

Lef1 Cas9-CKO Strategy

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Project Overview

Project Name

Lef1

Project type

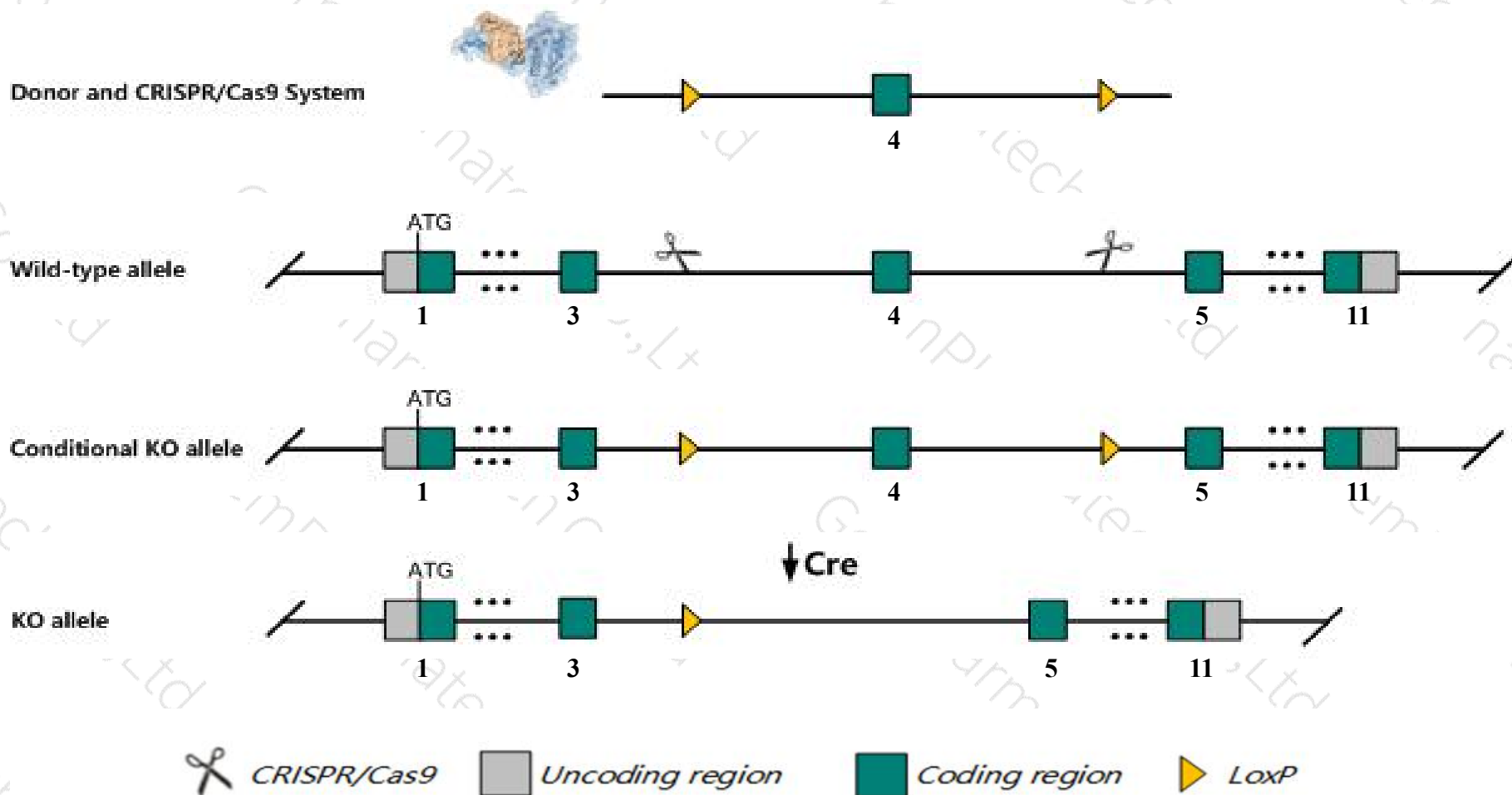
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Lef1* gene. The schematic diagram is as follows:



Technical routes

- The *Lef1* gene has 8 transcripts. According to the structure of *Lef1* gene, exon4 of *Lef1-201* (ENSMUST00000029611.13) transcript is recommended as the knockout region. The region contains 133bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Lef1* 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 null allele are small and die postnatally showing lack of teeth, mammary and uterine glands, whiskers, body hair, dermal-associated fat, and a dentate gyrus, as well as defects in hippocampus morphology, hair follicle development, retinal vasculature, and vascular regression.
- The *Lef1* gene is located on the Chr3. 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)

