

Atoh8 Cas9-CKO Strategy

Designer:

JiaYu

Reviewer:

Xiaojing Li

Design Date:

2020-2-12

Project Overview

Project Name

Atoh8

Project type

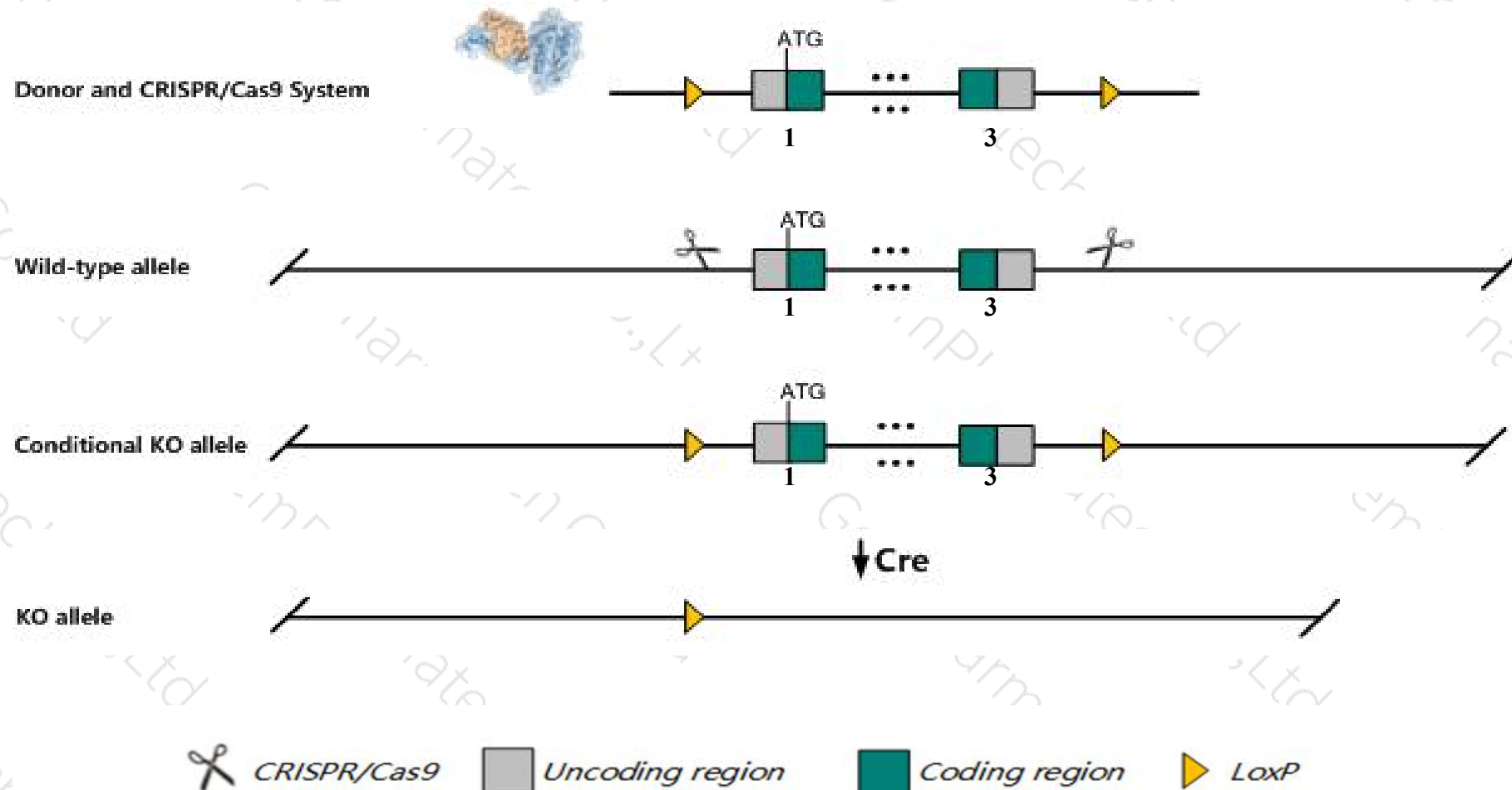
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Atoh8* gene. The schematic diagram is as follows:



- The *Atoh8* gene has 3 transcripts. According to the structure of *Atoh8* gene, exon1-exon3 of *Atoh8-201* (ENSMUST00000042646.7) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Atoh8* 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-in allele appear to be developmentally arrested at or slightly after gastrulation.
- The *Atoh8* 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)

Atoh8 atonal bHLH transcription factor 8 [Mus musculus (house mouse)]

Gene ID: 71093, updated on 31-Jan-2019

Summary



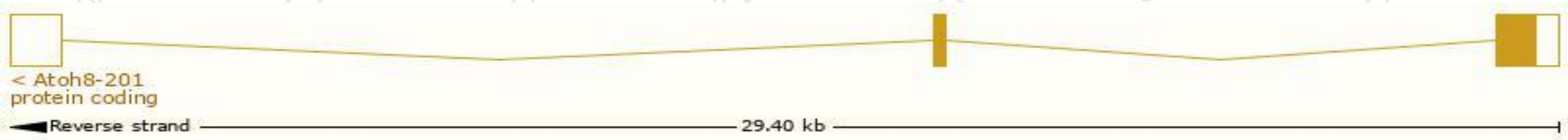
Official Symbol	Atoh8 provided by MGI
Official Full Name	atonal bHLH transcription factor 8 provided by MGI
Primary source	MGI:MGI:1918343
See related	Ensembl:ENSMUSG000000037621
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	4933425C05Rik, Hath6, Math6, bHLHa21, okadin
Expression	Biased expression in testis adult (RPKM 68.7), lung adult (RPKM 9.9) and 9 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

