Pld2 Cas9-CKO Strategy

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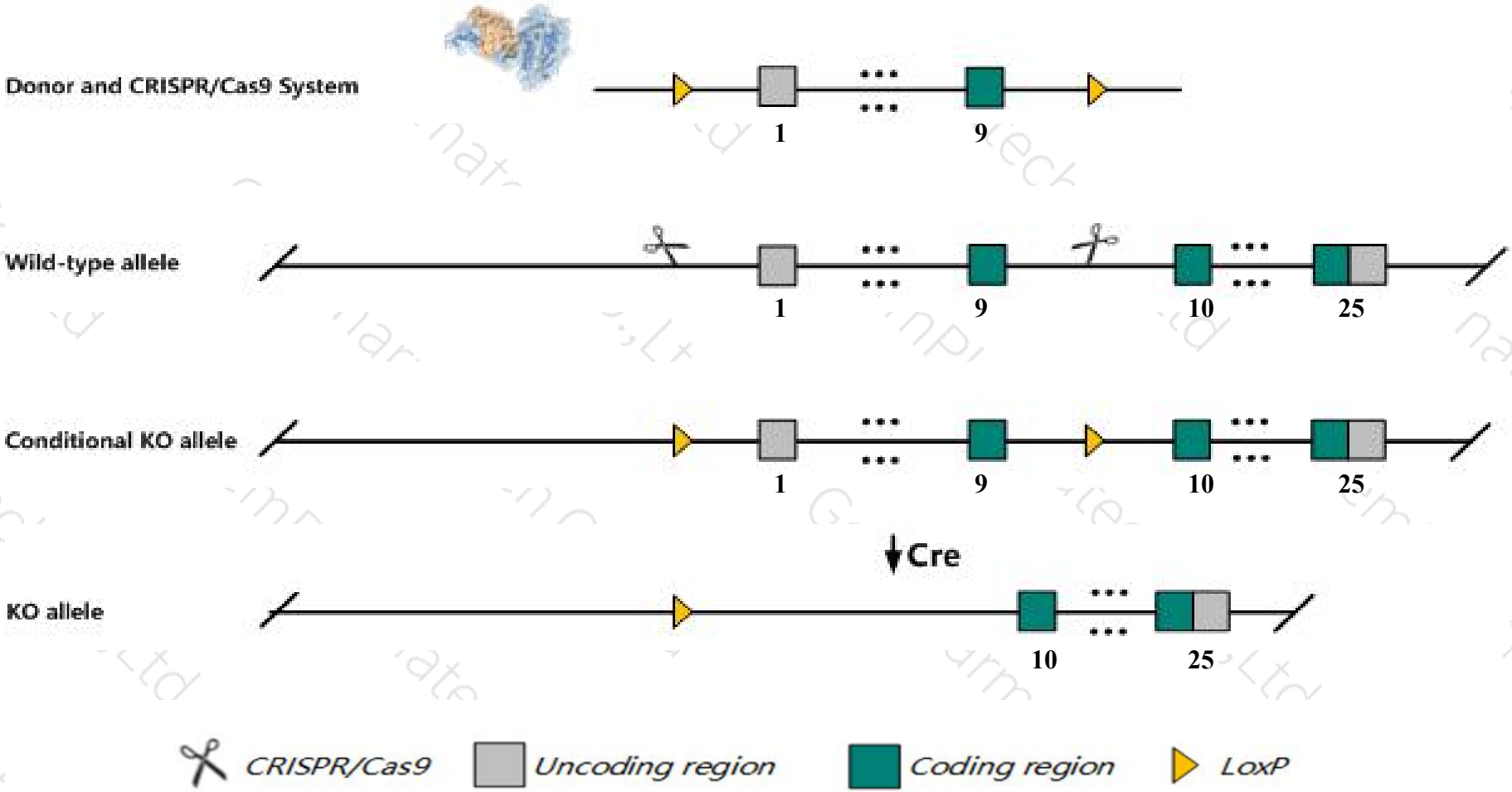
Project Overview

Project Name	***Pld2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pld2* gene. The schematic diagram is as follows:



Technical routes

- The *Pld2* gene has 7 transcripts. According to the structure of *Pld2* gene, exon1-exon9 of *Pld2-201* (ENSMUST00000018429.11) transcript is recommended as the knockout region.The region contains 860bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Pld2* 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 fail to exhibit Abeta42 suppression of LTP and show altered brain phosphatidic acid levels. Mice homozygous for a different knock-out allele show normal platelet function, hemostasis and thrombus formation.
- Transcript *Pld2-204* may not be affected.
- The KO region contains functional region of the *Gm10418* gene.Knockout the region may affect the function of *Gm10418* gene.
- The *Pld2* gene is located on the Chr11. 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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Pld2 phospholipase D2 [Mus musculus (house mouse)]

Gene ID: 18806, updated on 5-Mar-2019

Summary

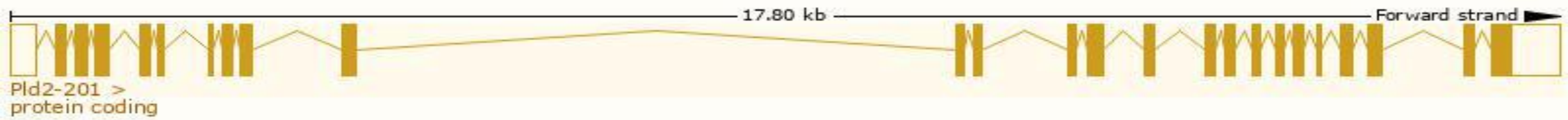
Official Symbol	Pld2 provided by MGI
Official Full Name	phospholipase D2 provided by MGI
Primary source	MGI:MGI:892877
See related	Ensembl:ENSMUSG00000020828
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	PLD1C
Summary	This gene is a member of the phospholipase D (PLD) superfamily. The encoded protein catalyzes the hydrolysis of phosphatidylcholine to phosphatidic acid and choline. Phosphatidic acid is an essential intracellular lipid second messenger for many signaling pathways and has been implicated in a variety of physiological processes including cytoskeletal organization and cell proliferation. A similar gene in human may also function as a guanine nucleotide exchange factor (GEF) for the small GTPase Rac2. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Oct 2014]
Expression	Ubiquitous expression in lung adult (RPKM 7.1), subcutaneous fat pad adult (RPKM 6.6) and 27 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

