

Klf4 Cas9-CKO Strategy

Designer:

Huan Wang

Reviewer:

Huan Fan

Design Date:

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Project Overview

Project Name

Klf4

Project type

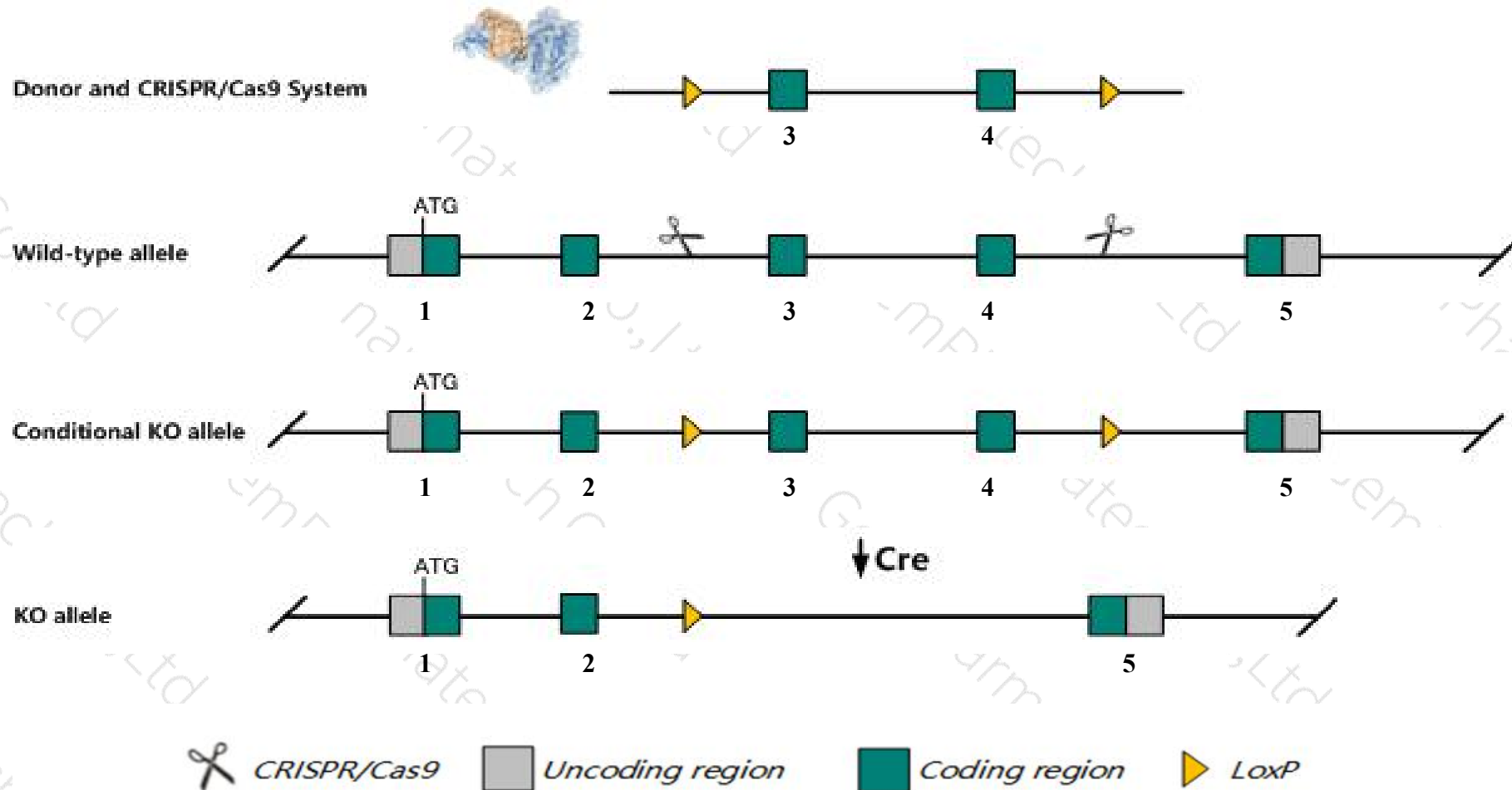
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Klf4* gene. The schematic diagram is as follows:



