

Pdia4 Cas9-CKO Strategy

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Reviewer:

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Design Date:

2020-2-18

Project Overview

Project Name

Pdia4

Project type

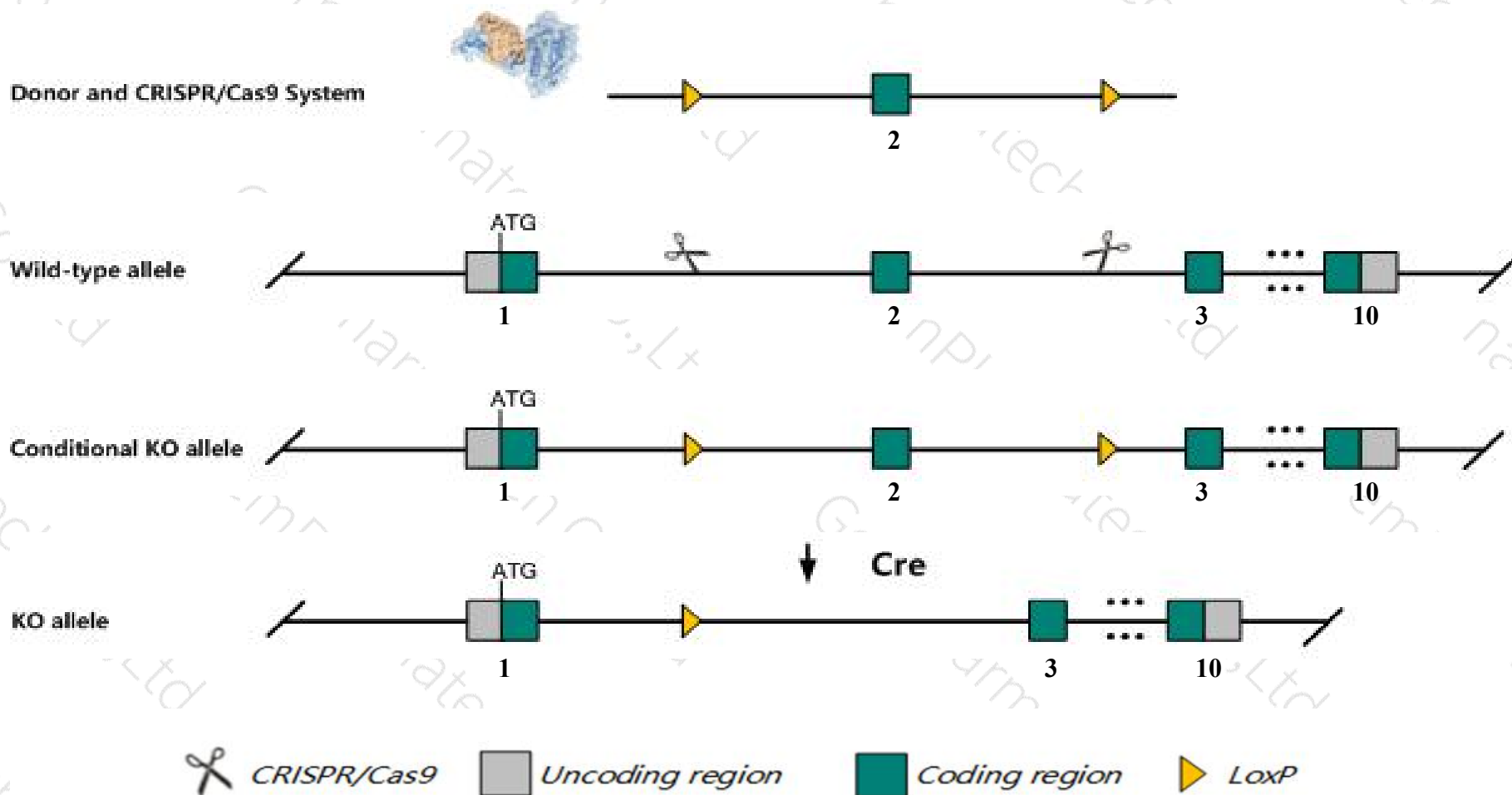
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pdia4* gene. The schematic diagram is as follows:



- The *Pdia4* gene has 2 transcripts. According to the structure of *Pdia4* gene, exon2 of *Pdia4-201* (ENSMUST00000077290.8) transcript is recommended as the knockout region. The region contains 169bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Pdia4* 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.