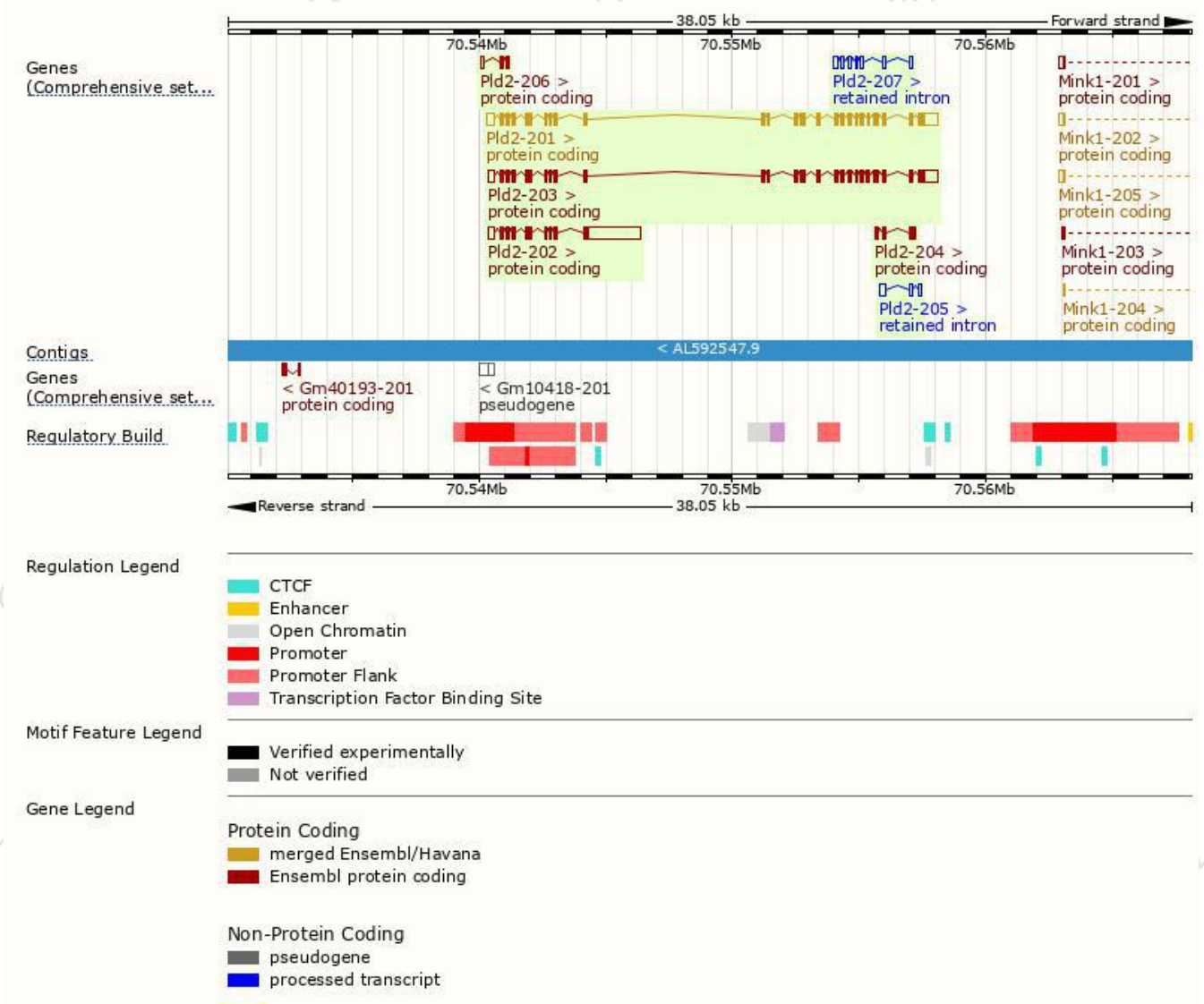
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pld2-201	ENSMUST00000018429.11	3659	933aa	Protein coding	CCDS24953	P97813 Q5SXG5	TSL:1 GENCODE basic APPRIS P3
Pld2-203	ENSMUST00000108557.9	3655	944aa	Protein coding	CCDS78979	Q6NV49	TSL:1 GENCODE basic APPRIS ALT2
Pld2-202	ENSMUST00000108556.1	3391	337aa	Protein coding	CCDS78980	Q80ZW1	TSL:1 GENCODE basic
Pld2-204	ENSMUST00000130678.1	467	132aa	Protein coding	-	J3QN26	CDS 5' incomplete TSL:5
Pld2-206	ENSMUST00000157075.7	376	76aa	Protein coding	-	J3QK47	CDS 3' incomplete TSL:2
Pld2-207	ENSMUST00000179806.1	875	No protein	Retained intron	-	-	TSL:3
Pld2-205	ENSMUST00000146248.1	513	No protein	Retained intron	-	-	TSL:2