Technical routes

- The *Klf4* gene has 3 transcripts. According to the structure of *Klf4* gene, exon3-exon4 of *Klf4-201* (ENSMUST00000107619.2) transcript is recommended as the knockout region. The region contains 1150bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Klf4* 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, Homozygotes for targeted null mutations die shortly after birth due to a skin defect that results in loss of fluids. Mutants also show a dramatic decrease in the number of goblet cells of the colon.
- The *Klf4* gene is located on the Chr4. 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)

Klf4 Kruppel-like factor 4 (gut) [Mus musculus (house mouse)]

Gene ID: 16600, updated on 26-Mar-2019

Summary



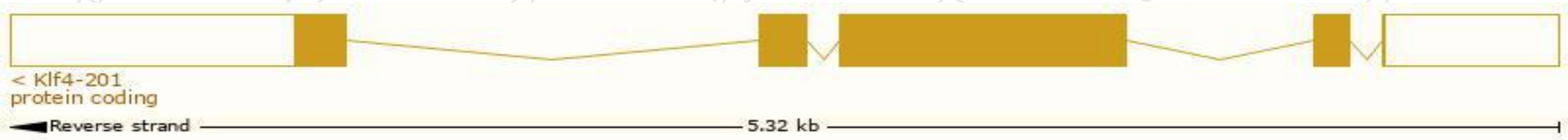
Official Symbol	Klf4 provided by MGI
Official Full Name	Kruppel-like factor 4 (gut) provided by MGI
Primary source	MGI:MGI:1342287
See related	Ensembl:ENSMUSG000000003032
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	EZF, Gklf, Zie
Expression	Biased expression in colon adult (RPKM 221.0), stomach adult (RPKM 155.0) and 9 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

