

Calcr Cas9-CKO Strategy

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Project Overview

Project Name

Calcr

Project type

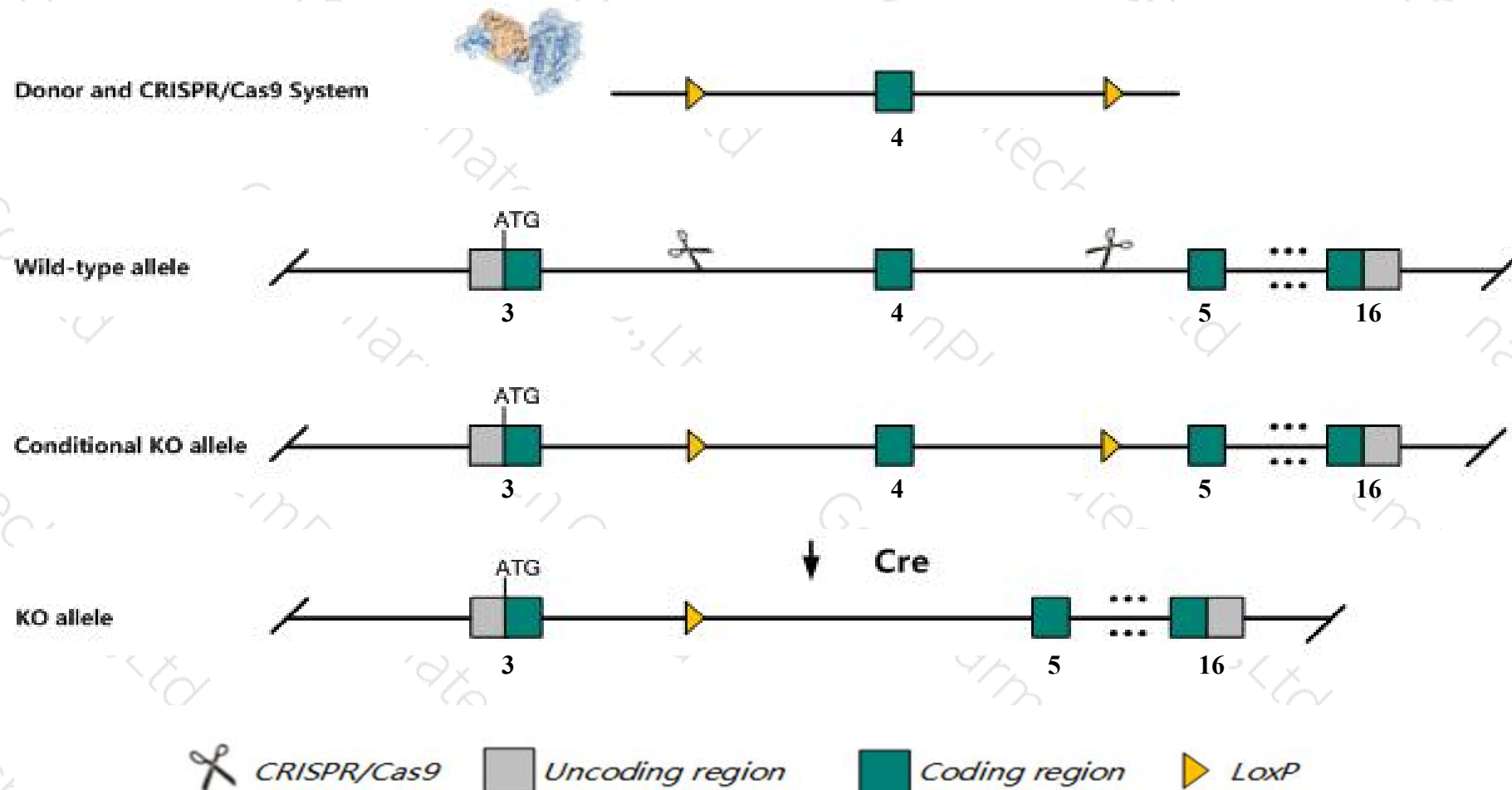
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Calcr* gene has 5 transcripts. According to the structure of *Calcr* gene, exon4 of *Calcr-201* (ENSMUST00000075644.12) transcript is recommended as the knockout region. The region contains 79bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Calcr* 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, Haploinsufficiency may result in increased bone density due to increased bone formation. Homozygous inactivation may result in embryonic lethality. Mice homozygous for another disruption allele at this locus show a normal phenotype.
- The floxed region is near to the N-terminal of *Mir653* and *Mir489* gene, this strategy may influence the regulatory function of the N-terminal of these gene.
- The *Calcr* 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)

