

Trpv5 Cas9-CKO Strategy

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Reviewer:

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Design Date:

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Project Overview

Project Name

Trpv5

Project type

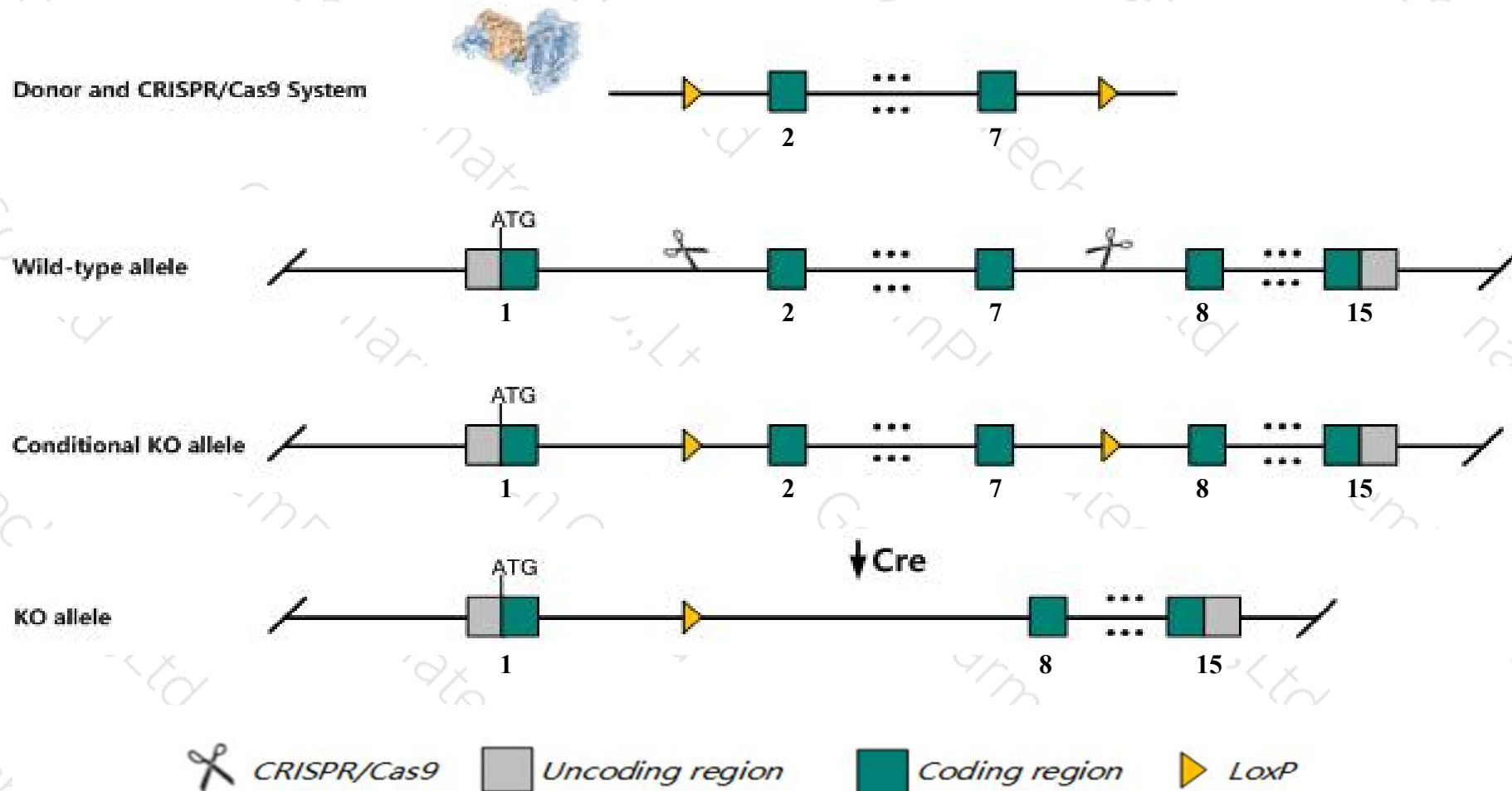
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Trpv5* gene. The schematic diagram is as follows:



Technical routes

- The *Trpv5* gene has 2 transcripts. According to the structure of *Trpv5* gene, exon2-exon7 of *Trpv5-201* (ENSMUST00000031901.10) transcript is recommended as the knockout region. The region contains 778bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Trpv5* 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, Homozygous mutant mice exhibit increased calcium excretion and reduced bone thickenss.
- The *Trpv5* 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.

