Pth2r Cas9-CKO Strategy

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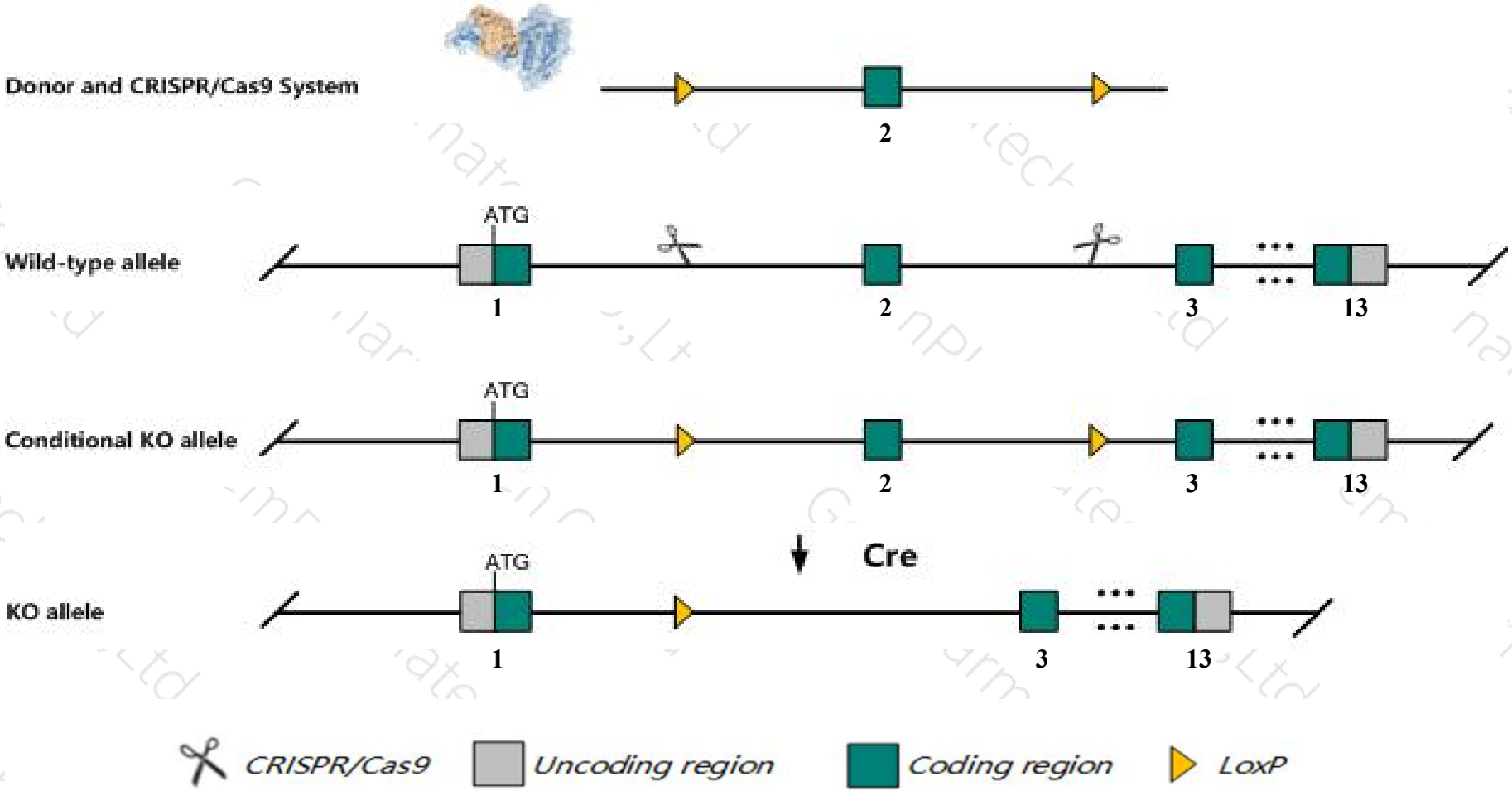
Project Overview

Project Name	***Pth2r***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pth2r* gene. The schematic diagram is as follows:



Technical routes

- The *Pth2r* gene has 2 transcripts. According to the structure of *Pth2r* gene, exon2 of *Pth2r-201* (ENSMUST00000027083.6) transcript is recommended as the knockout region.The region contains 103bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Pth2r* 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, homozygous KO reduces core body temperature in pregnant and lactating females and affects locomotor and grooming behavior of lactating females.

