

Cdk9 Cas9-CKO Strategy

Designer: Huan Wang

Reviewer: Jinling Wang

Design Date: 2024-1-9

Overview

Target Gene Name

- Cdk9

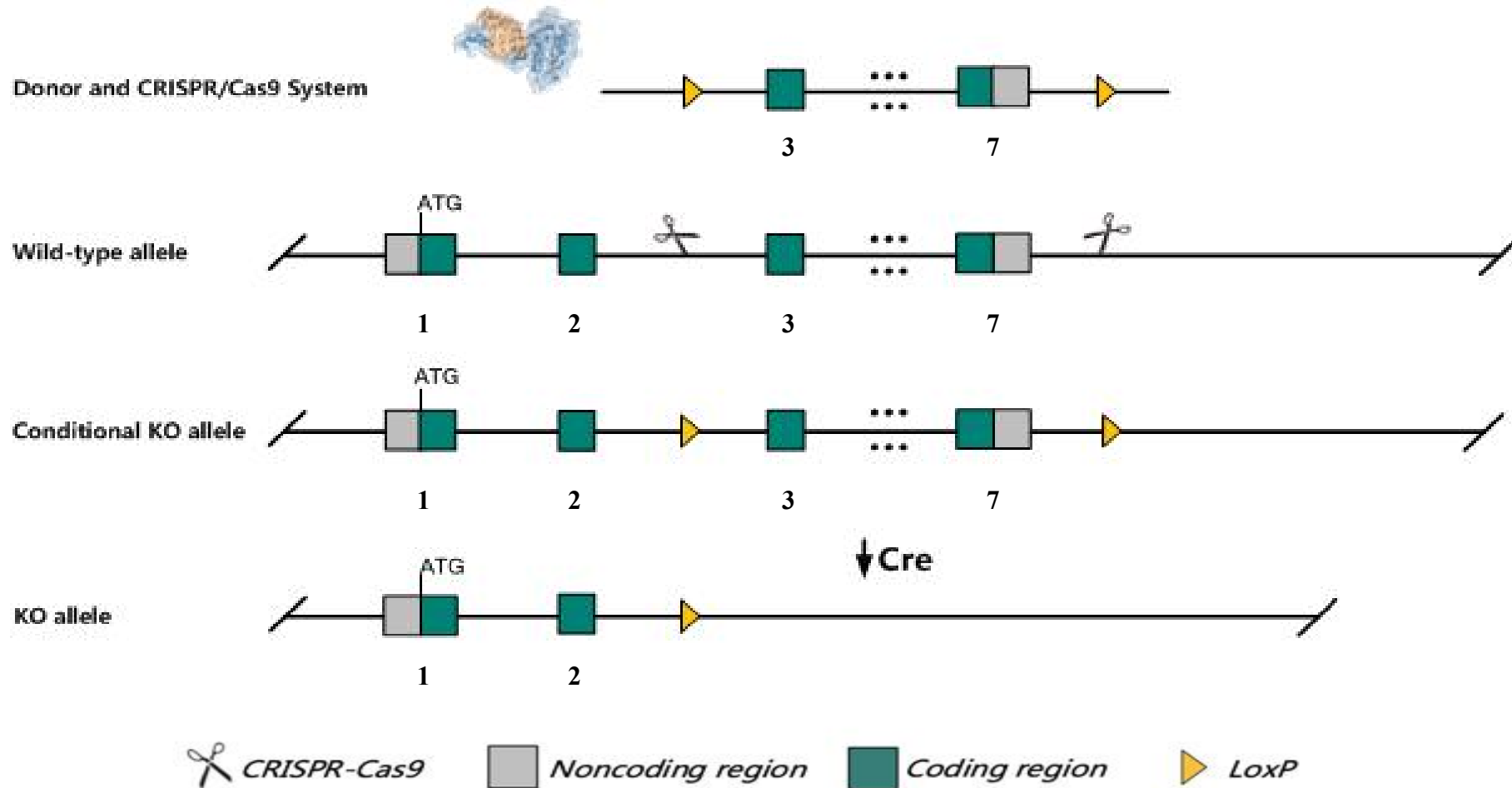
Project Type

- Cas9-CKO

Genetic Background

- C57BL/6JGpt

Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the *Cdk9* gene.