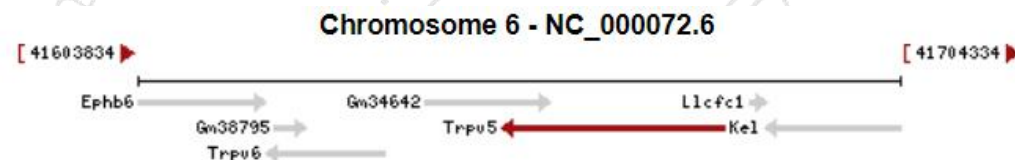
Gene information (NCBI)

Trpv5 transient receptor potential cation channel, subfamily V, member 5 [*Mus musculus* (house mouse)]

Gene ID: 194352, updated on 25-Nov-2019

Summary

Official Symbol	Trpv5 provided by MGI
Official Full Name	transient receptor potential cation channel, subfamily V, member 5 provided by MGI
Primary source	MGI:MGI:2429764
See related	Ensembl:ENSMUSG000000036899
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	CAT2; ECAC1; D630033B11
Expression	Biased expression in kidney adult (RPKM 1.9), placenta adult (RPKM 0.2) and 2 other tissues See more
Orthologs	human all

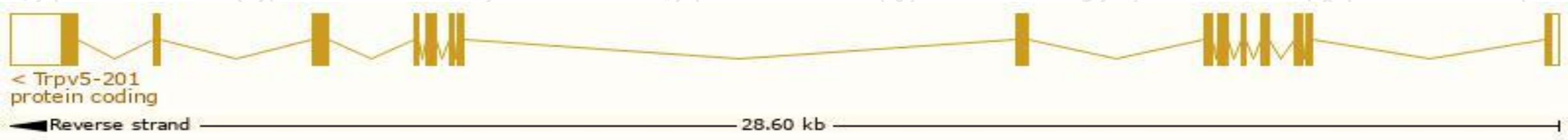


Transcript information (Ensembl)

