Emilin2 Cas9-CKO Strategy

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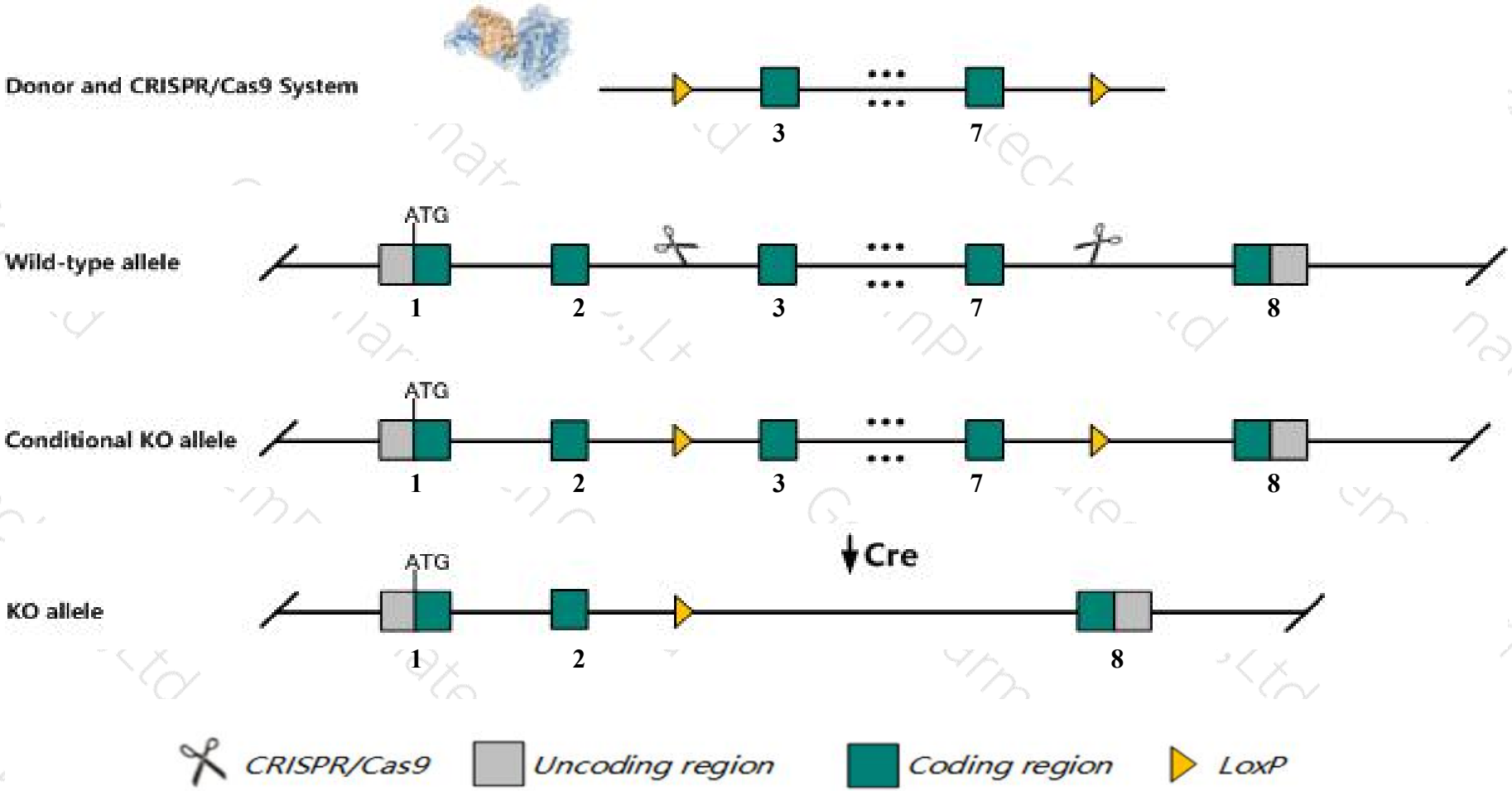
Date:2020-02-21

Project Overview

Project Name	***Emilin2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Emilin2* gene. The schematic diagram is as follows:



Technical routes

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- The *Emilin2* gene has 11 transcripts. According to the structure of *Emilin2* gene, exon3-exon7 of *Emilin2-203* (ENSMUST00000233057.1) transcript is recommended as the knockout region.The region contains 2621bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Emilin2* 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 knock-out allele exhibit disruptions in platelet activation, thrombus formation and clot retraction.
- Transcript *Emilin2*-208 may not be affected.
- The *Emilin2* gene is located on the Chr17. 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）

