Nfe2 Cas9-KO Strategy

Designer: Ruirui Zhang

Reviewer: Xueting Zhang

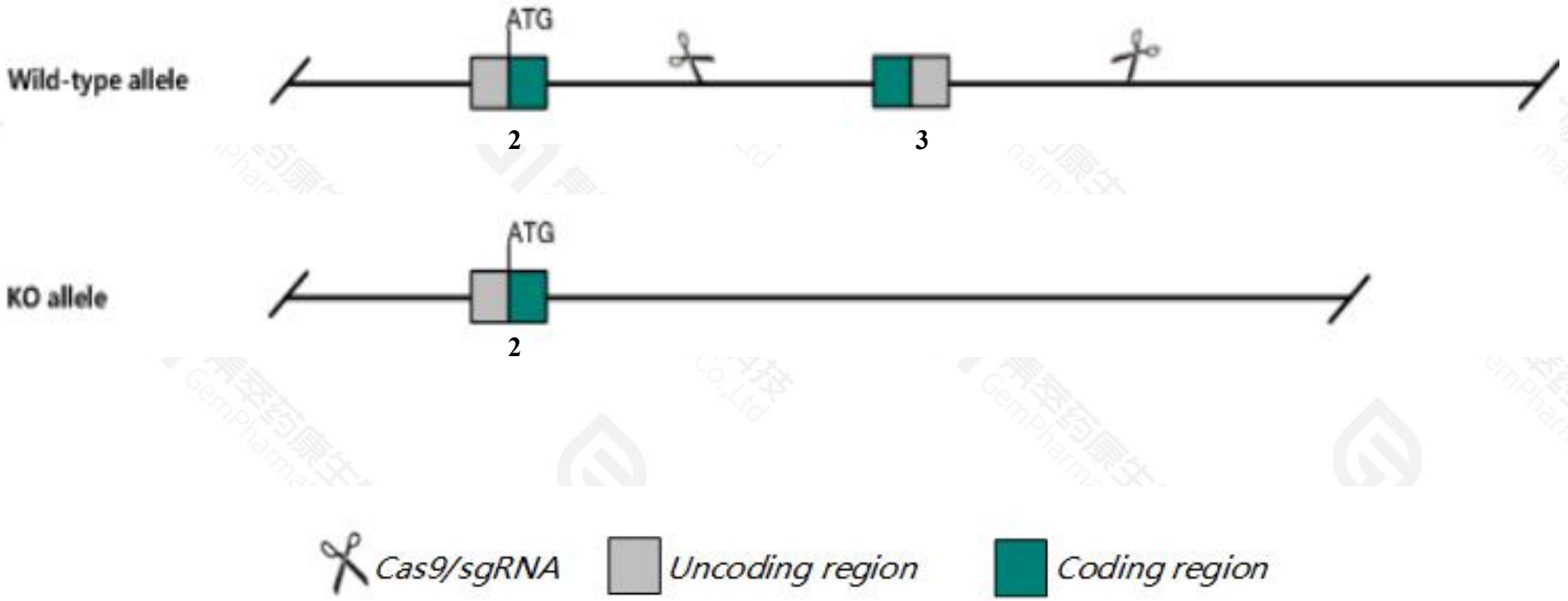
Design Date: 2021/7/6

Project Overview

Project Name	***Nfe2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Nfe2* gene. The schematic diagram is as follows:



Technical routes

➢ The *Nfe2* gene has 10 transcripts. According to the structure of *Nfe2* gene, exon3 of *Nfe2-201*(ENSMUST00000075192.13) transcript is recommended as the knockout region.The region contains most of the coding sequence.Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Nfe2* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

Notice

➢ According to the existing MGI data,homozygotes for a targeted null mutation lack platelets and most die as neonates from internal bleeding. Survivors exhibit hypochromia, reticulocytosis, and splenomegaly.

➢The *Hnrnpa1* gene is close to the *Nfe2* gene, and this strategy may affect the 3'terminal regulation function of *Hnrnpa1* gene.