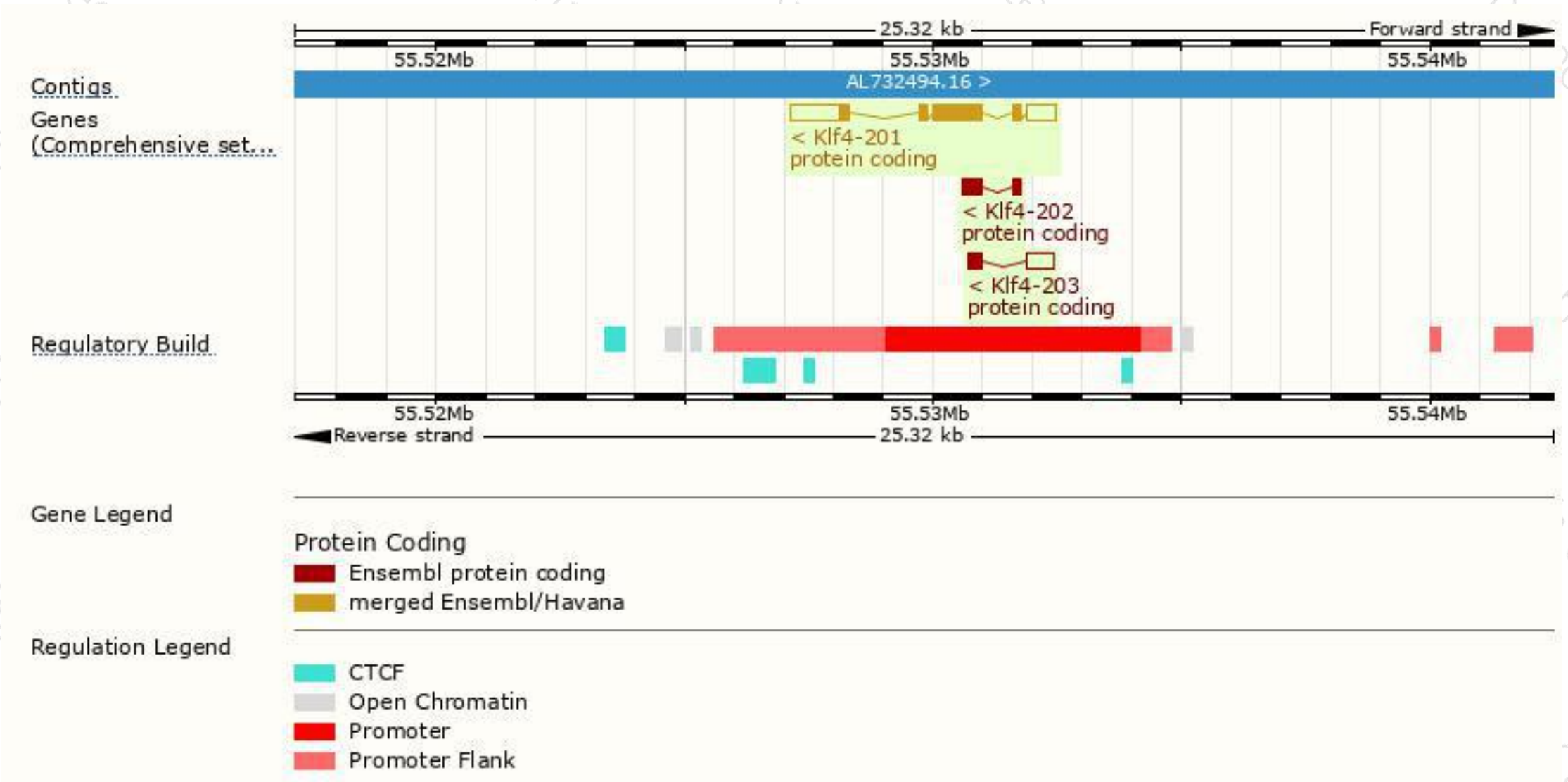
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Klf4-201	ENSMUST00000107619.2	3029	483aa	Protein coding	CCDS18195	F2YID5 Q60793	TSL:1 GENCODE basic APPRIS P1
Klf4-203	ENSMUST00000132746.1	859	82aa	Protein coding	-	B7ZCH1	CDS 3' incomplete TSL:3
Klf4-202	ENSMUST00000129250.1	522	160aa	Protein coding	-	B7ZCH2	CDS 3' incomplete TSL:2

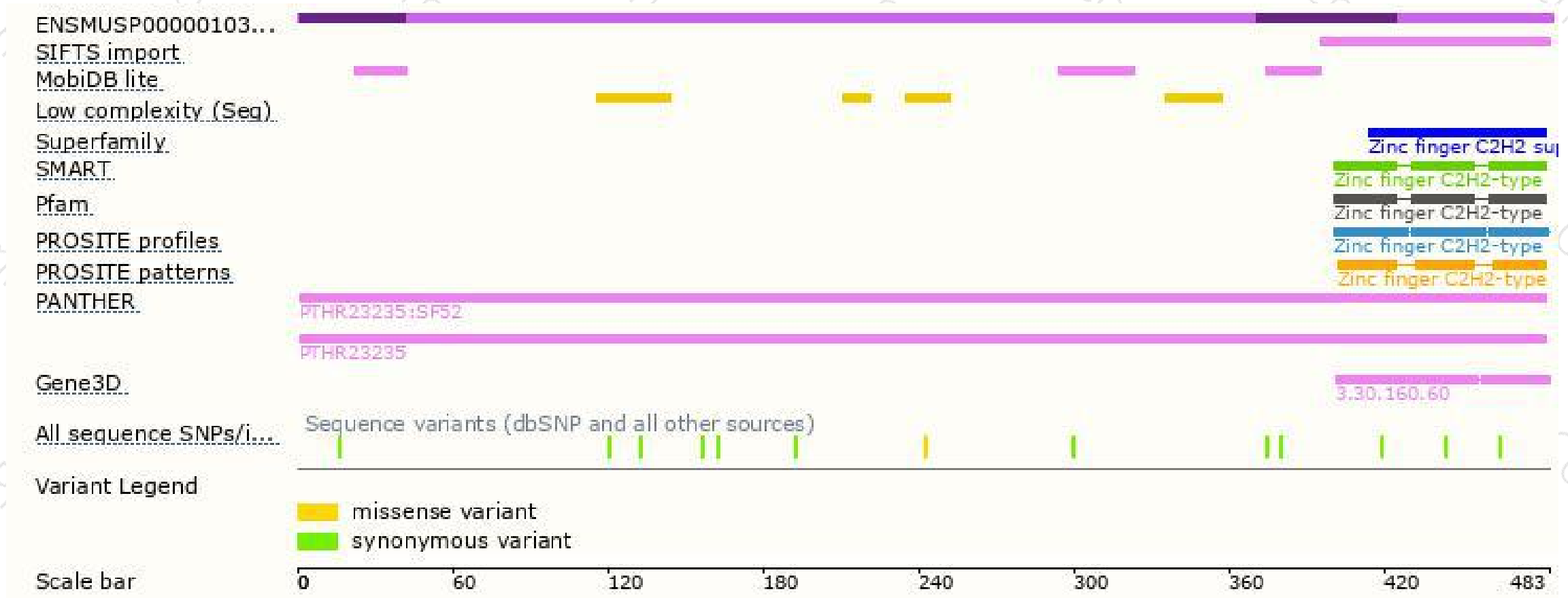
The strategy is based on the design of *Klf4-201* transcript,The transcription is shown below