- The *Pth2r* gene is located on the Chr1. 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）

集萃药康 GemPharmatech

Pth2r parathyroid hormone 2 receptor [*Mus musculus* (house mouse)]

Gene ID: 213527, updated on 12-Aug-2019

Summary

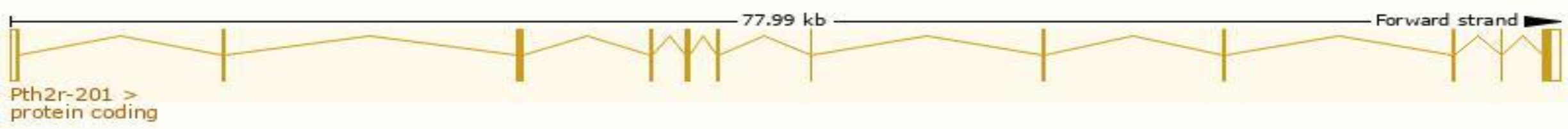
Official Symbol	Pth2r provided by MGI
Official Full Name	parathyroid hormone 2 receptor provided by MGI
Primary source	MGI:MGI:2180917
See related	Ensembl:ENSMUSG00000025946
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Pthr2
Expression	Low expression observed in reference dataset See more
Orthologs	human all

Transcript information（Ensembl）

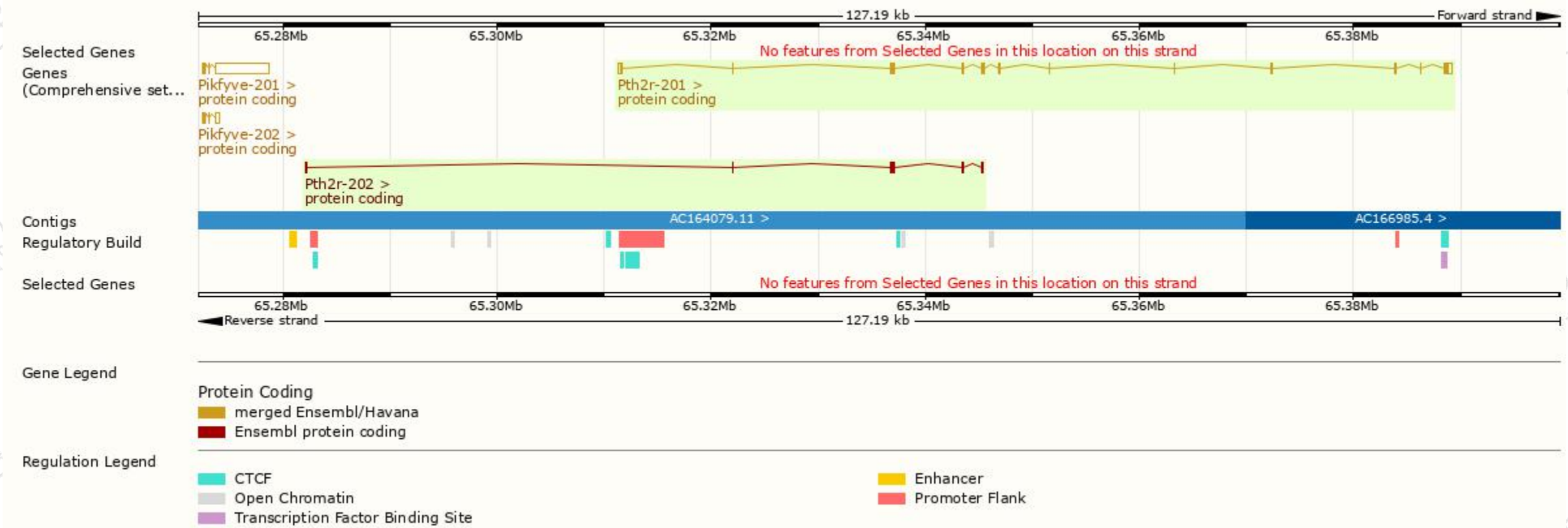
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pth2r-201	ENSMUST00000027083.6	2414	546aa	Protein coding	CCDS15017	Q91V95	TSL:1 GENCODE basic APPRIS P1
Pth2r-202	ENSMUST00000140190.7	631	174aa	Protein coding	-	D3YV95	CDS 3' incomplete TSL:3