➢ The *Nfe2* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

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Nfe2 nuclear factor, erythroid derived 2 [*Mus musculus* (house mouse)]

Download Datasets

Gene ID: 18022, updated on 23-Jun-2021

Summary

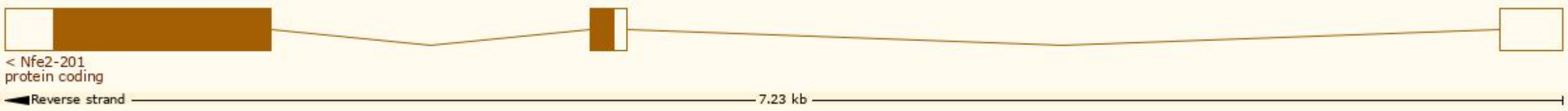
Official Symbol	Nfe2 provided by MGI
Official Full Name	nuclear factor, erythroid derived 2 provided by MGI
Primary source	MGI:MGI:97308
See related	Ensembl:ENSMUSG00000058794
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	p4; p45; NF-E; NF-E2; p45NF; p45nf-; p45NFE2; p45nf-e2; NF-E2/P45
Expression	Biased expression in liver E14.5 (RPKM 63.0), liver E14 (RPKM 51.5) and 2 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

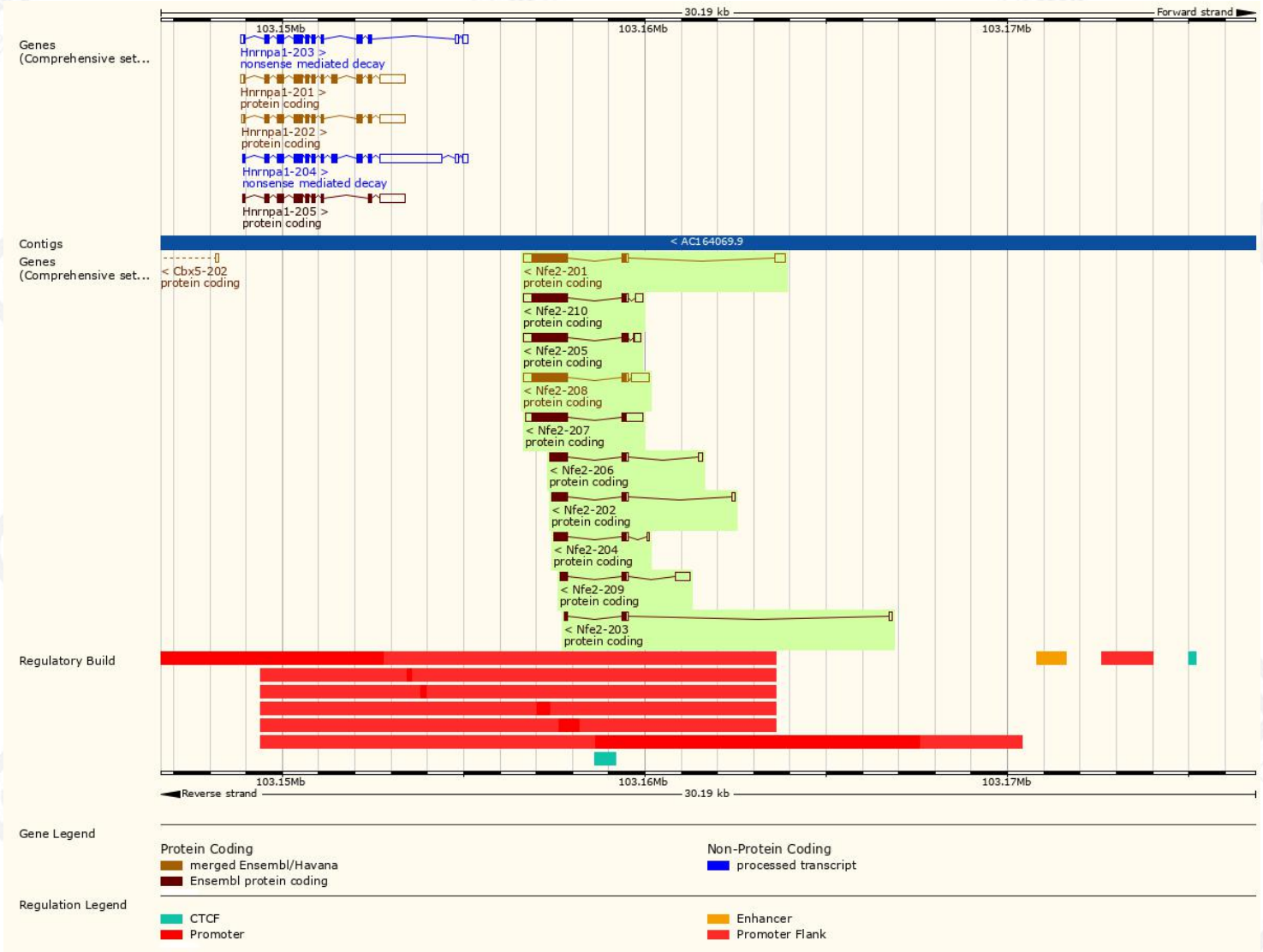
The gene has 10 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt Match	Flags
Nfe2-210	ENSMUST00000156927.8	1608	373aa	Protein coding	CCDS27900	A0A0R4J0Y5	GENCODE basic APPRIS P1 TSL:5
Nfe2-209	ENSMUST00000154510.8	784	108aa	Protein coding	-	D3Z4E8	TSL:2 CDS 3' incomplete
Nfe2-208	ENSMUST00000149111.8	1878	373aa	Protein coding	CCDS27900	A0A0R4J0Y5	GENCODE basic APPRIS P1 TSL:1
Nfe2-207	ENSMUST00000134554.2	1761	373aa	Protein coding	CCDS27900	A0A0R4J0Y5	GENCODE basic APPRIS P1 TSL:2
Nfe2-206	ENSMUST00000134193.8	774	202aa	Protein coding	-	D3YUM3	TSL:3 CDS 3' incomplete
Nfe2-205	ENSMUST00000133600.8	1605	396aa	Protein coding	-	D3Z224	GENCODE basic TSL:5
Nfe2-204	ENSMUST00000132836.8	607	164aa	Protein coding	-	D3Z0J1	TSL:2 CDS 3' incomplete
Nfe2-203	ENSMUST00000132438.2	362	71aa	Protein coding	-	D3Z706	TSL:2 CDS 3' incomplete
Nfe2-202	ENSMUST00000131364.8	708	185aa	Protein coding	-	D3Z6L9	TSL:5 CDS 3' incomplete
Nfe2-201	ENSMUST00000075192.13	1697	373aa	Protein coding	CCDS27900	A0A0R4J0Y5	GENCODE basic APPRIS P1 TSL:1

