Afap1l2 Cas9-CKO Strategy

Designer: JiaYu

Reviewer: Xiaojing Li

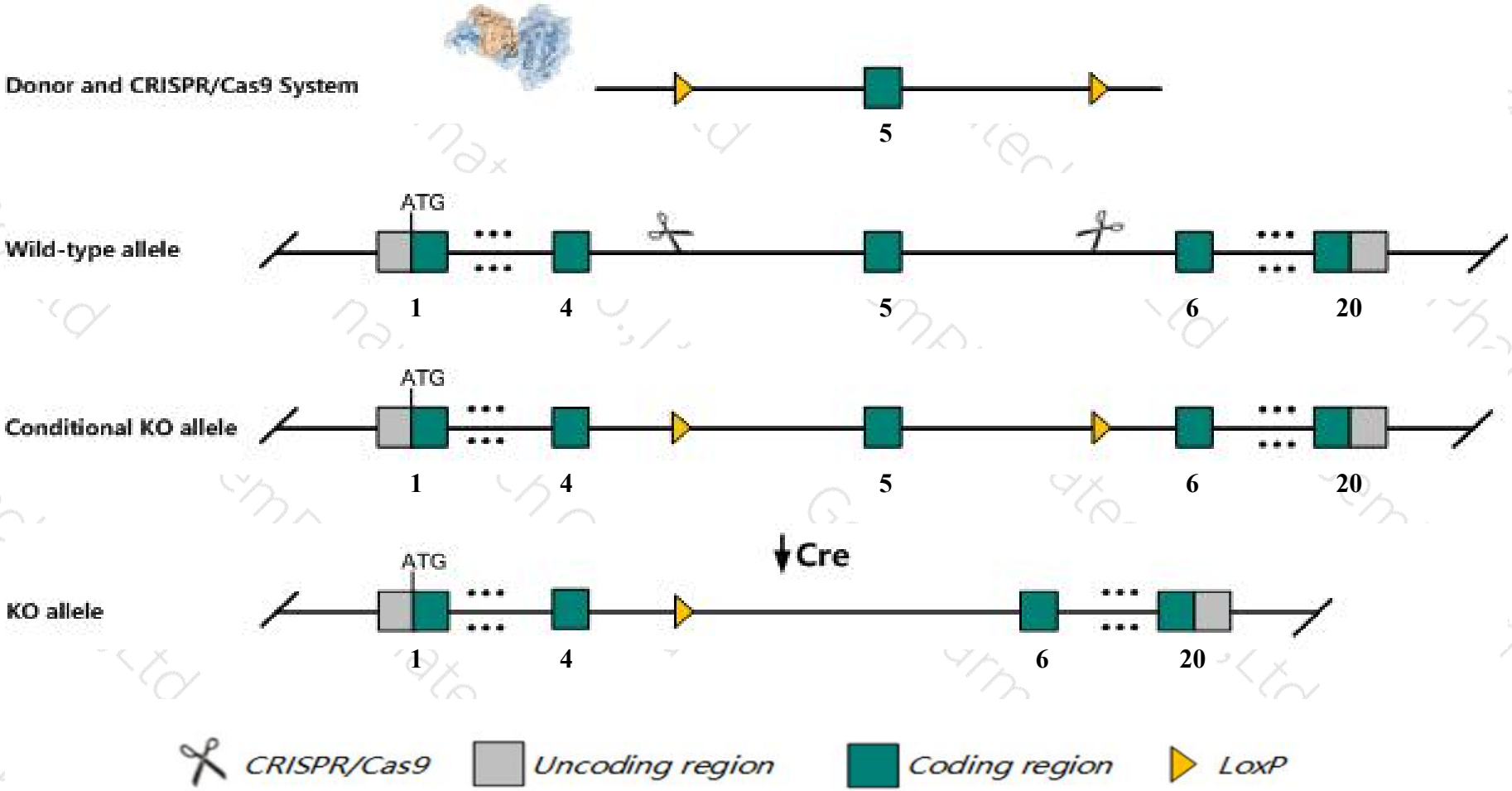
Design Date: 2020-8-24

Project Overview

Project Name	***Afap1l2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Afap1l2* gene. The schematic diagram is as follows:



Technical routes

➢ The *Afap1l2* gene has 8 transcripts. According to the structure of *Afap1l2* gene, exon5 of *Afap1l2-201*(ENSMUST00000111584.8) transcript is recommended as the knockout region.The region contains 95bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Afap1l2* 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,mice homozygous for a null allele exhibit a thicker tracheal epithelium and a transient tracheal epithelial hyperplasia during airway repair after injury.