Technical Information

- The *Cdk9* gene has 9 transcripts. According to the structure of *Cdk9* gene, exon3-exon7 of *Cdk9*-201 (ENSMUST00000009699.16) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Cdk9* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.

Gene Information

Cdk9 cyclin dependent kinase 9 [Mus musculus (house mouse)]

Gene ID: 107951, updated on 18-May-2023

Summary

Official Symbol	Cdk9 provided by MGI
Official Full Name	cyclin dependent kinase 9 provided by MGI
Primary source	MGI:MGI:1328368
See related	Ensembl:ENSMUSG00000009555
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	PITALRE
Summary	Enables several functions, including RNA polymerase II CTD heptapeptide repeat kinase activity; chromatin binding activity; and nucleic acid binding activity. Involved in positive regulation of histone H2B ubiquitination and positive regulation of histone phosphorylation. Located in nucleus. Part of cyclin/CDK positive transcription elongation factor complex. Is expressed in several structures, including alimentary system; early embryo; genitourinary system; hemolymphoid system; and nervous system. Orthologous to human CDK9 (cyclin dependent kinase 9). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Ubiquitous expression in ovary adult (RPKM 28.4), thymus adult (RPKM 26.4) and 28 other tissues See more
Orthologs	human all

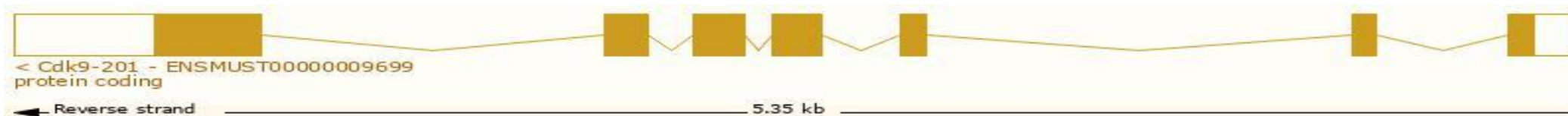
Source: <https://www.ncbi.nlm.nih.gov/>

Transcript Information

The gene has 9 transcripts, all transcripts are shown below:

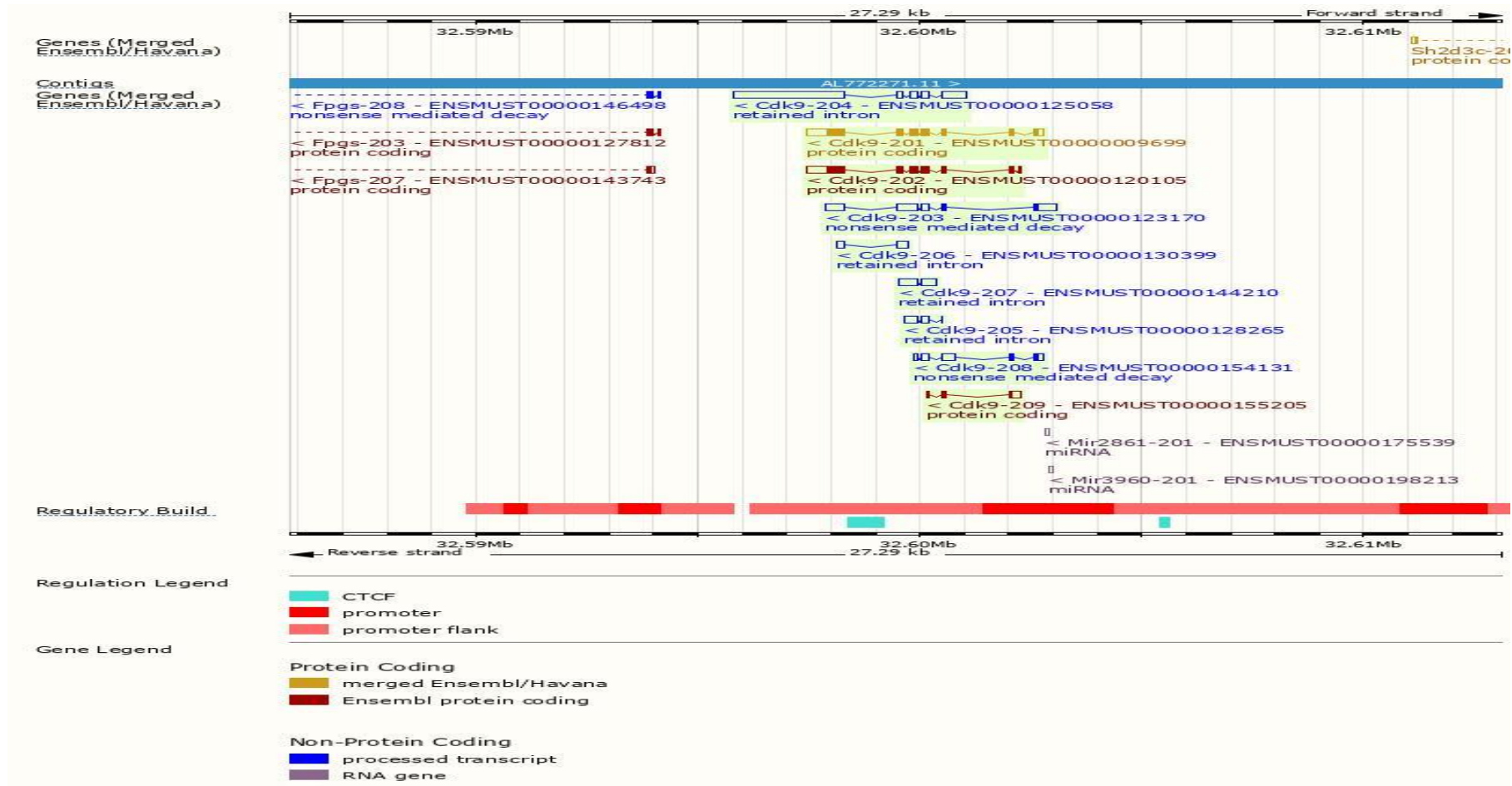
Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
ENSMUST0000009699.16	Cdk9-201	1733	372aa	Protein coding	CCDS15927	Q99J95-1	Ensembl Canonical Gencode basic APPRIS P1 TSL:1
ENSMUST00000120105.8	Cdk9-202	1565	321aa	Protein coding		Q99J95-2	Gencode basic TSL:1
ENSMUST00000155205.2	Cdk9-209	366	54aa	Protein coding		B0R021	TSL:3 CDS 3' incomplete
ENSMUST00000123170.8	Cdk9-203	1689	36aa	Nonsense mediated decay		D6RGN3	TSL:1
ENSMUST00000154131.2	Cdk9-208	846	61aa	Nonsense mediated decay		D6RHP4	TSL:3
ENSMUST00000125058.8	Cdk9-204	3565	No protein	Retained intron		-	TSL:1
ENSMUST00000144210.2	Cdk9-207	720	No protein	Retained intron		-	TSL:2
ENSMUST00000128265.8	Cdk9-205	482	No protein	Retained intron		-	TSL:2
ENSMUST00000130399.2	Cdk9-206	444	No protein	Retained intron		-	TSL:1

The strategy is based on the design of *Cdk9*-201 transcript, the transcription is shown below:

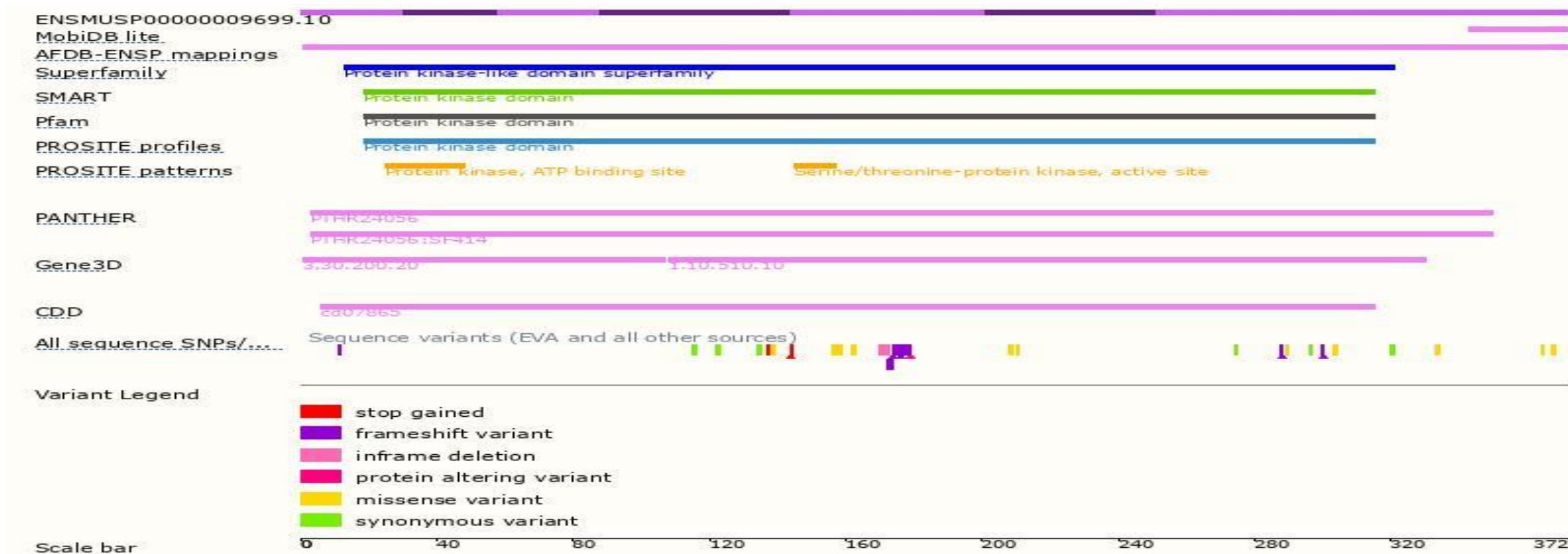


Source: <https://www.ensembl.org>

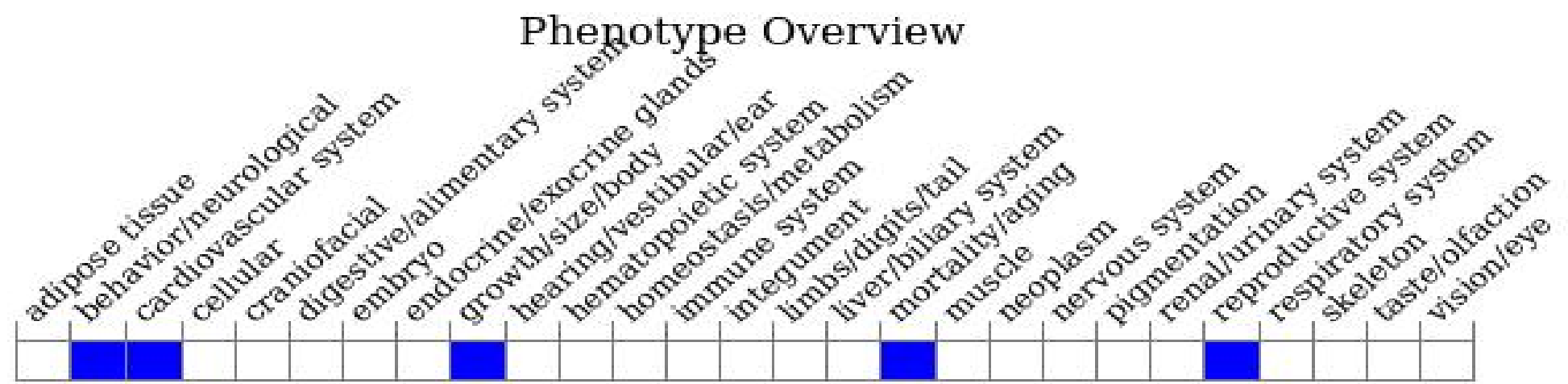
Genomic Information



Protein Information



Mouse Phenotype Information (MGI)



•

Important Information

- The floxed region is about 1.5kb away from *Fpgs* gene. Knockout the region may affect the function of *Fpgs* gene.
- *Cdk9* is located on Chr2. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.