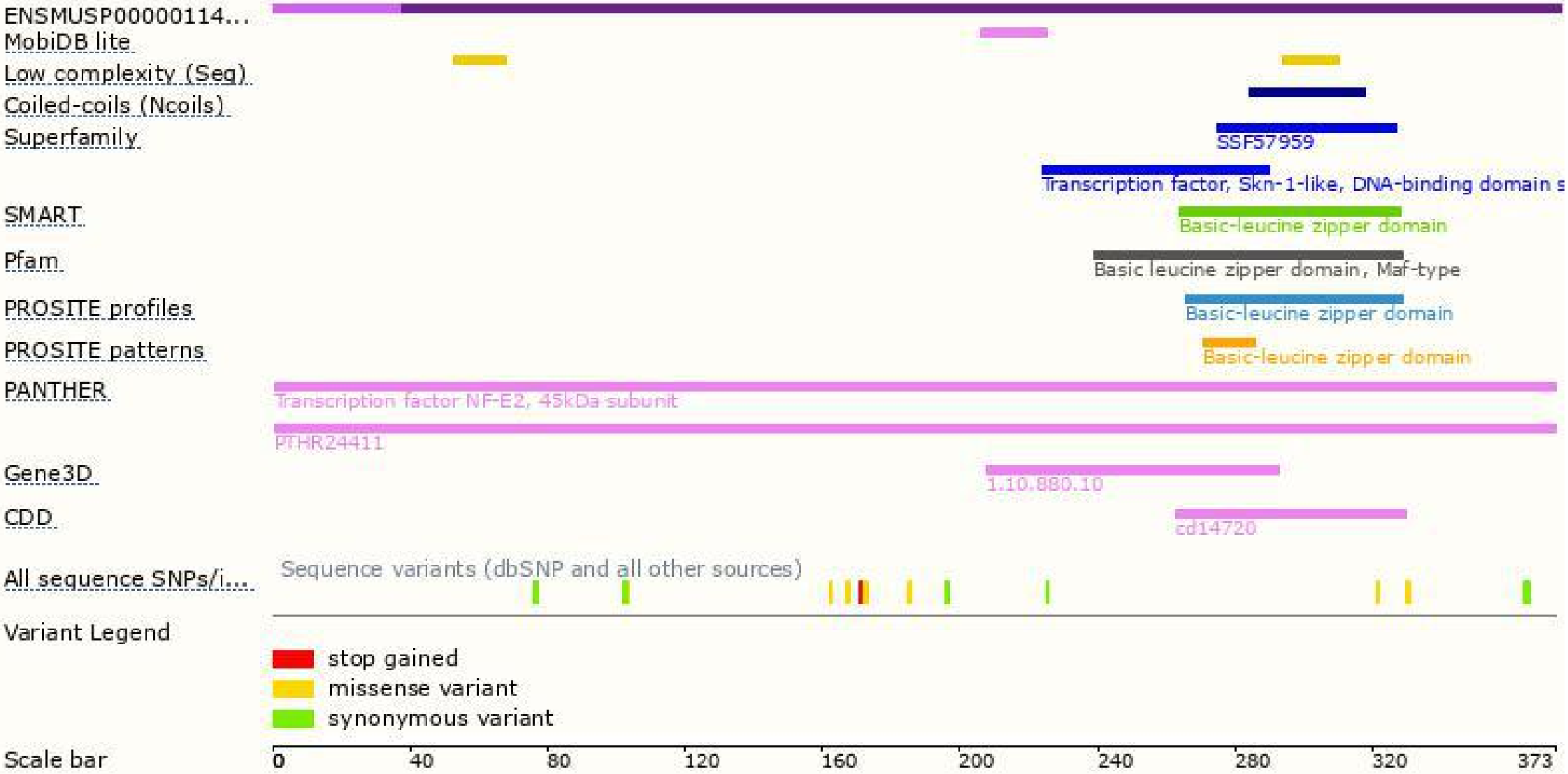
The strategy is based on the design of *Nfe2-201* transcript,the transcription is shown below:



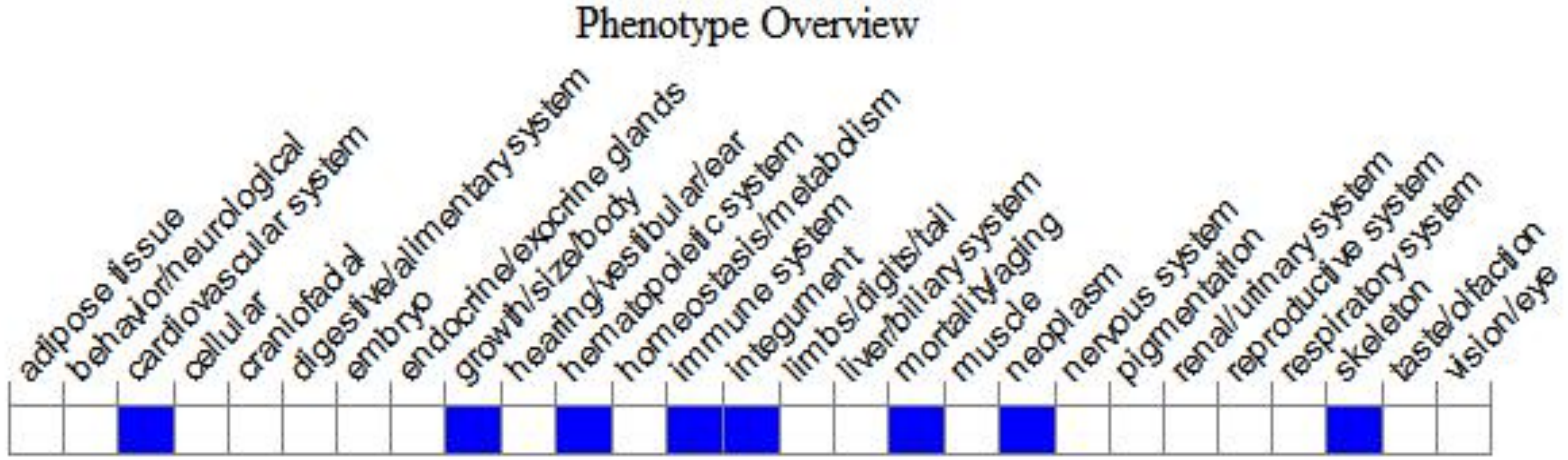
Genomic location distribution



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 a targeted null mutation lack platelets and most die as neonates from internal bleeding. Survivors exhibit hypochromia, reticulocytosis, and splenomegaly.

If you have any questions, you are welcome to inquire.

Tel: 025-5864 1534

集萃药康生物科技
GemPharmatech Co.,Ltd