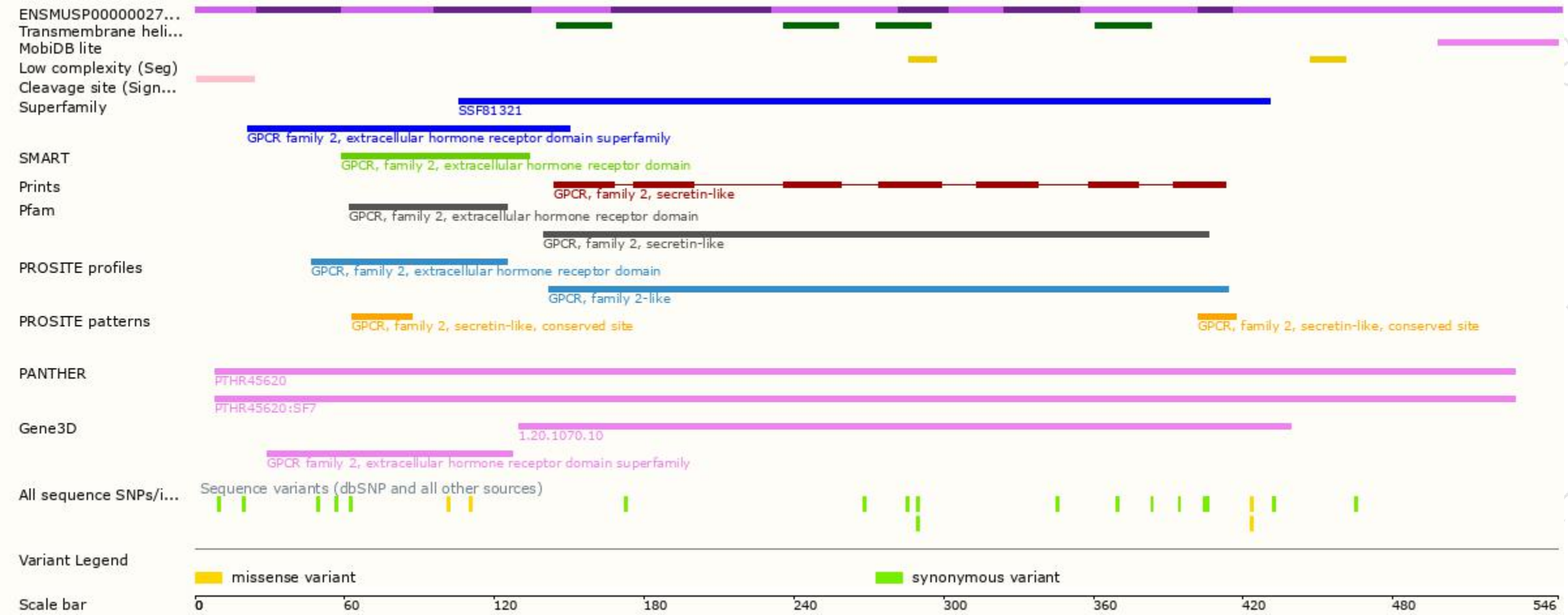
The strategy is based on the design of *Pth2r-201* transcript,The transcription is shown below



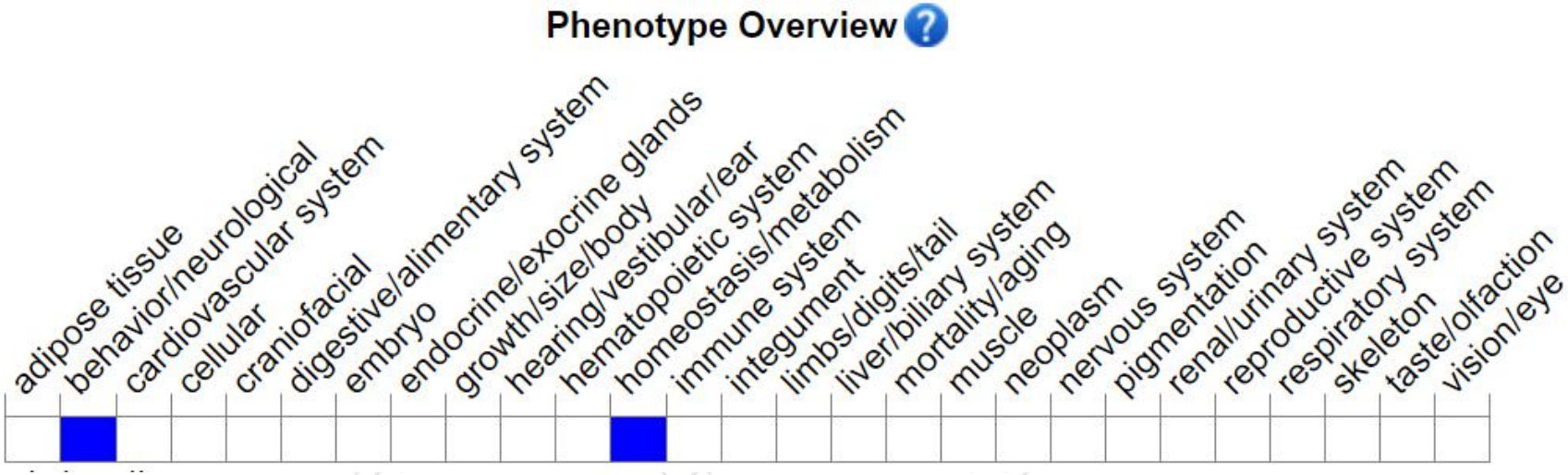
Genomic location distribution



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/).

Homozygous KO reduces core body temperature in pregnant and lactating females and affects locomotor and grooming behavior of lactating females.

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

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