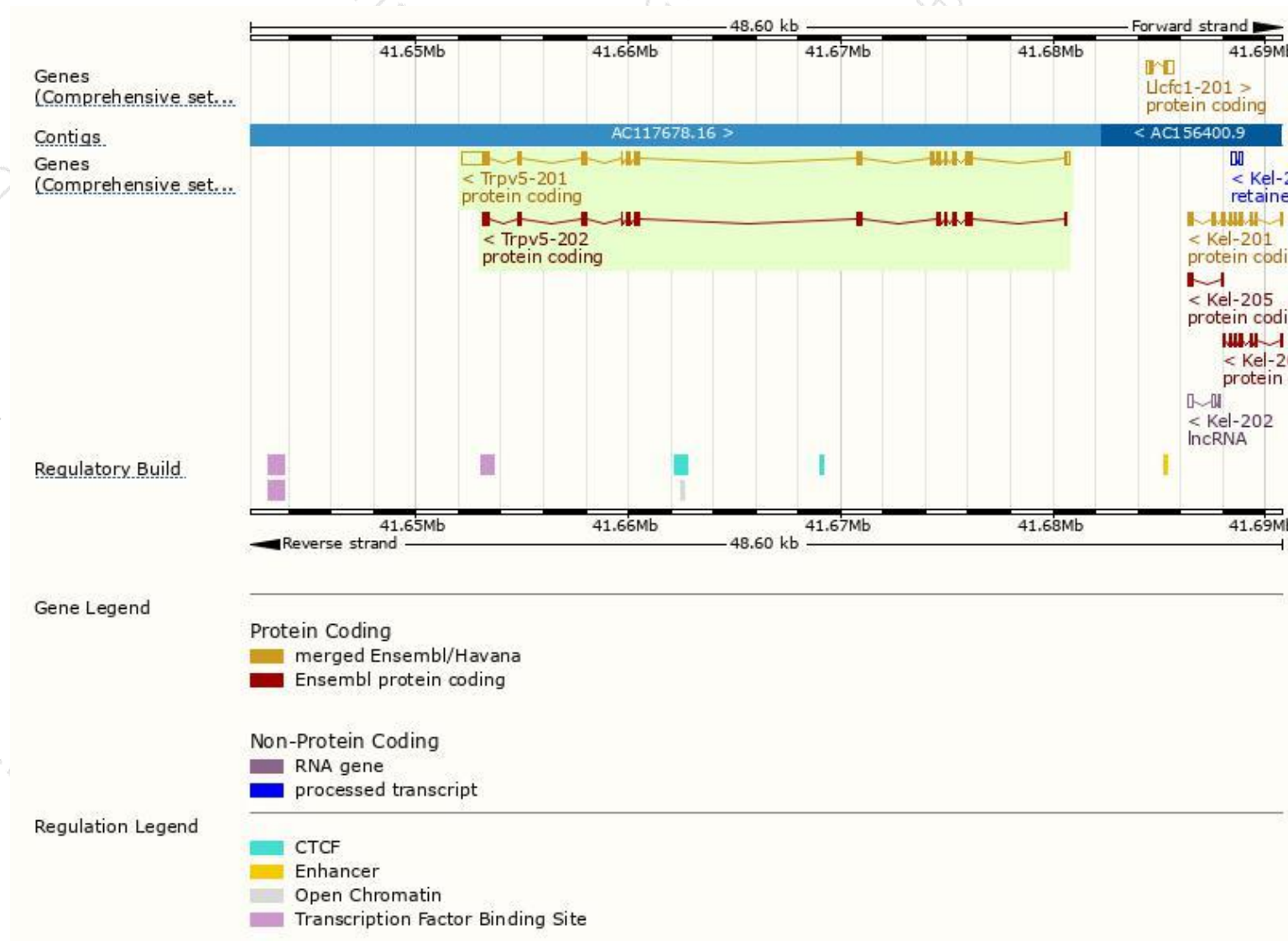
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Trpv5-201	ENSMUST00000031901.10	3243	723aa	Protein coding	CCDS20053	P69744	TSL:1 GENCODE basic APPRIS P2
Trpv5-202	ENSMUST00000193503.1	2025	674aa	Protein coding	-	A0A0A6YW69	TSL:5 GENCODE basic APPRIS ALT2