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

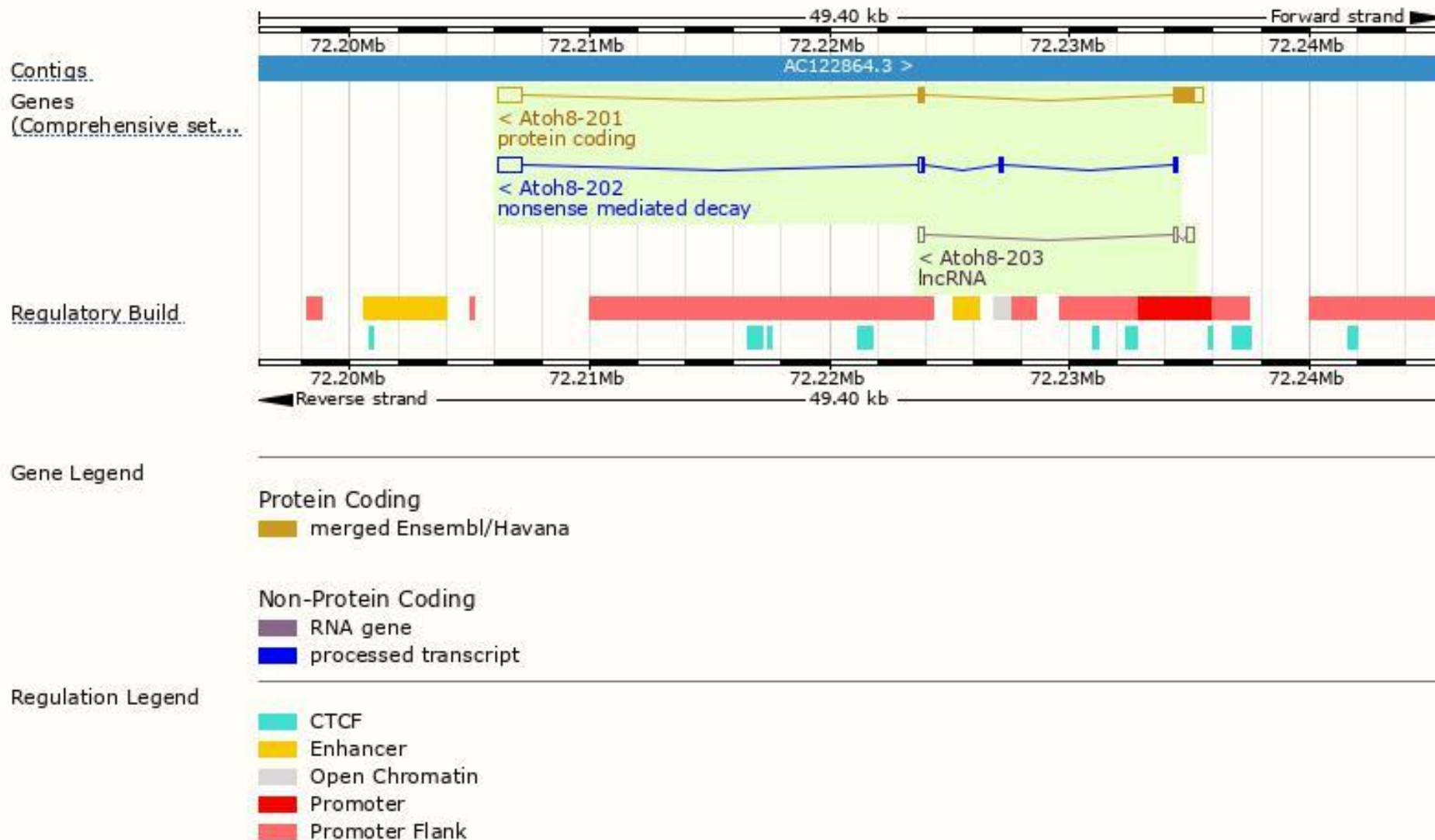
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Atoh8-201	ENSMUST00000042646.7	2355	322aa	Protein coding	CCDS39511	Q99NA2	TSL:1 GENCODE basic APPRIS P1
Atoh8-202	ENSMUST00000206425.1	1467	114aa	Nonsense mediated decay	-	A0A0U1RP16	CDS 5' incomplete TSL:1
Atoh8-203	ENSMUST00000206553.1	659	No protein	lncRNA	-	-	TSL:5

The strategy is based on the design of *Atoh8-201* transcript,The transcription is shown below