- According to the existing MGI data, Mice homozygous for a conditional allele activated in platelets exhibit decreased platelet aggregation and increased bleeding time.
- The *Pdia4* gene is located on the Chr6. 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)

Pdia4 protein disulfide isomerase associated 4 [Mus musculus (house mouse)]

Gene ID: 12304, updated on 24-Feb-2019

Summary



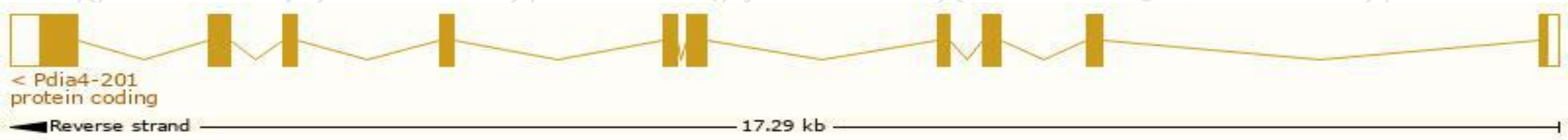
Official Symbol	Pdia4 provided by MGI
Official Full Name	protein disulfide isomerase associated 4 provided by MGI
Primary source	MGI:MGI:104864
See related	Ensembl:ENSMUSG00000025823
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI987846, Cai, ERp-72, Erp72
Expression	Ubiquitous expression in placenta adult (RPKM 75.3), genital fat pad adult (RPKM 46.1) and 25 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

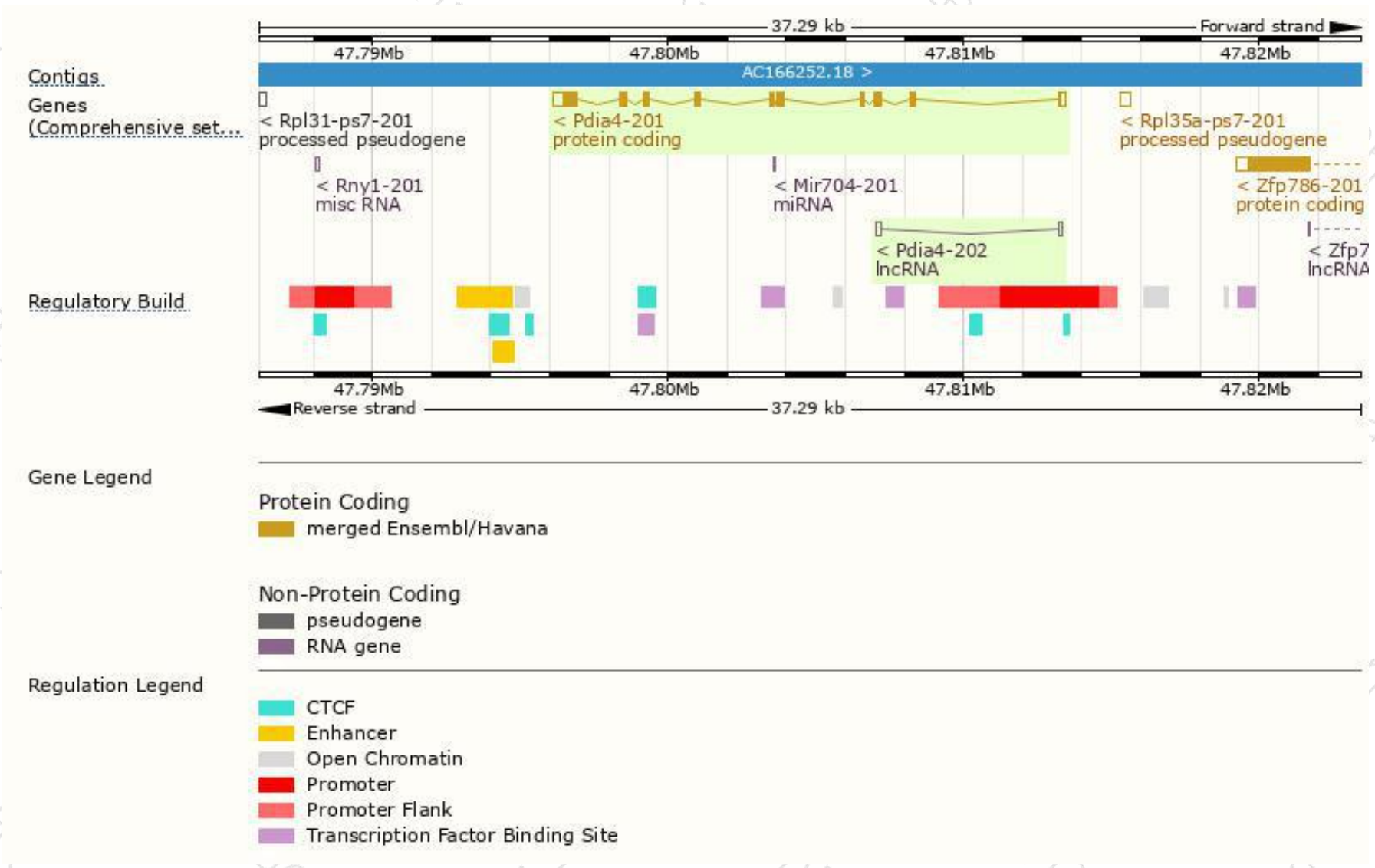
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pdia4-201	ENSMUST00000077290.8	2399	641aa	Protein coding	CCDS20097	A0A0R4J0Z1	TSL:1 GENCODE basic APPRIS P1
Pdia4-202	ENSMUST00000204029.1	347	No protein	lncRNA	-	-	TSL:2