Lef1 lymphoid enhancer binding factor 1 [Mus musculus (house mouse)]

Gene ID: 16842, updated on 19-Mar-2019

Summary



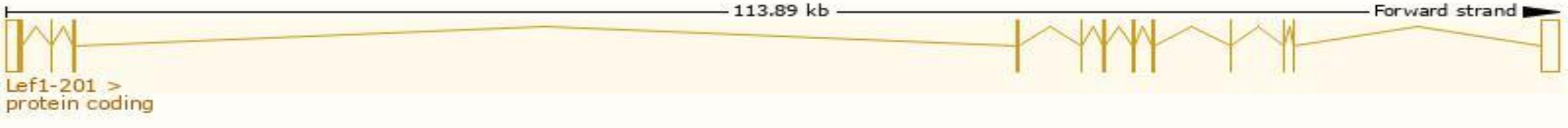
Official Symbol	Lef1 provided by MGI
Official Full Name	lymphoid enhancer binding factor 1 provided by MGI
Primary source	MGI:MGI:96770
See related	Ensembl:ENSMUSG00000027985
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	3000002B05, AI451430, Lef-1
Expression	Biased expression in thymus adult (RPKM 63.3), spleen adult (RPKM 14.2) and 8 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

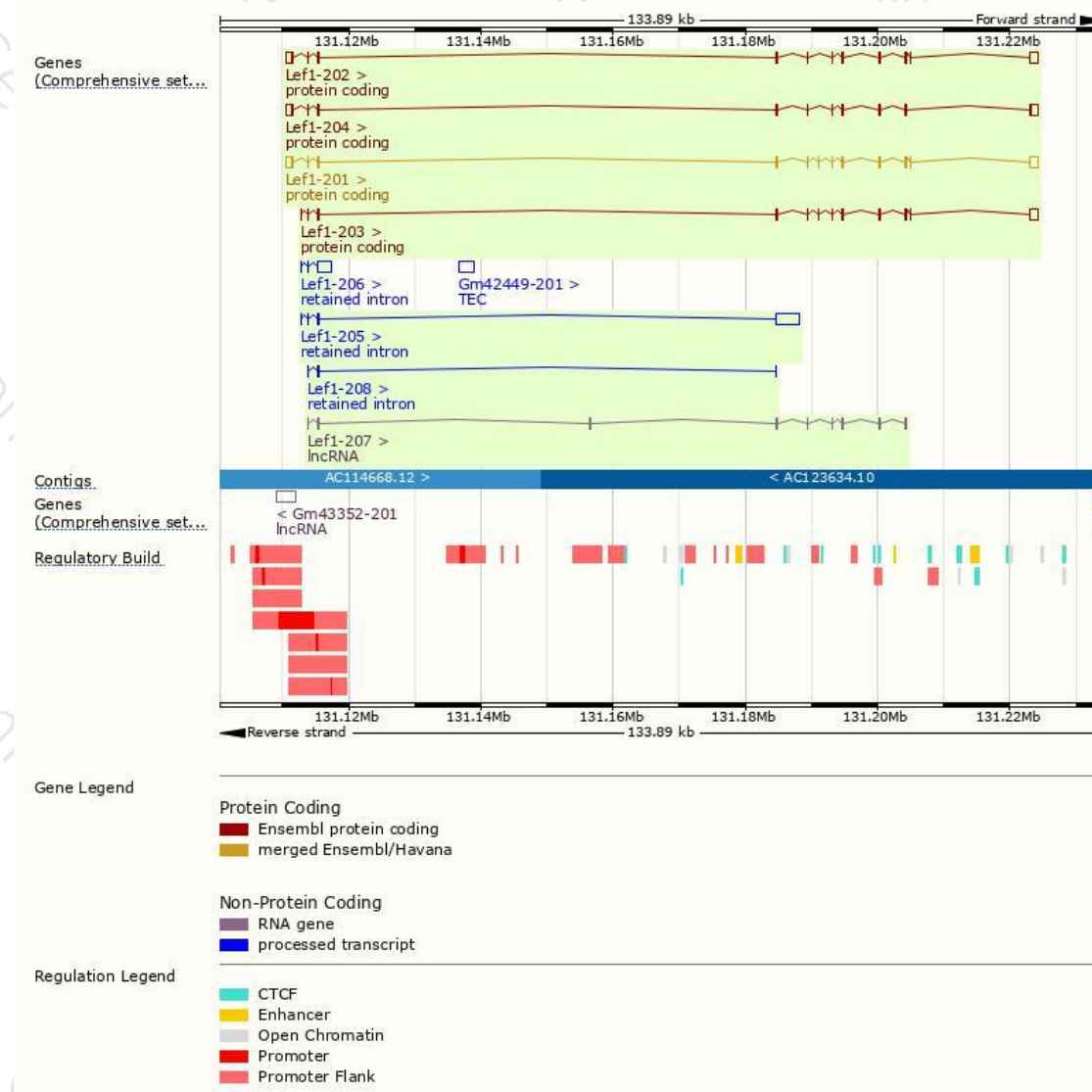
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Lef1-201	ENSMUST00000029611.13	3482	397aa	Protein coding	CCDS17842	P27782 Q3TYB0	TSL:1 GENCODE basic APPRIS P3
Lef1-204	ENSMUST00000106341.8	3357	384aa	Protein coding	CCDS71319	Q8BGZ9	TSL:1 GENCODE basic APPRIS ALT 1
Lef1-203	ENSMUST00000098611.3	2298	331aa	Protein coding	CCDS71320	Q8C402	TSL:1 GENCODE basic
Lef1-202	ENSMUST00000066849.12	3398	369aa	Protein coding	-	D3Z654	TSL:5 GENCODE basic APPRIS ALT 1
Lef1-207	ENSMUST00000198624.1	918	No protein	Processed transcript	-	-	TSL:5
Lef1-205	ENSMUST00000132737.5	3656	No protein	Retained intron	-	-	TSL:1
Lef1-206	ENSMUST00000136147.7	2066	No protein	Retained intron	-	-	TSL:1
Lef1-208	ENSMUST00000200166.4	345	No protein	Retained intron	-	-	TSL:2