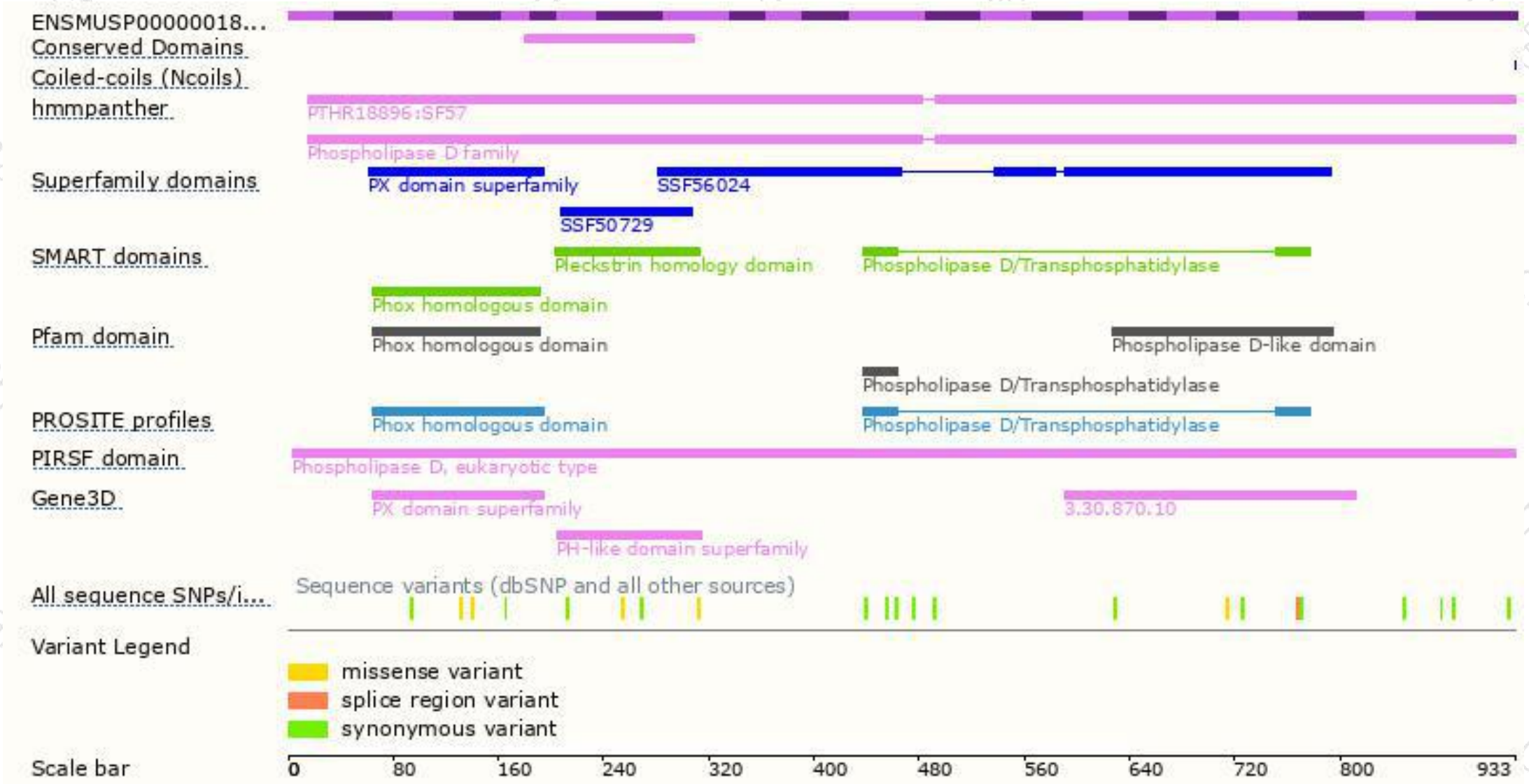
The strategy is based on the design of *Pld2-201* transcript,The transcription is shown below



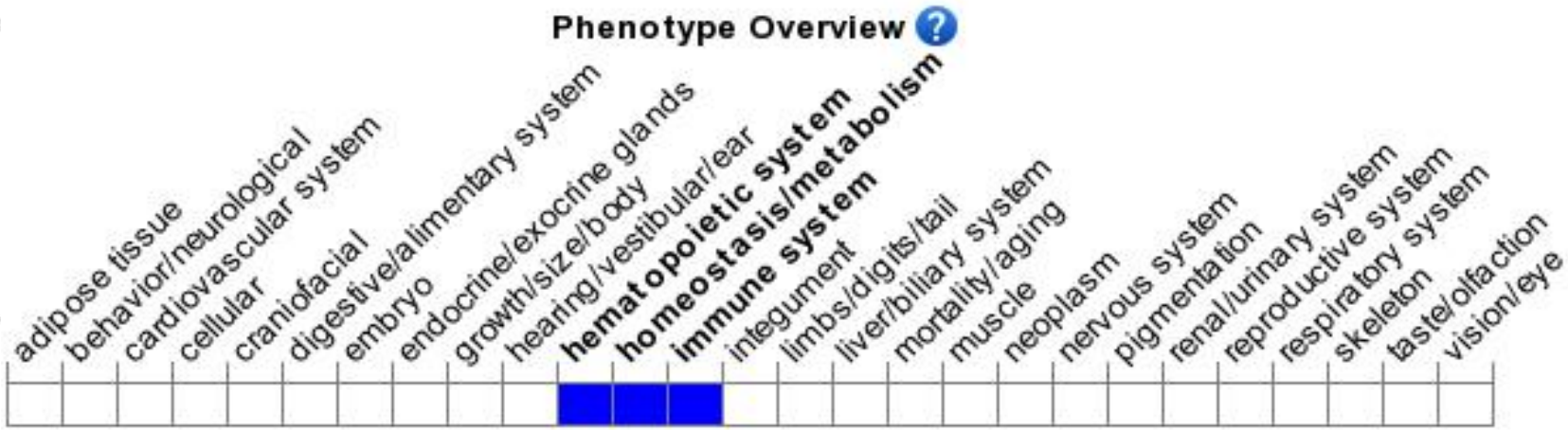
Genomic location distribution



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/).

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

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