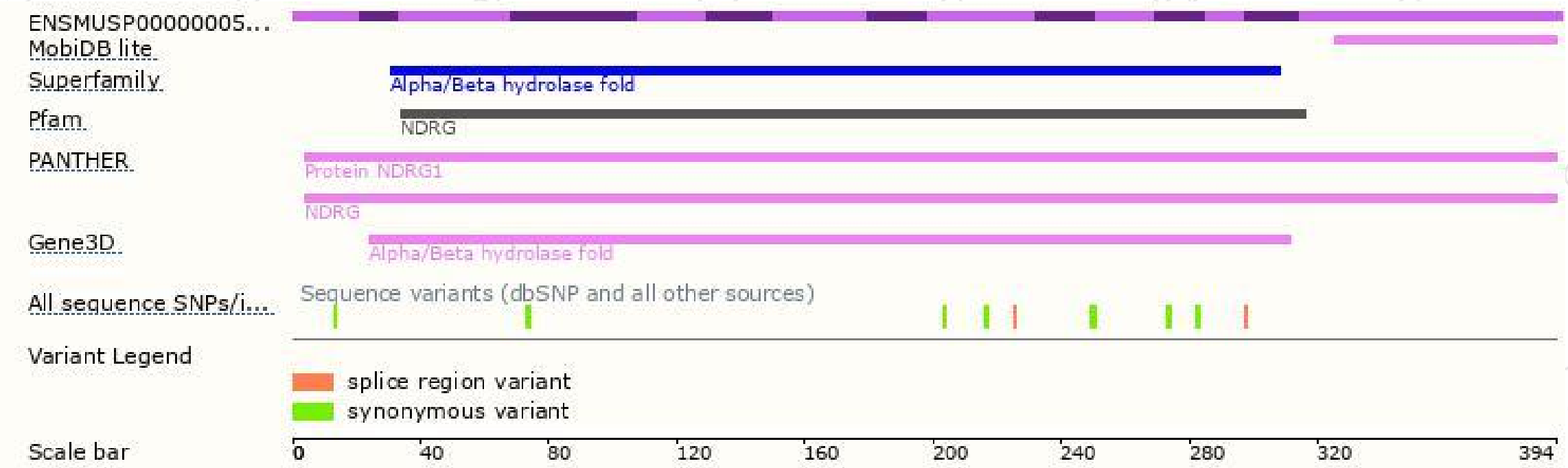
The strategy is based on the design of *Ndrg1-201* transcript,The transcription is shown below



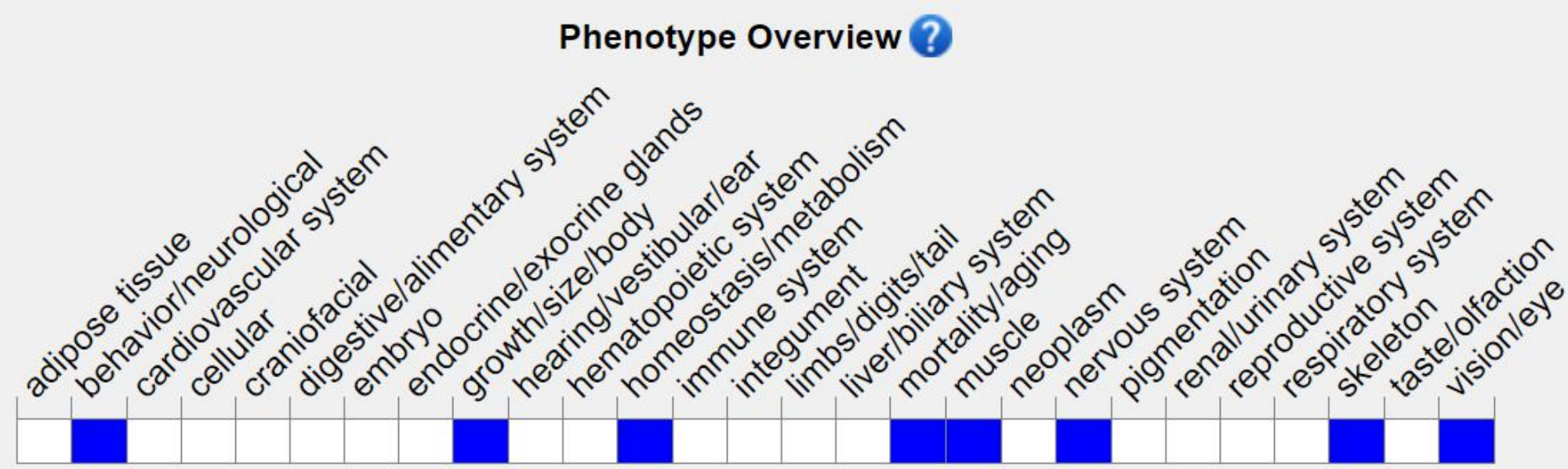
Genomic location distribution



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 null mice exhibit a progressive demyelinating disorder of the peripheral nerves with hindlimb weakness, some mice die between 1 to 10 months.

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