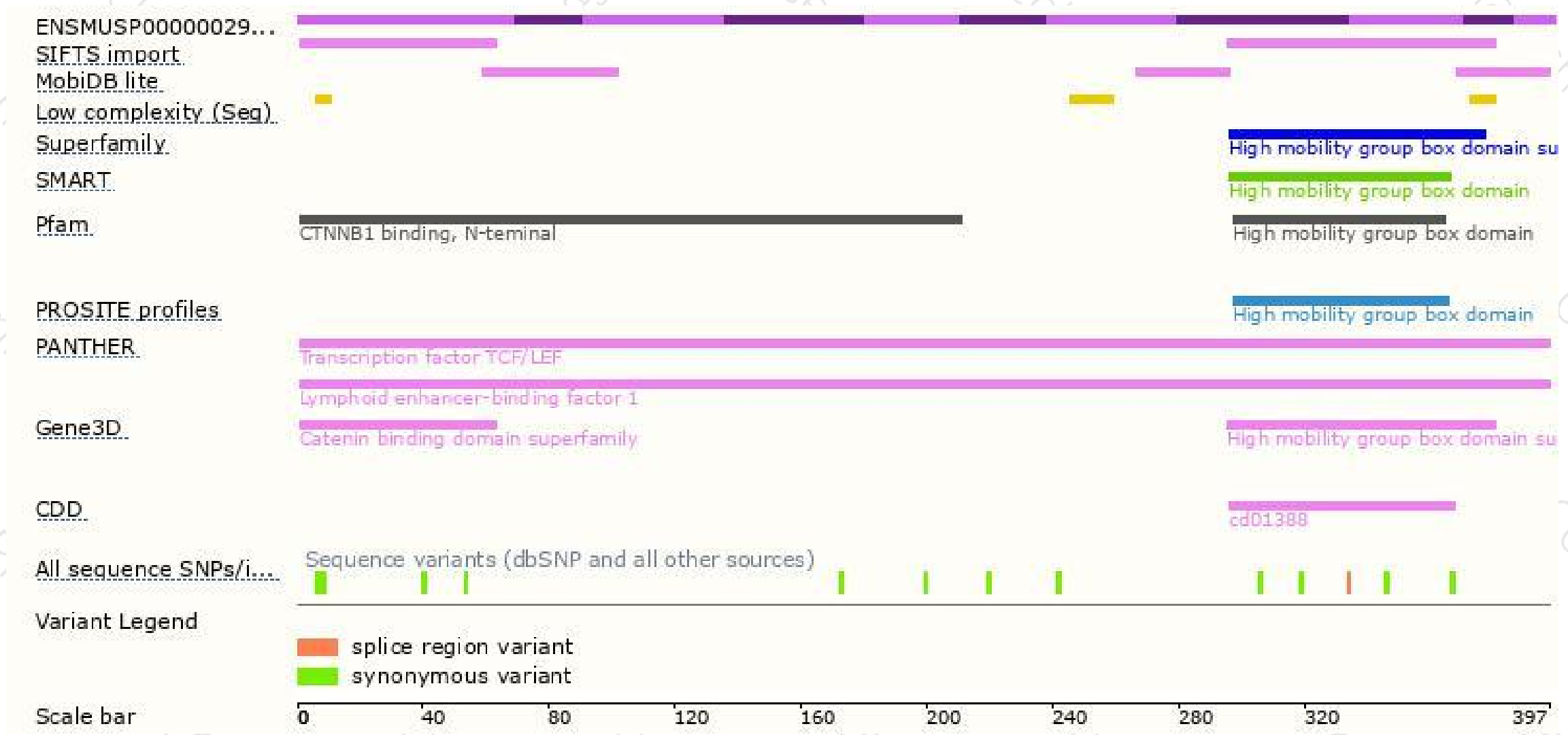
The strategy is based on the design of *Lef1-201* transcript,The transcription is shown below