Calcr calcitonin receptor [Mus musculus (house mouse)]

Gene ID: 12311, updated on 19-Mar-2019

Summary



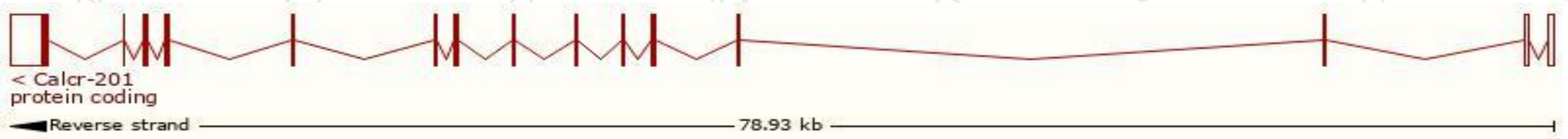
Official Symbol	Calcr provided by MGI
Official Full Name	calcitonin receptor provided by MGI
Primary source	MGI:MGI:101950
See related	Ensembl:ENSMUSG00000023964
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Clr, Ct-r
Expression	Biased expression in whole brain E14.5 (RPKM 1.5), CNS E18 (RPKM 1.2) and 5 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

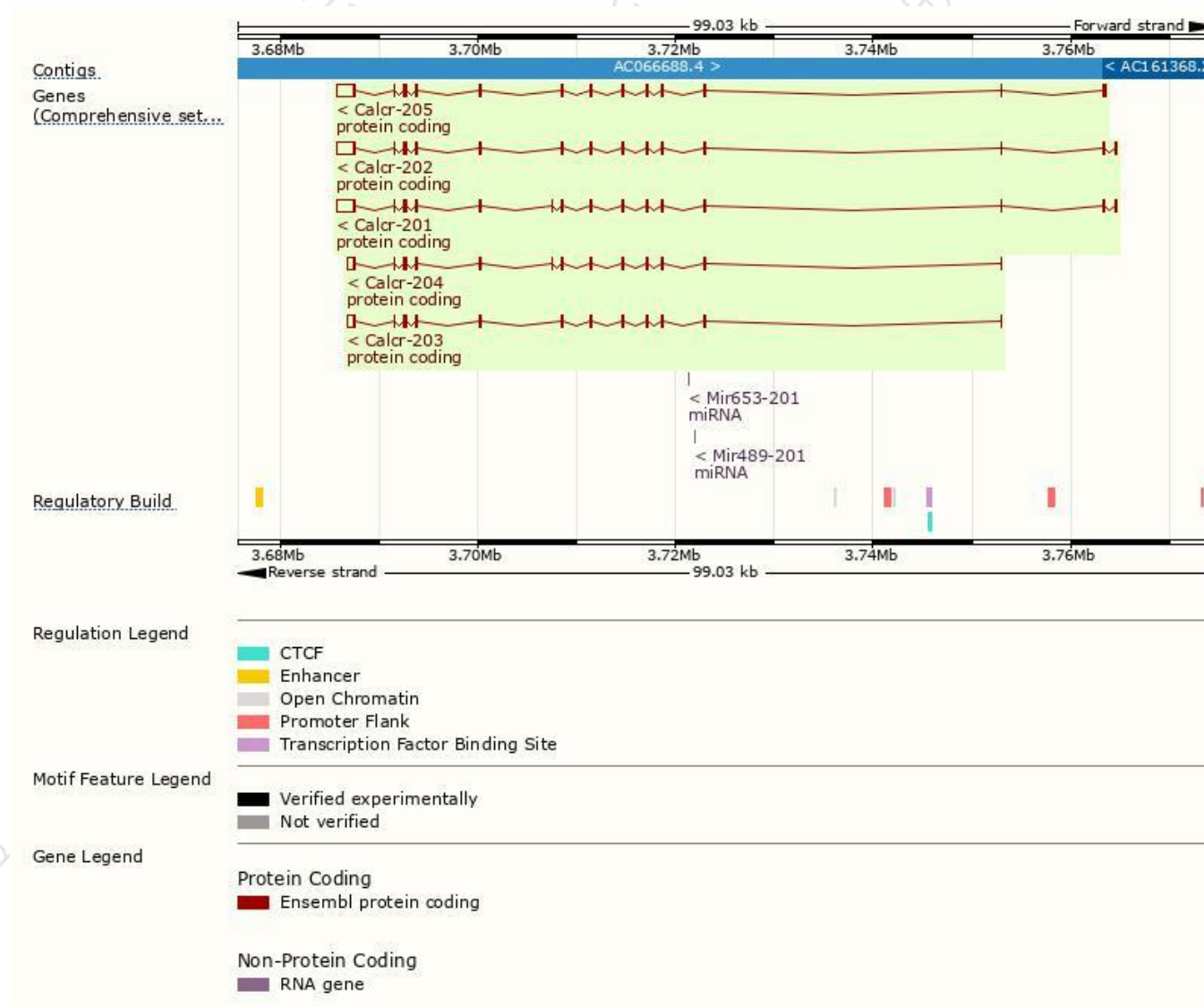
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Calcr-201	ENSMUST00000075644.12	3763	533aa	Protein coding	CCDS39417	Q60755	TSL:1 GENCODE basic APPRIS P4
Calcr-202	ENSMUST00000115622.7	3721	496aa	Protein coding	CCDS39416	Q60755	TSL:1 GENCODE basic APPRIS ALT2
Calcr-205	ENSMUST00000171613.7	3518	496aa	Protein coding	CCDS39416	Q60755	TSL:1 GENCODE basic APPRIS ALT2
Calcr-204	ENSMUST00000170266.2	2175	533aa	Protein coding	CCDS39417	Q60755	TSL:1 GENCODE basic APPRIS P4
Calcr-203	ENSMUST00000168592.8	2064	496aa	Protein coding	CCDS39416	Q60755	TSL:5 GENCODE basic APPRIS ALT2