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Emilin2 elastin microfibril interfacer 2 [*Mus musculus* (house mouse)]

Gene ID: 246707, updated on 24-Oct-2019

Summary

Official Symbol Emilin2 provided by MGI
Official Full Name elastin microfibril interfacer 2 provided by MGI
Primary source MGI:MGI:2389136
See related Ensembl:ENSMUSG00000024053
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 FOAP-10
Expression Broad expression in subcutaneous fat pad adult (RPKM 10.2), mammary gland adult (RPKM 9.7) and 21 other tissues See more
Orthologs human all

Genomic context

Location: 17; 17 E1.3

See Emilin2 in Genome Data Viewer

Exon count: 9

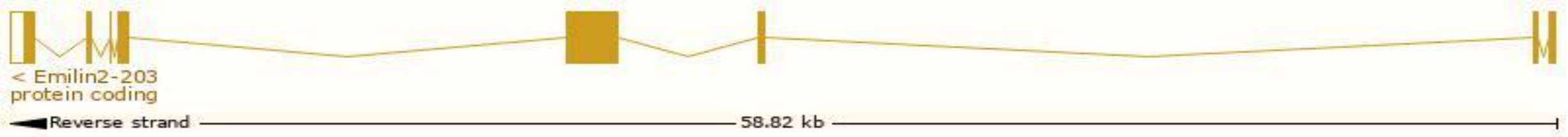
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	17	NC_000083.6 (71252172..71311556, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	17	NC_000083.5 (71601516..71660305, complement)

Transcript information（Ensembl）

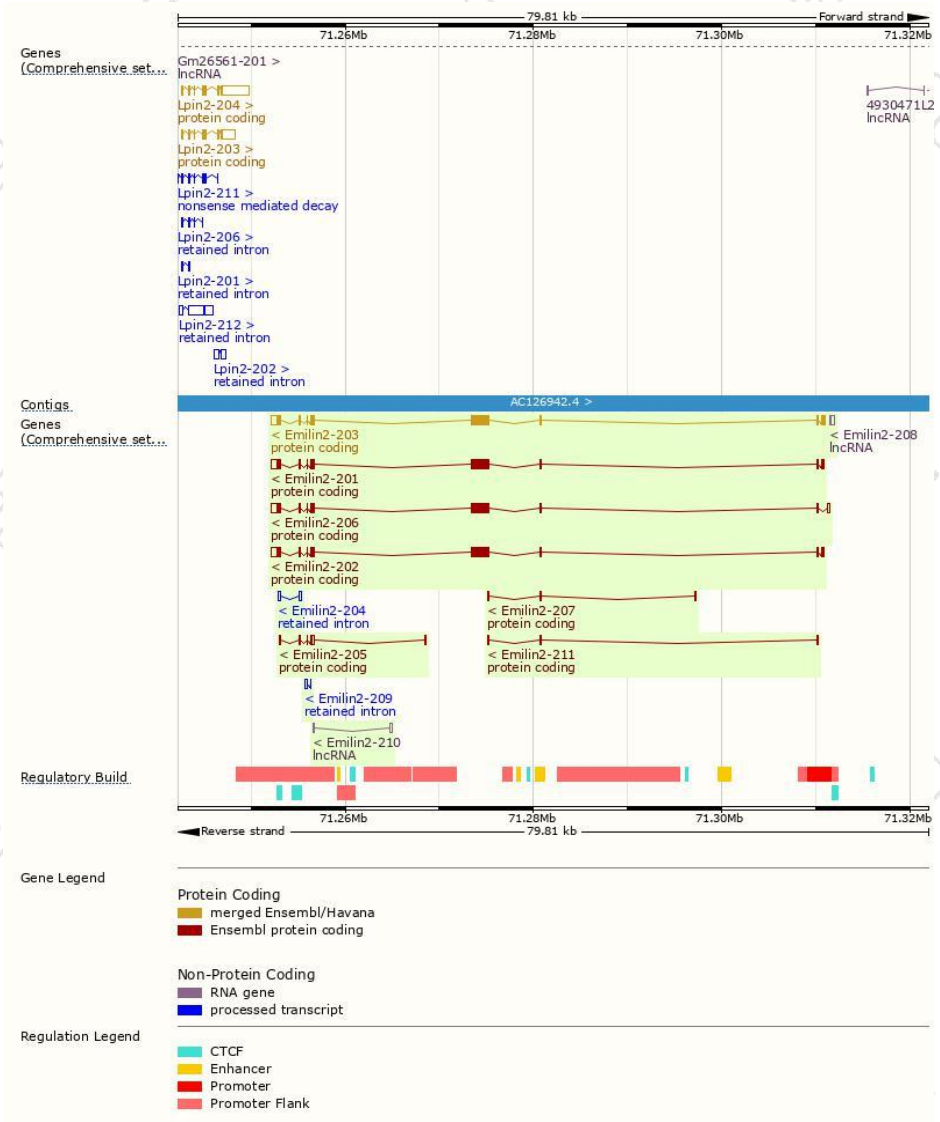
The gene has 11 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Emilin2-203	ENSMUST00000233057.1	3939	1074aa	Protein coding	CCDS37686	Q3U1J9 Q8K482	GENCODE basic APPRIS P2
Emilin2-201	ENSMUST00000024849.10	3906	1073aa	Protein coding	-	Q3TDP9	TSL:1 GENCODE basic APPRIS ALT2
Emilin2-206	ENSMUST00000233245.1	3883	993aa	Protein coding	-	A0A3B2WB50	GENCODE basic
Emilin2-202	ENSMUST00000232777.1	3873	1063aa	Protein coding	-	A0A3B2W4E4	GENCODE basic APPRIS ALT2
Emilin2-205	ENSMUST00000233148.1	758	122aa	Protein coding	-	A0A3B2WD46	CDS 3' incomplete
Emilin2-211	ENSMUST00000233728.1	405	98aa	Protein coding	-	A0A3B2W437	CDS 3' incomplete
Emilin2-207	ENSMUST00000233343.1	404	95aa	Protein coding	-	A0A3B2W846	CDS 3' incomplete
Emilin2-204	ENSMUST00000233083.1	479	No protein	Retained intron	-	-	
Emilin2-209	ENSMUST00000233677.1	363	No protein	Retained intron	-	-	
Emilin2-210	ENSMUST00000233698.1	399	No protein	lncRNA	-	-	
Emilin2-208	ENSMUST00000233659.1	368	No protein	lncRNA	-	-	