➢Some amino acids will remain at the N-terminus and some functions may be retained.

➢Transcript 208 CDS 5' and 3' incomplete the influences is unknown.

➢ The *Afap1l2* gene is located on the Chr19. 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

Afap1l2 actin filament associated protein 1-like 2 [Mus musculus (house mouse)]

Gene ID: 226250, updated on 13-Mar-2020

Summary

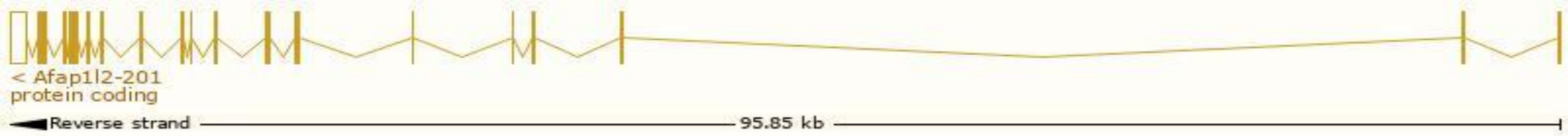
Official Symbol	Afap1l2 provided by MGI
Official Full Name	actin filament associated protein 1-like 2 provided byMGI
Primary source	MGI:MGI:2147658
See related	Ensembl:ENSMUSG00000025083
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AU041783, C86904, mKIAA1914
Expression	Broad expression in placenta adult (RPKM 15.2), cerebellum adult (RPKM 10.4) and 21 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

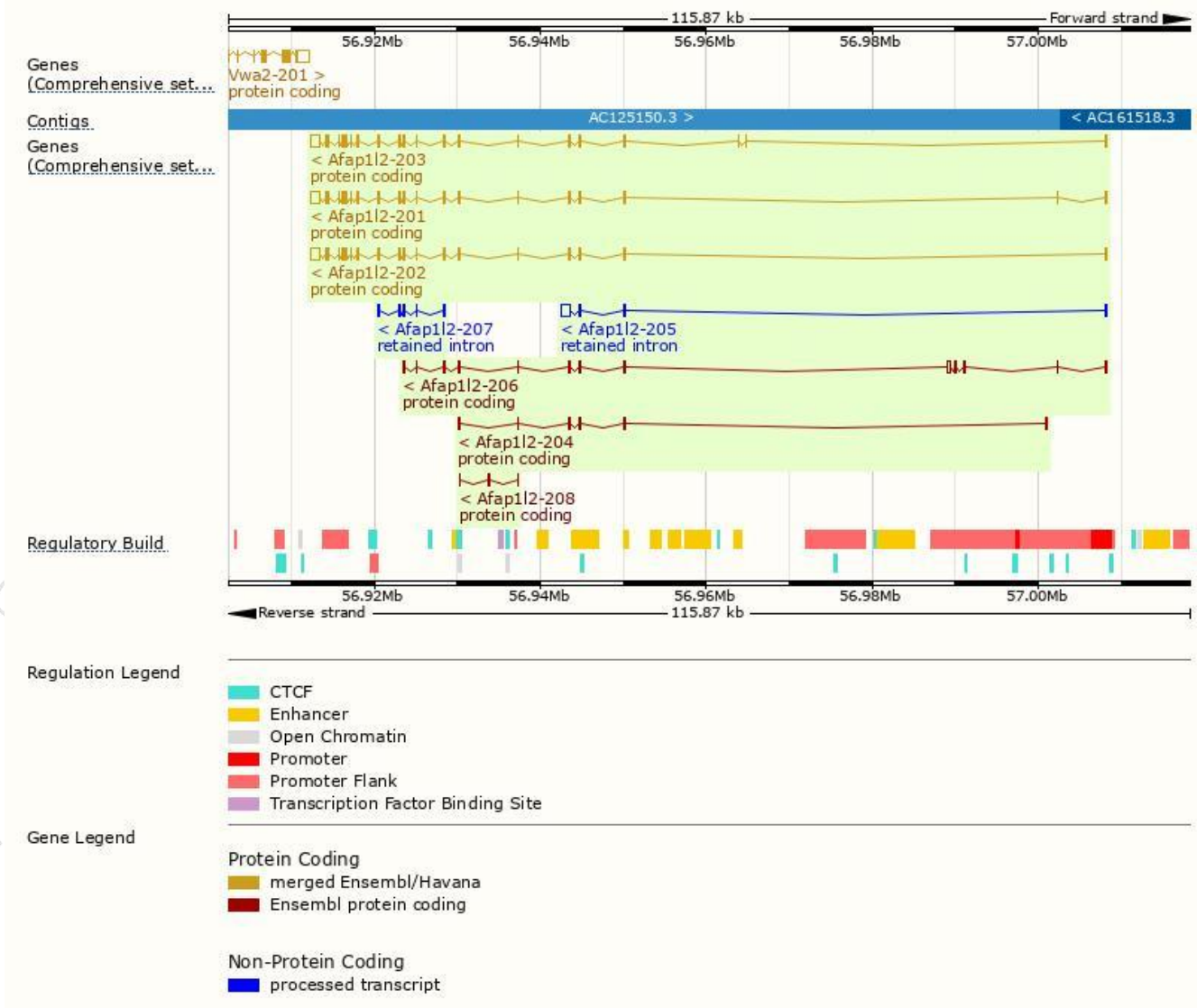
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Afap1l2-203	ENSMUST00000122359.7	3667	769aa	Protein coding	CCDS29924	A0A0R4J1L6	TSL:1 GENCODE basic APPRIS P3
Afap1l2-201	ENSMUST00000111584.8	3597	843aa	Protein coding	CCDS50477	Q5DTU0	TSL:1 GENCODE basic APPRIS ALT2
Afap1l2-202	ENSMUST00000118800.7	3531	825aa	Protein coding	CCDS50476	Q5DTU0	TSL:1 GENCODE basic APPRIS ALT2
Afap1l2-206	ENSMUST00000148049.7	1724	264aa	Protein coding	-	D3YYQ6	CDS 3' incomplete TSL:1
Afap1l2-204	ENSMUST00000126964.7	681	134aa	Protein coding	-	D3YZD3	CDS 3' incomplete TSL:2
Afap1l2-208	ENSMUST00000225394.1	93	31aa	Protein coding	-	A0A286YCN9	CDS 5' and 3' incomplete
Afap1l2-205	ENSMUST00000132851.1	1327	No protein	Retained intron	-	-	TSL:2
Afap1l2-207	ENSMUST00000155467.1	776	No protein	Retained intron	-	-	TSL:2