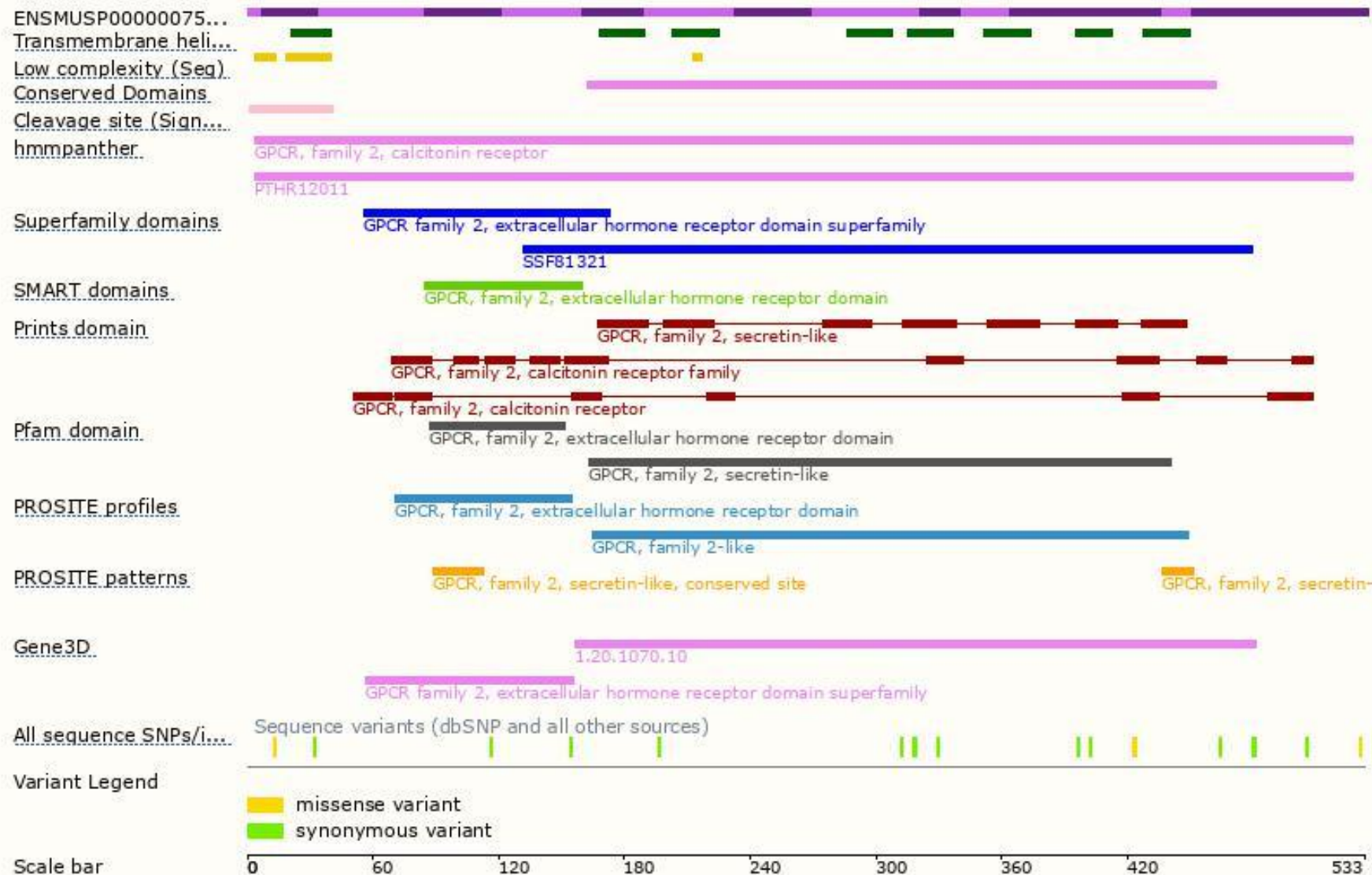
The strategy is based on the design of *Calcr-201* transcript,The transcription is shown below