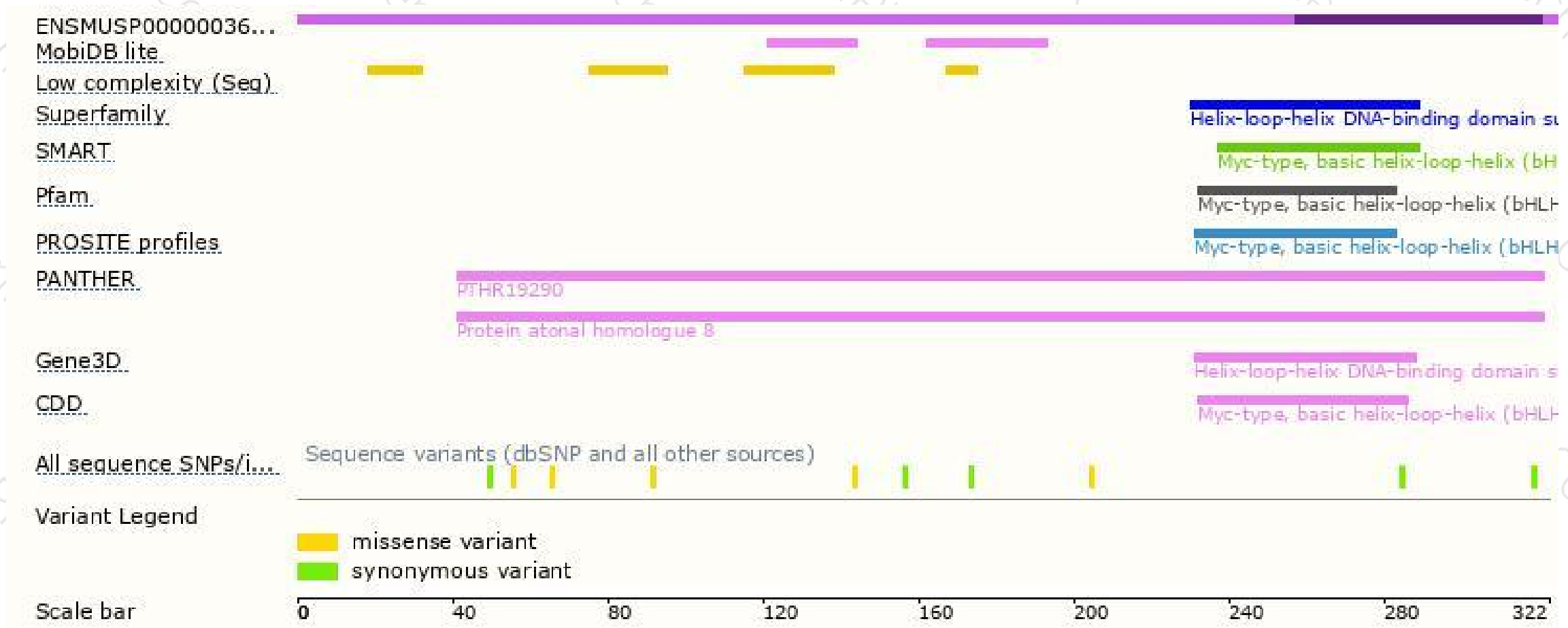




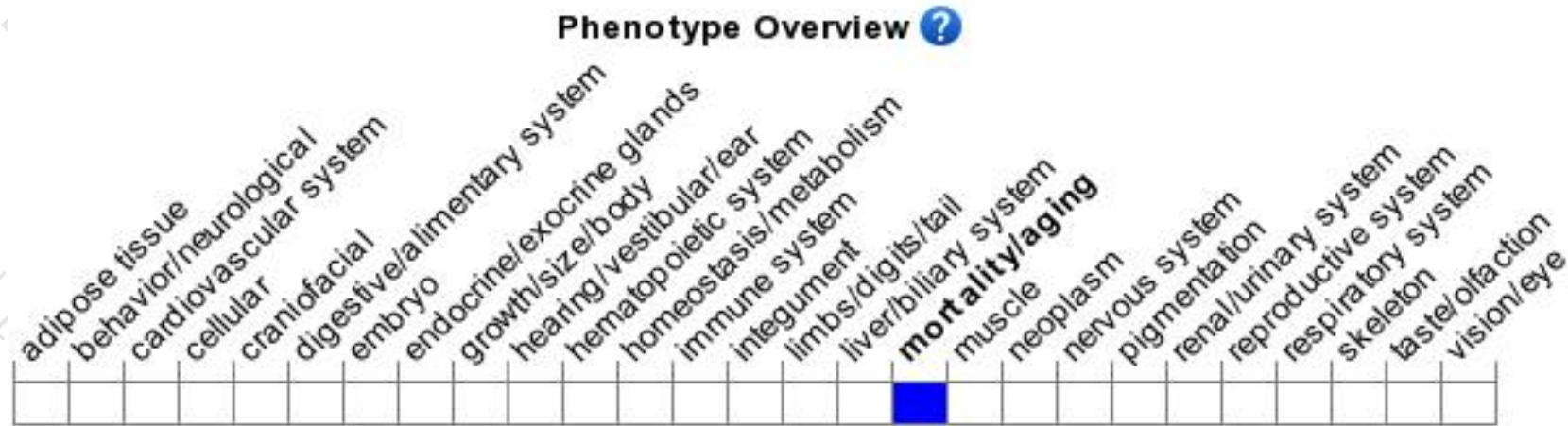
Genomic location distribution



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-in allele appear to be developmentally arrested at or slightly after gastrulation.

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

Tel: 400-9660890

