

Htra1 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Htra1

Project type

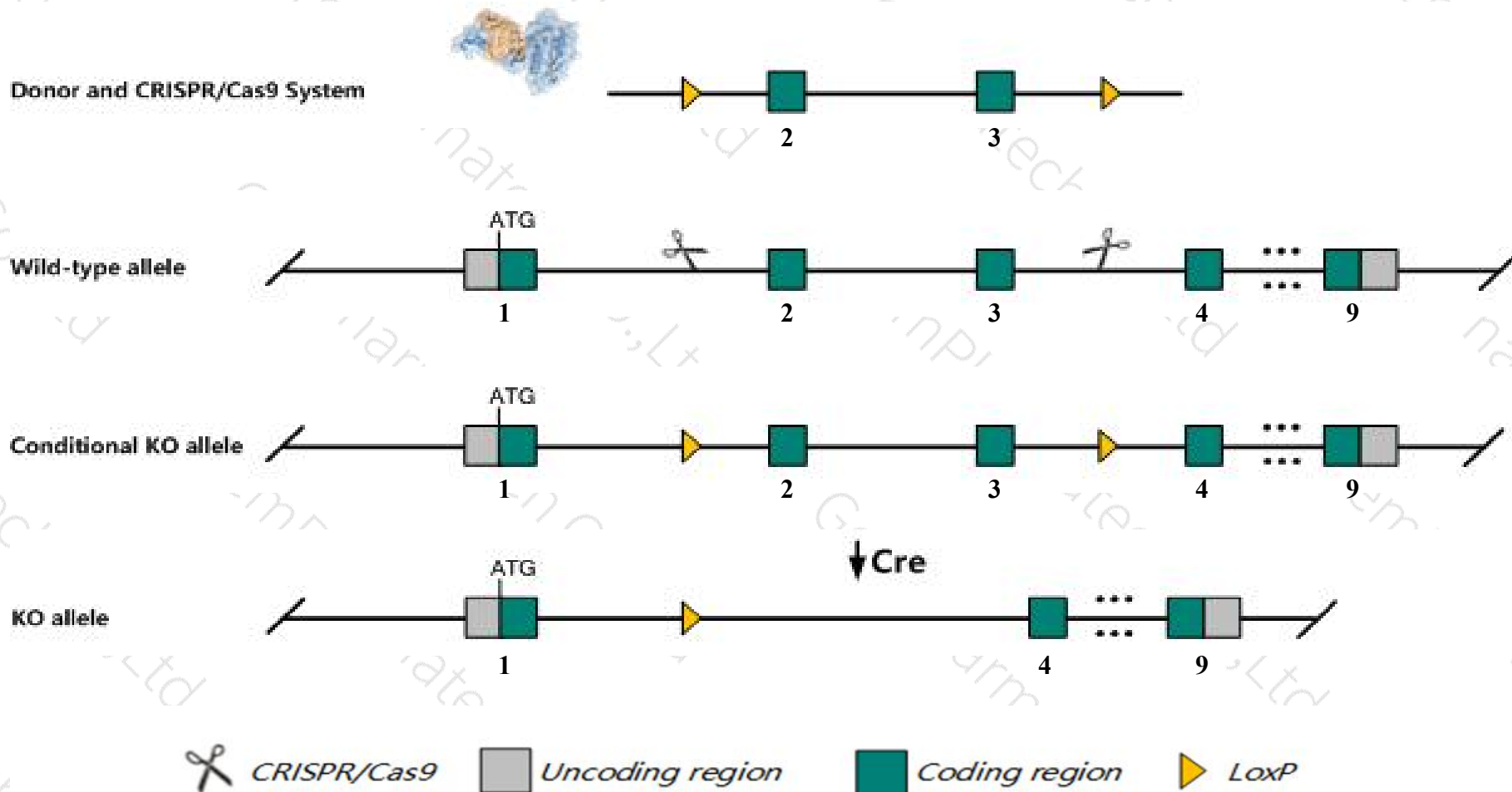
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



- The *Htra1* gene has 5 transcripts. According to the structure of *Htra1* gene, exon2-exon3 of *Htra1-201* (ENSMUST00000006367.7) transcript is recommended as the knockout region. The region contains 305bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Htra1* 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 knock-out allele exhibit normal retinal morphology. Mice homozygous for a different allele exhibit increased bone volume and increased trabecular bone thickness without body weight gain.
- The *Htral* gene is located on the Chr7. 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)