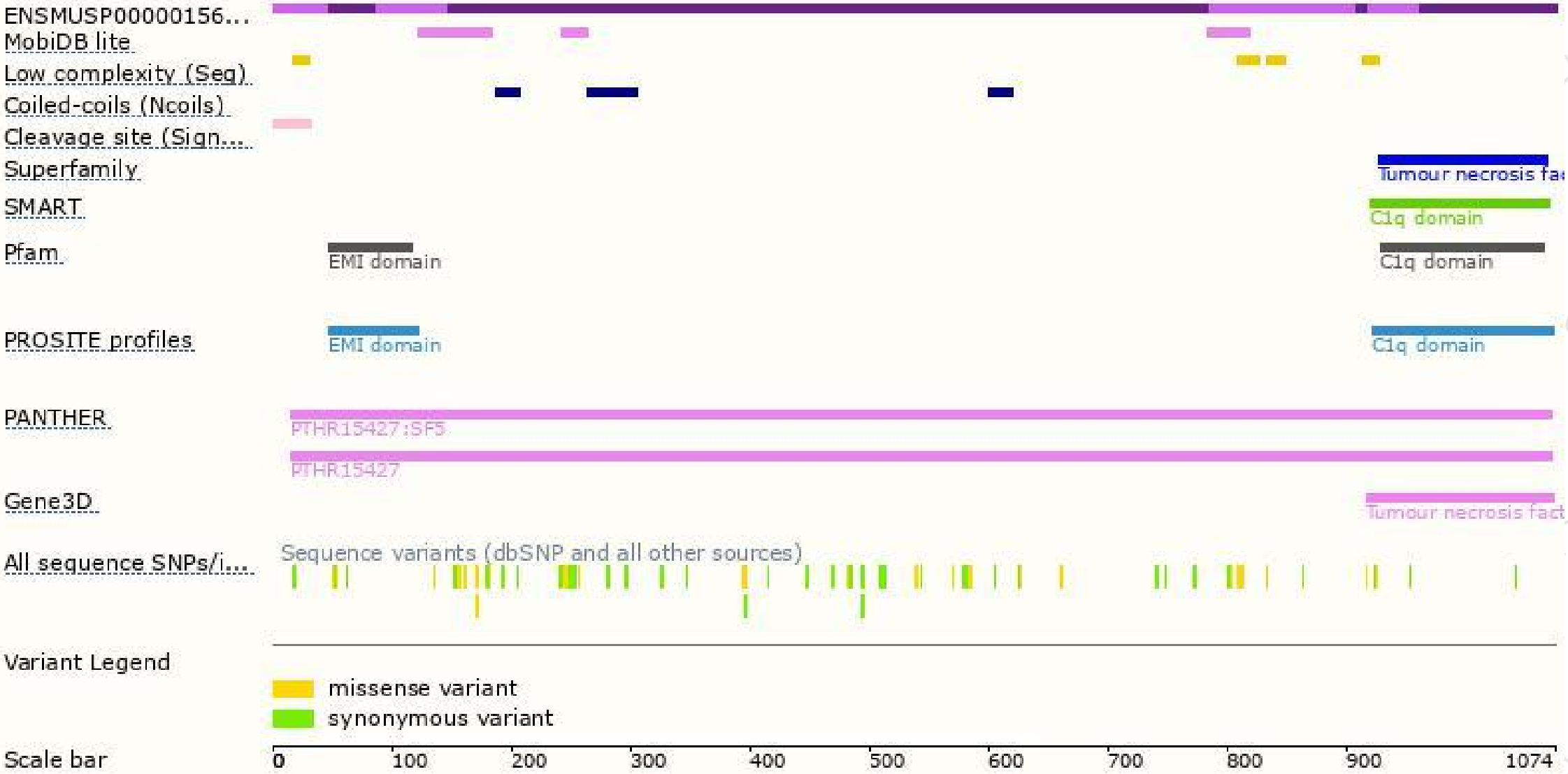
The strategy is based on the design of *Emilin2-203* transcript,The transcription is shown below



