

***Rab7* Cas9-CKO Strategy**

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Project Overview

Project Name

Rab7

Project type

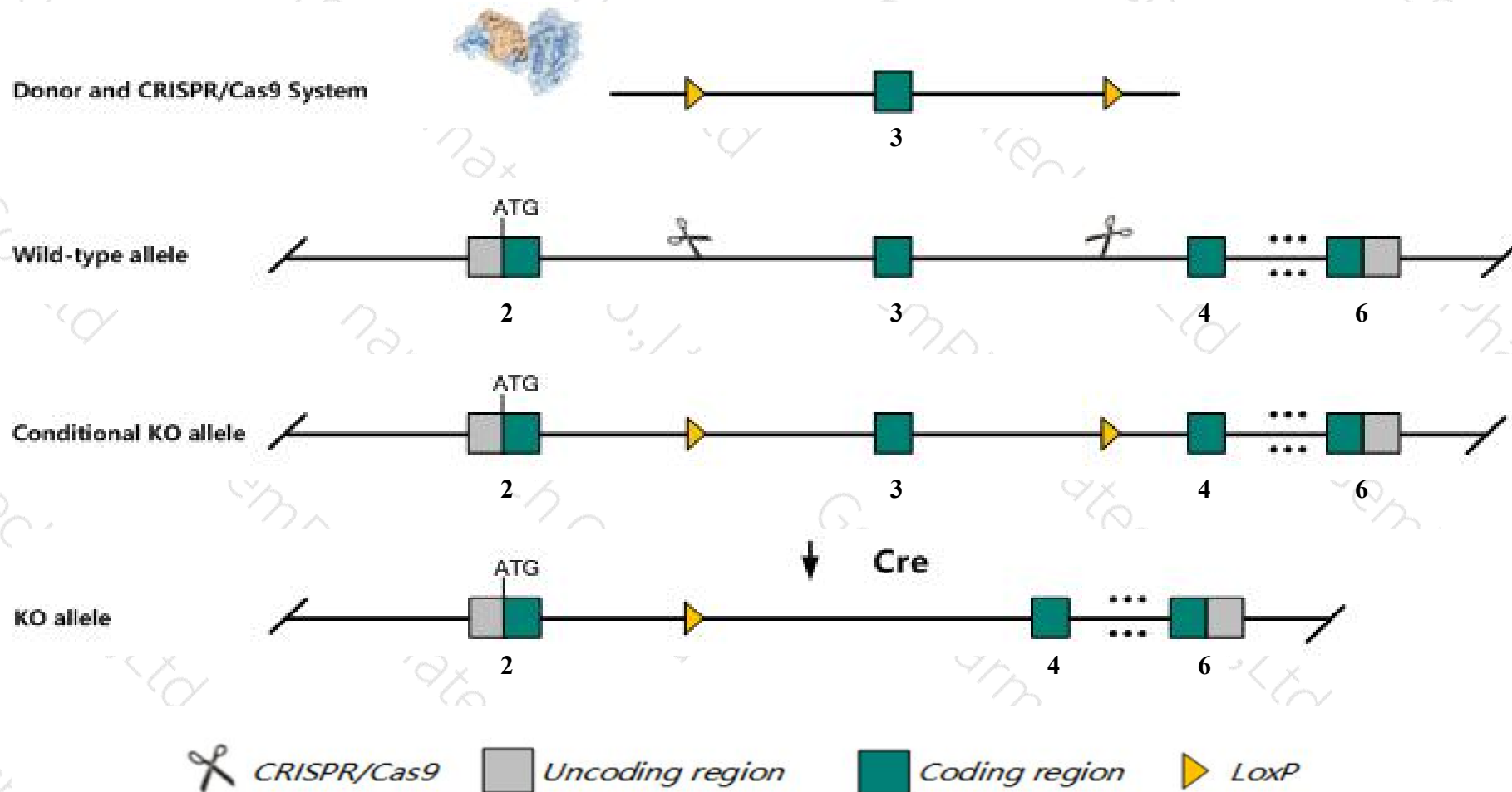
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rab7* gene. The schematic diagram is as follows:



Technical routes

- The *Rab7* gene has 7 transcripts. According to the structure of *Rab7* gene, exon3 of *Rab7*-205(ENSMUST00000113600.9) transcript is recommended as the knockout region. The region contains 127bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rab7* 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 targeted alleles exhibit abnormal endocytosis within the visceral endoderm, failure of elongation along the primitive streak and incomplete gastrulation.
- The *Rab7* 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)

Rab7 RAB7, member RAS oncogene family [Mus musculus (house mouse)]

Gene ID: 19349, updated on 13-Mar-2020

Summary



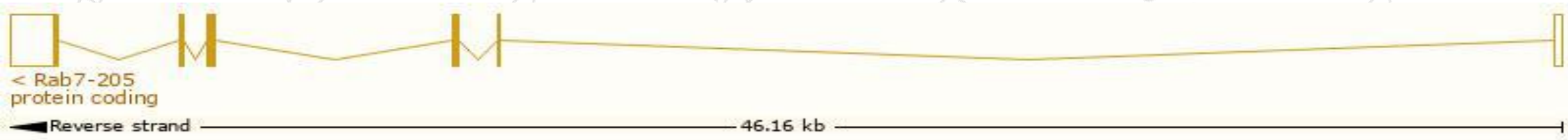
Official Symbol	Rab7 provided by MGI
Official Full Name	RAB7, member RAS oncogene family provided by MGI
Primary source	MGI:MGI:105068
See related	Ensembl:ENSMUSG00000079477
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Rab7a
Expression	Ubiquitous expression in bladder adult (RPKM 76.7), kidney adult (RPKM 66.7) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