Htra1 HtrA serine peptidase 1 [Mus musculus (house mouse)]

Gene ID: 56213, updated on 24-Feb-2019

Summary



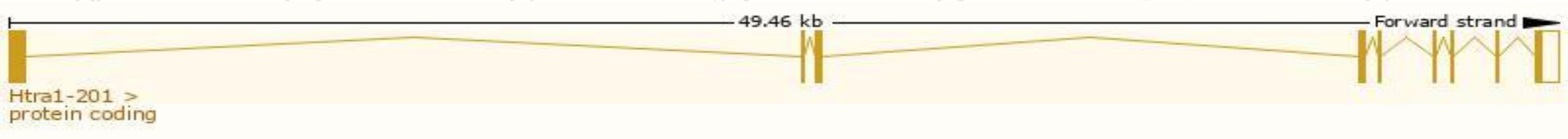
Official Symbol	Htra1 provided by MGI
Official Full Name	HtrA serine peptidase 1 provided by MGI
Primary source	MGI:MGI:1929076
See related	Ensembl:ENSMUSG00000006205
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI429470, HTRA, L56, Prss11, RSPP11
Expression	Biased expression in ovary adult (RPKM 297.0), mammary gland adult (RPKM 75.1) and 14 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

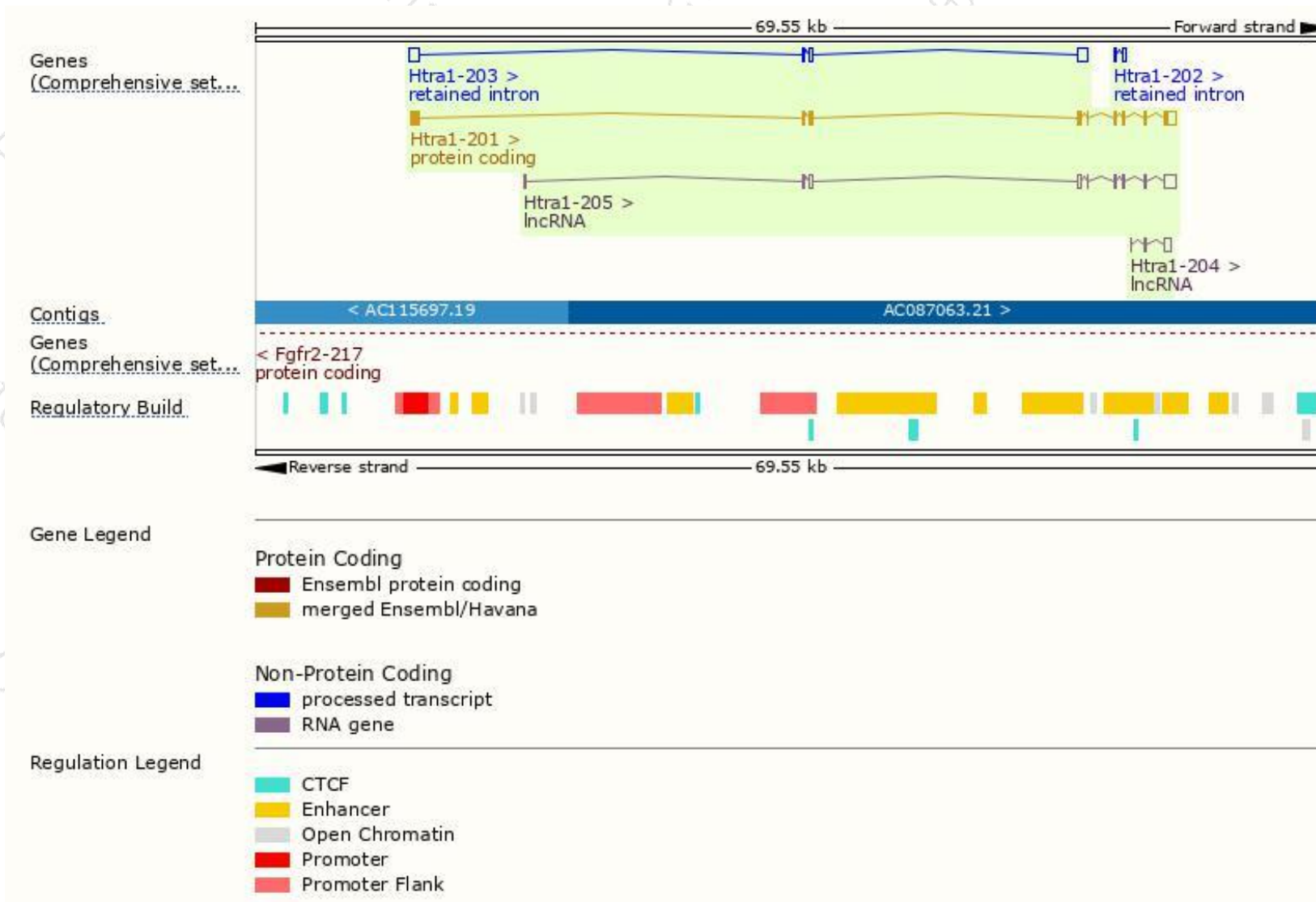
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Htra1-201	ENSMUST00000006367.7	2041	480aa	Protein coding	CCDS21908	Q9R118	TSL:1 GENCODE basic APPRIS P1
Htra1-205	ENSMUST00000153290.7	1606	No protein	Processed transcript	-	-	TSL:1
Htra1-204	ENSMUST00000150905.1	582	No protein	Processed transcript	-	-	TSL:3
Htra1-203	ENSMUST00000150717.7	1572	No protein	Retained intron	-	-	TSL:5
Htra1-202	ENSMUST00000140741.1	309	No protein	Retained intron	-	-	TSL:3