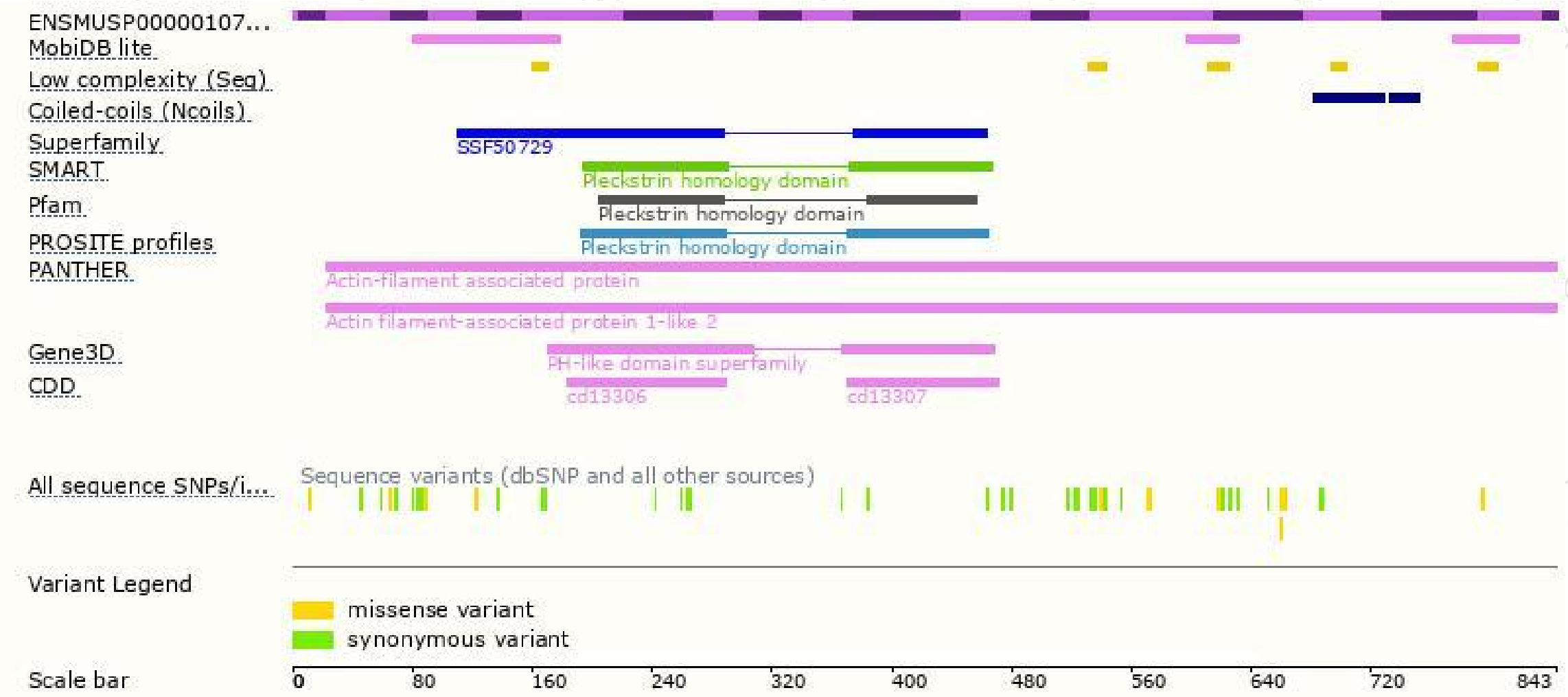
The strategy is based on the design of *Afap1l2-201* transcript,the transcription is shown below:



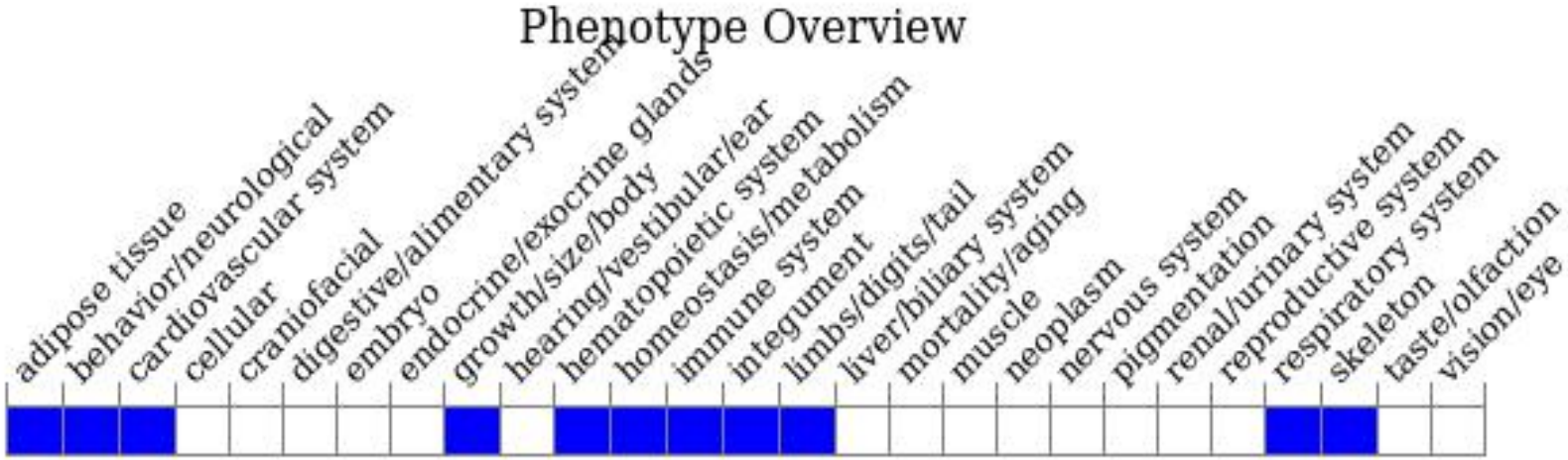
Genomic location distribution



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,mice homozygous for a null allele exhibit a thicker tracheal epithelium and a transient tracheal epithelial hyperplasia during airway repair after injury.

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