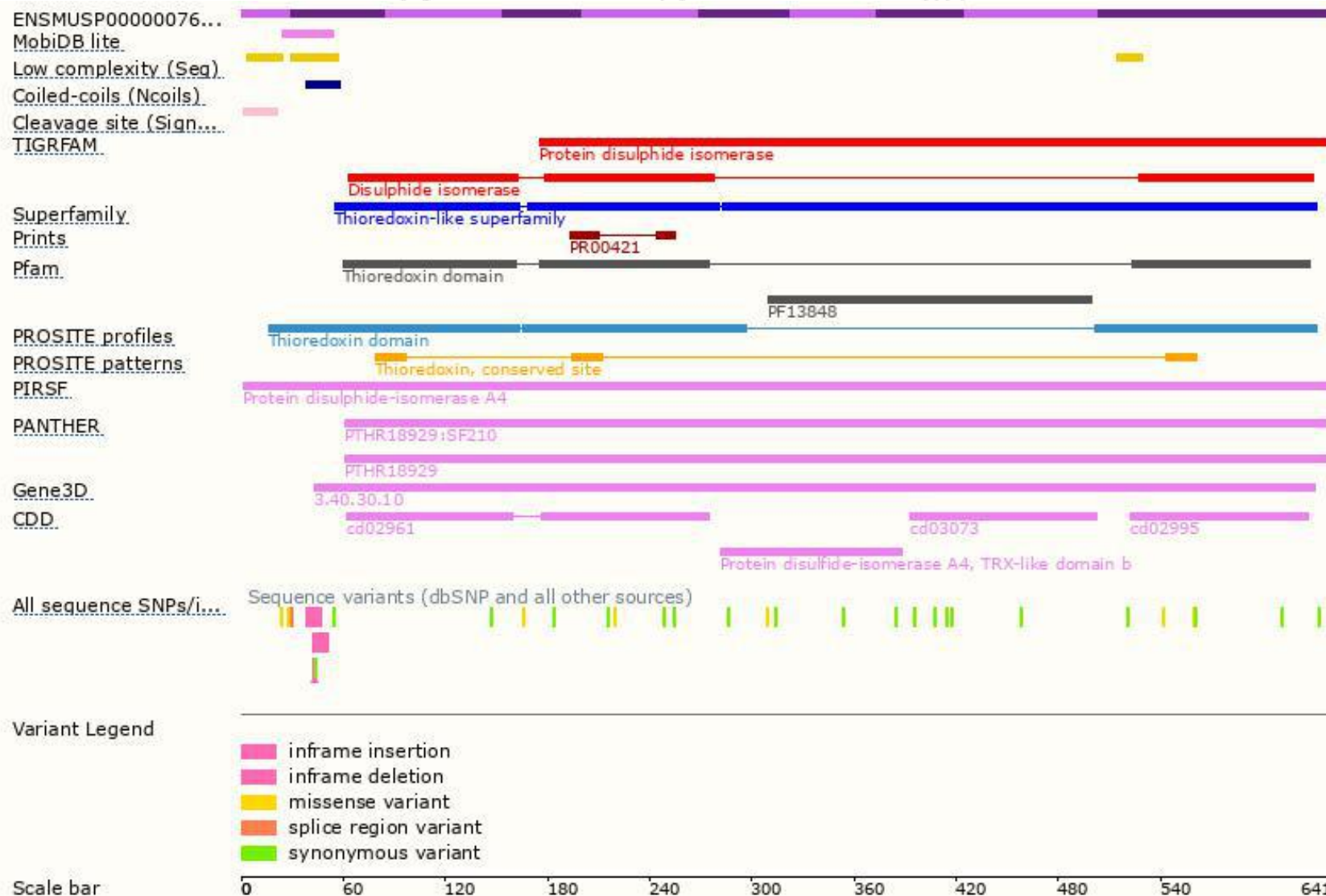
The strategy is based on the design of *Pdia4-201* transcript,The transcription is shown below