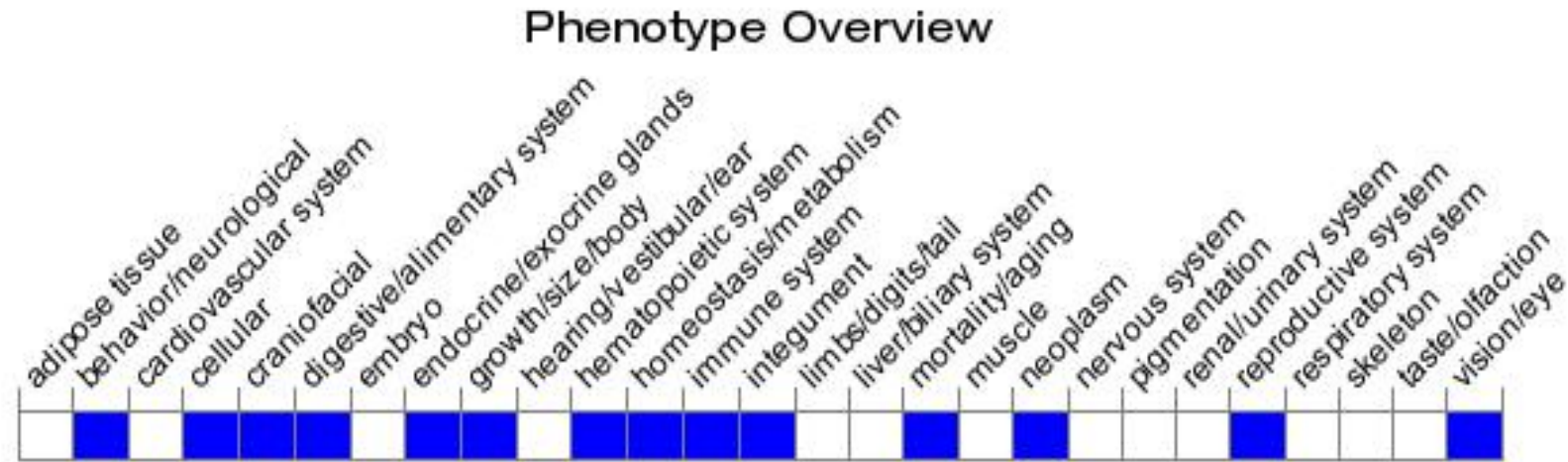
Genomic location distribution



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/>).

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

Tel: 400-9660890