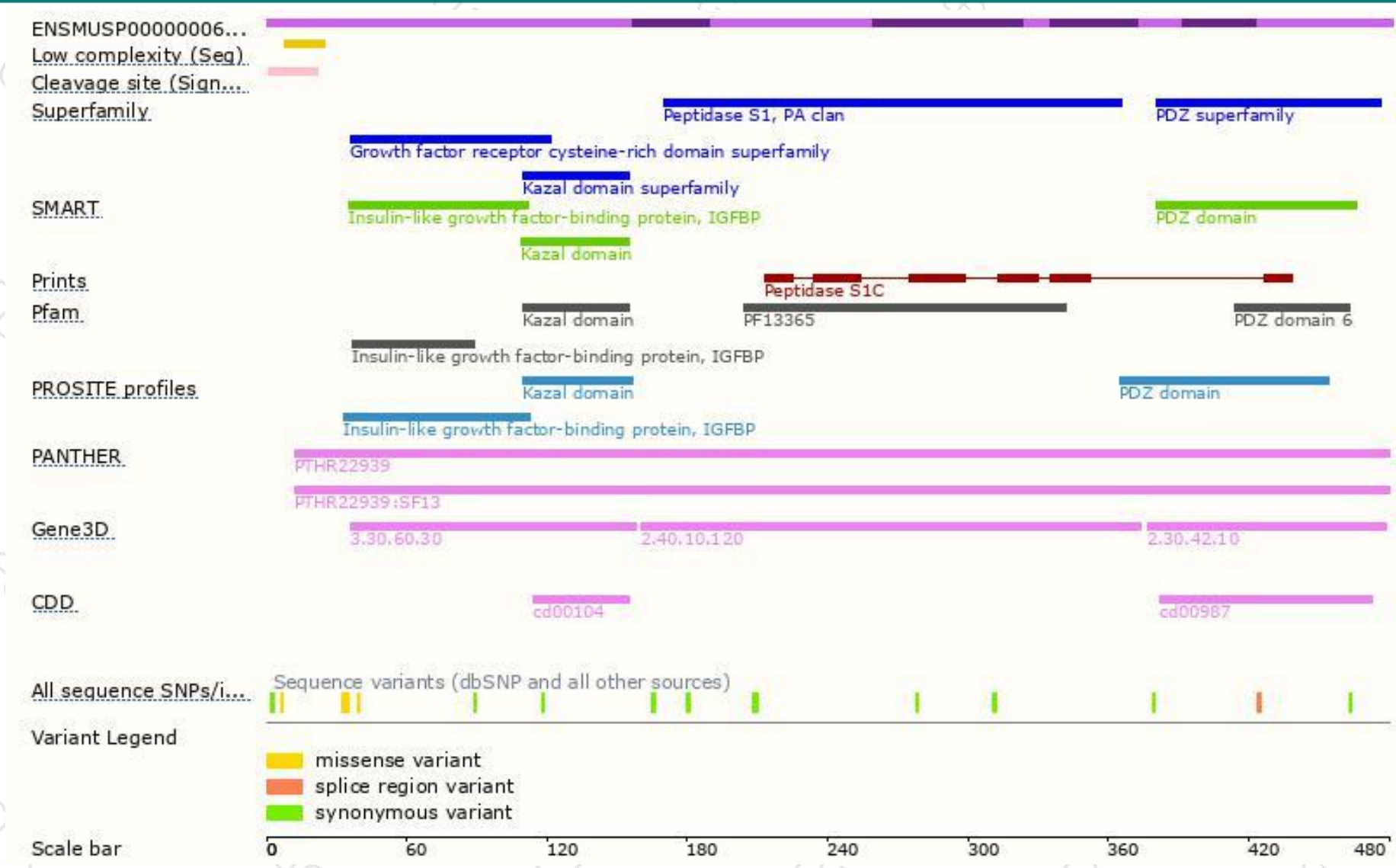
The strategy is based on the design of *Htra1-201* transcript,The transcription is shown below