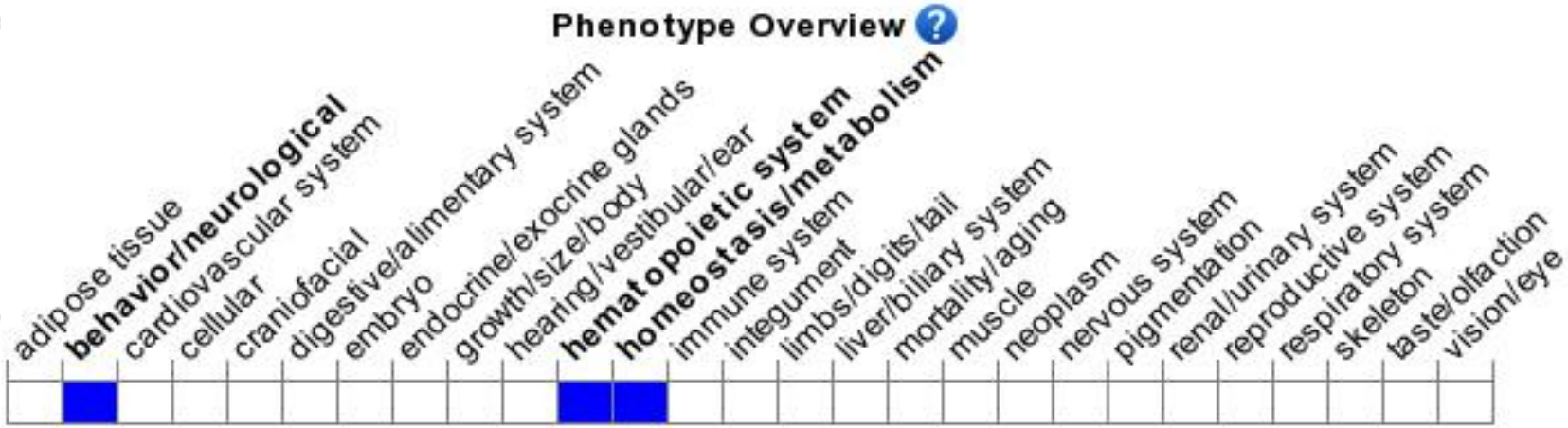
Genomic location distribution



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,Mice homozygous for a knock-out allele exhibit disruptions in platelet activation, thrombus formation and clot retraction.

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

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