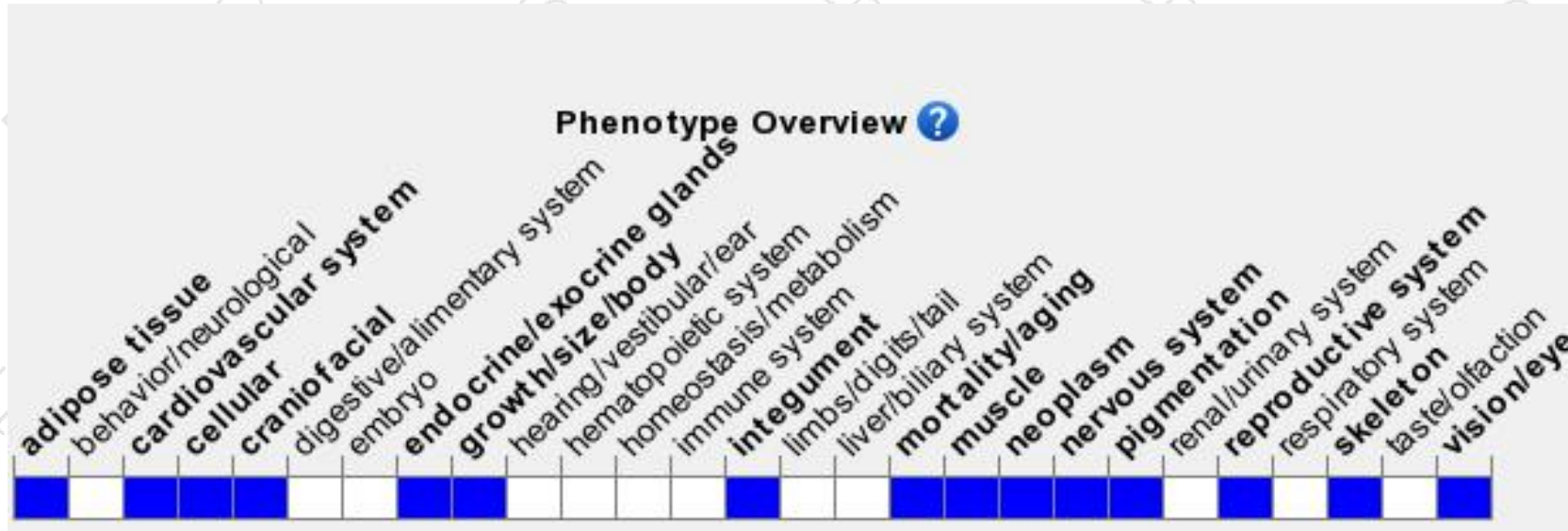
Genomic location distribution



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 null allele are small and die postnatally showing lack of teeth, mammary and uterine glands, whiskers, body hair, dermal-associated fat, and a dentate gyrus, as well as defects in hippocampus morphology, hair follicle development, retinal vasculature, and vascular regression.

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

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