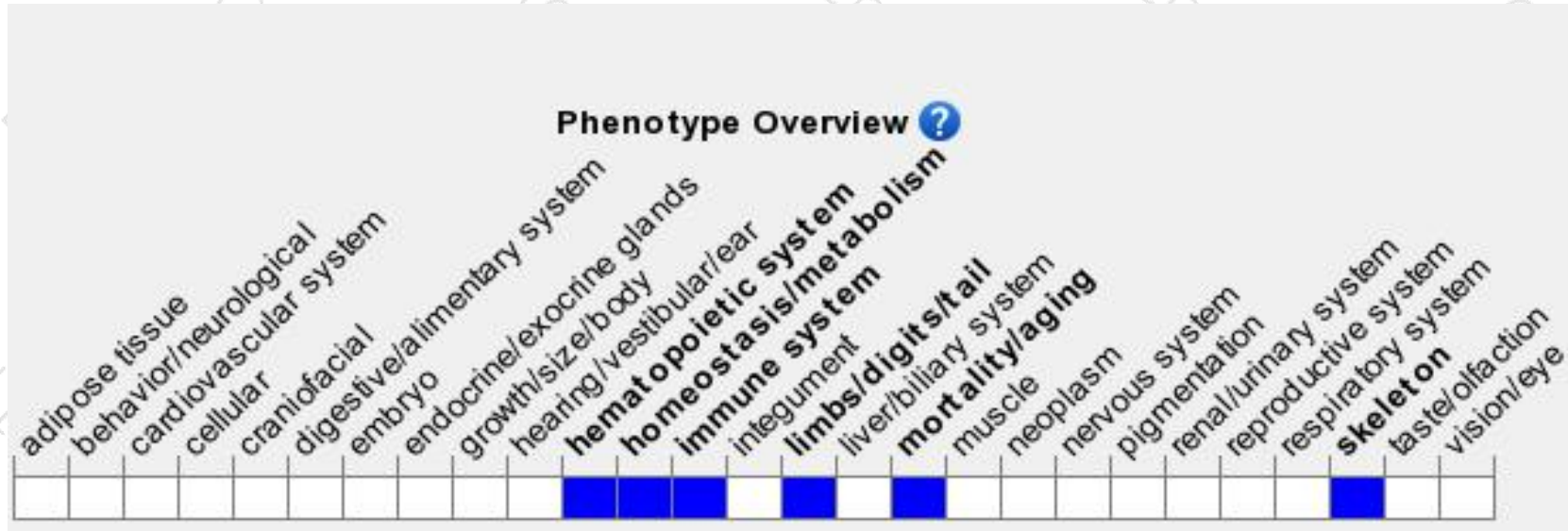
Genomic location distribution



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, Haploinsufficiency may result in increased bone density due to increased bone formation. Homozygous inactivation may result in embryonic lethality. Mice homozygous for another disruption allele at this locus show a normal phenotype.

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

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