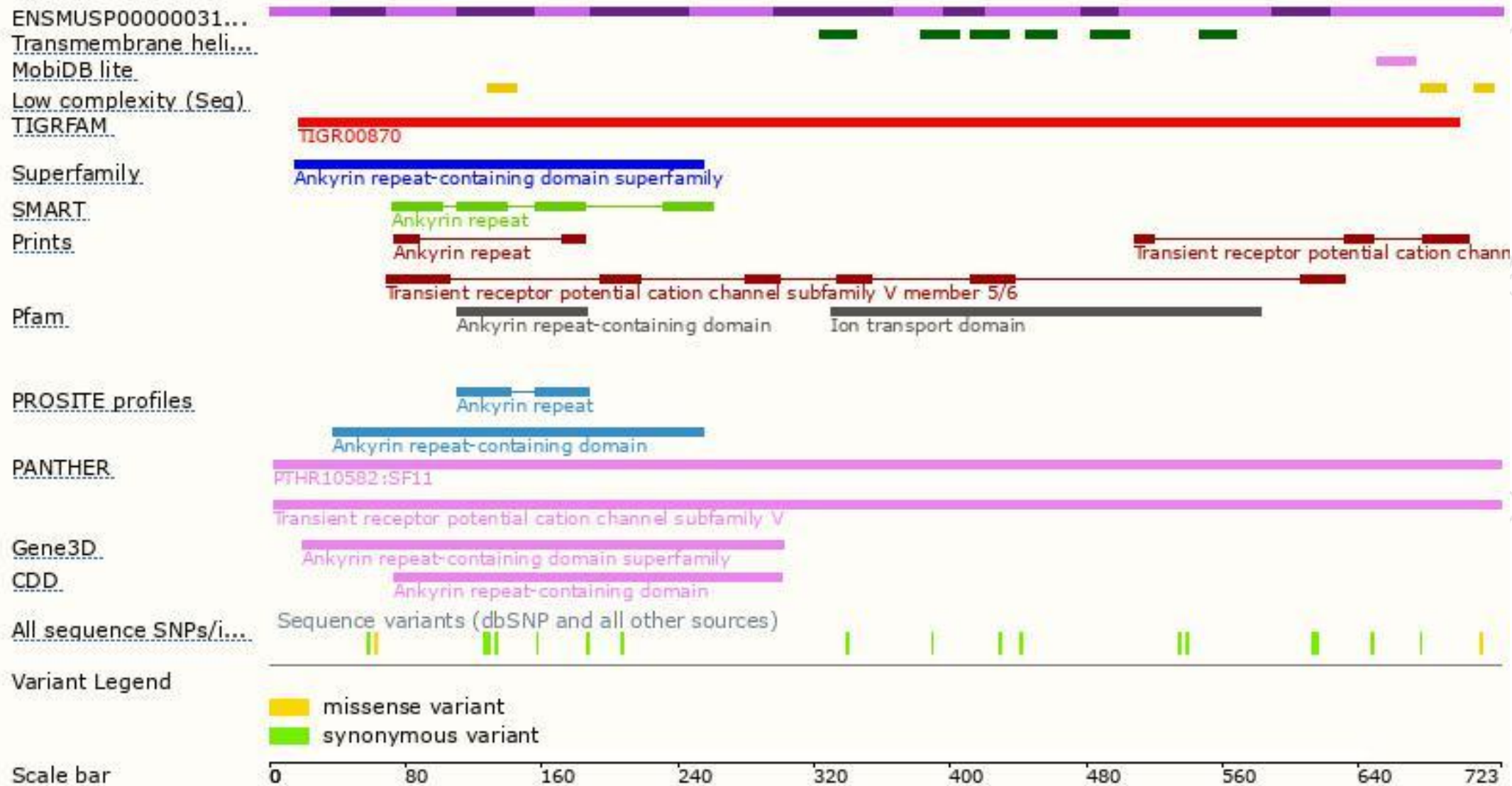
The strategy is based on the design of *Trpv5-201* transcript,The transcription is shown below



Genomic location distribution

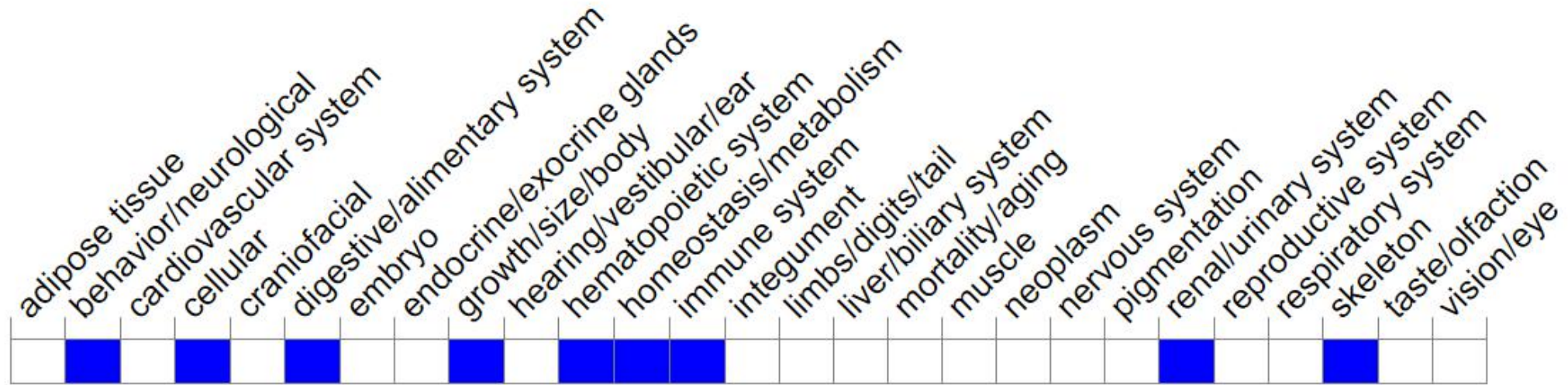


Protein domain



Mouse phenotype description(MGI)

Phenotype Overview ?



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, Homozygous mutant mice exhibit increased calcium excretion and reduced bone thickenss.

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

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