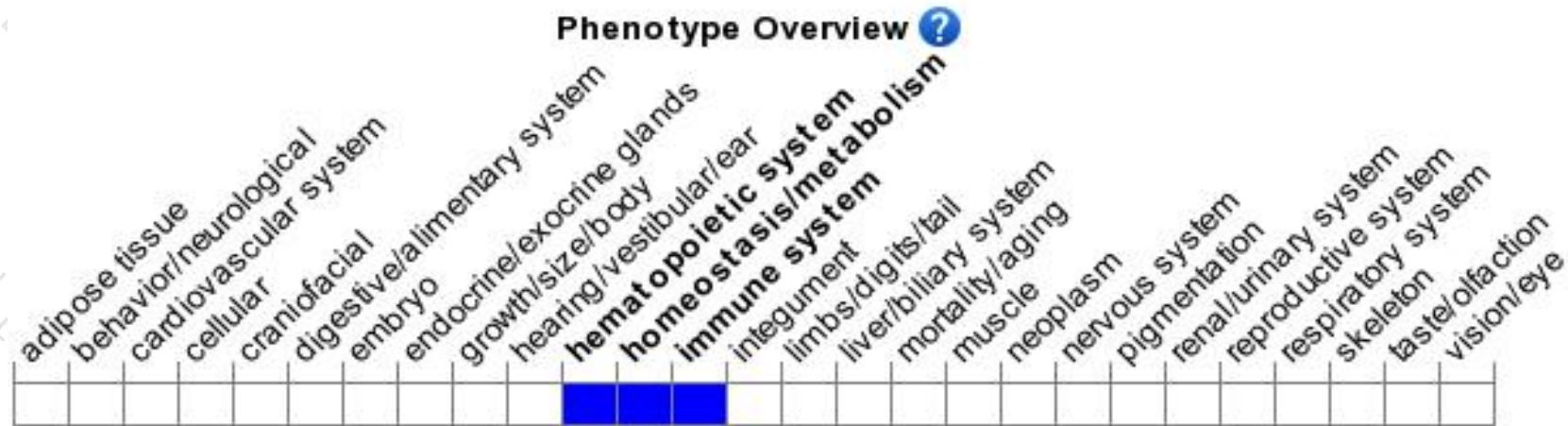
Genomic location distribution



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 conditional allele activated in platelets exhibit decreased platelet aggregation and increased bleeding time.

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

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