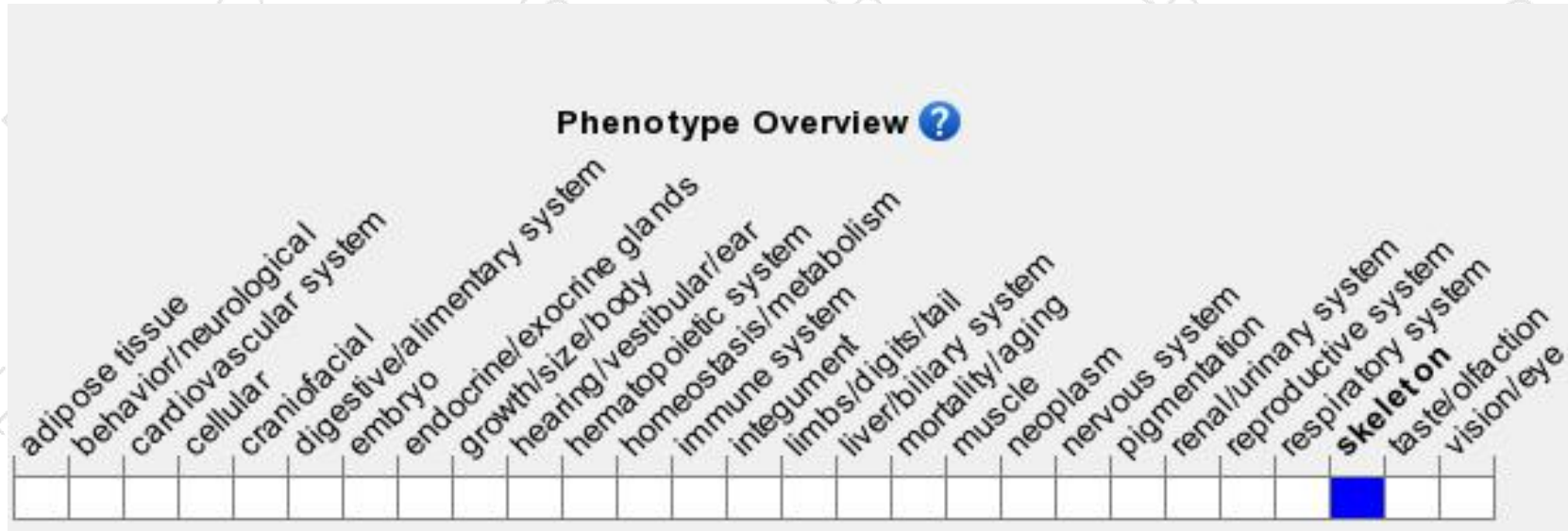
Genomic location distribution



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 knock-out allele exhibit normal retinal morphology. Mice homozygous for a different allele exhibit increased bone volume and increased trabecular bone thickness without body weight gain.

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

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