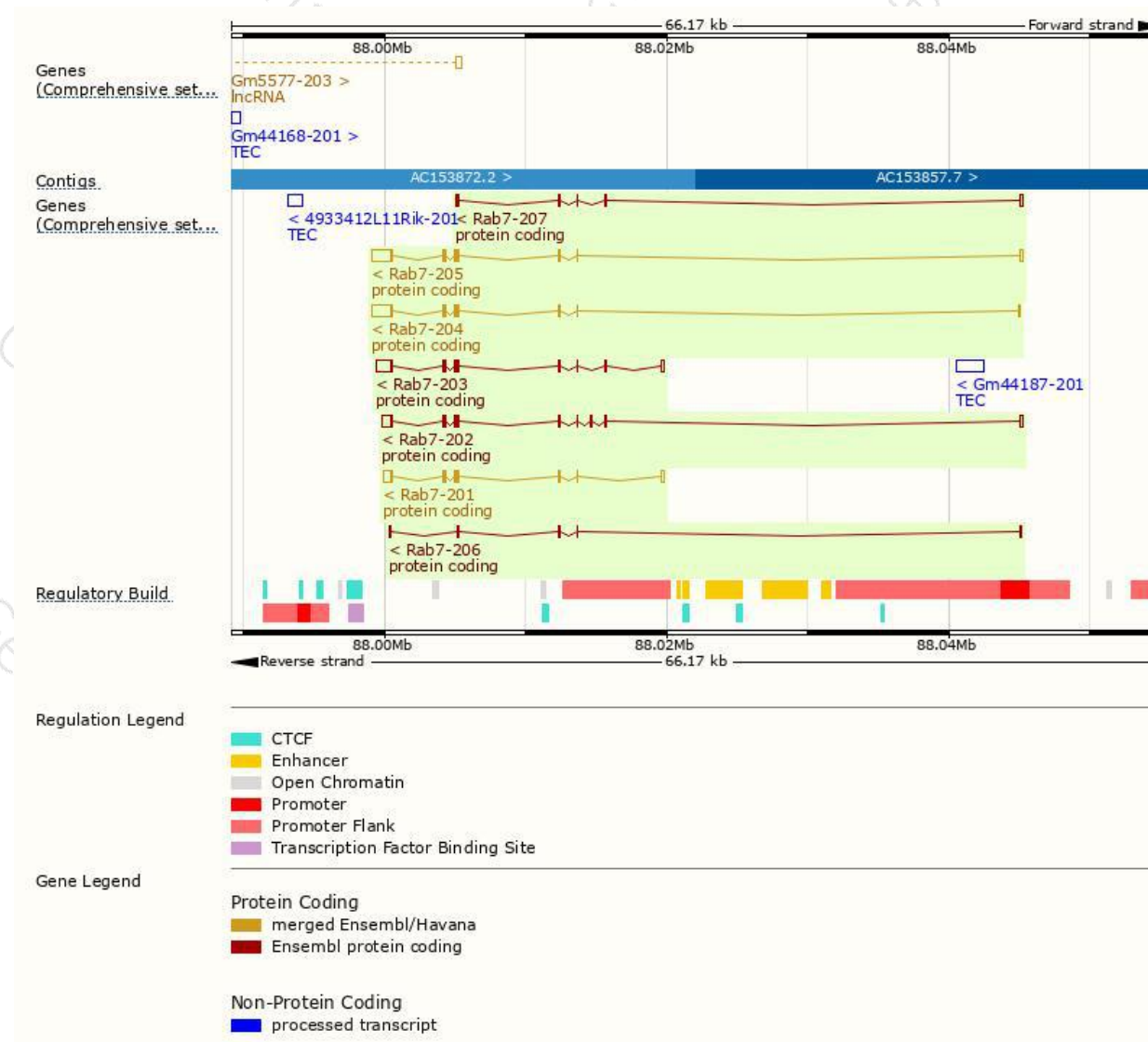
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rab7-205	ENSMUST00000113600.9	2159	207aa	Protein coding	CCDS39552	P51150 Q4FJQ0	TSL:1 GENCODE basic APPRIS P1
Rab7-204	ENSMUST00000113598.7	2031	207aa	Protein coding	CCDS39552	P51150 Q4FJQ0	TSL:1 GENCODE basic APPRIS P1
Rab7-203	ENSMUST00000113597.7	1859	207aa	Protein coding	CCDS39552	P51150 Q4FJQ0	TSL:1 GENCODE basic APPRIS P1
Rab7-202	ENSMUST00000113596.7	1657	207aa	Protein coding	CCDS39552	P51150 Q4FJQ0	TSL:1 GENCODE basic APPRIS P1
Rab7-201	ENSMUST00000095048.5	1275	207aa	Protein coding	CCDS39552	P51150 Q4FJQ0	TSL:1 GENCODE basic APPRIS P1
Rab7-207	ENSMUST00000204126.1	550	98aa	Protein coding	-	A0A0N4SVG9	CDS 3' incomplete TSL:3
Rab7-206	ENSMUST00000203674.2	424	88aa	Protein coding	-	A0A0N4SVR6	TSL:5 GENCODE basic

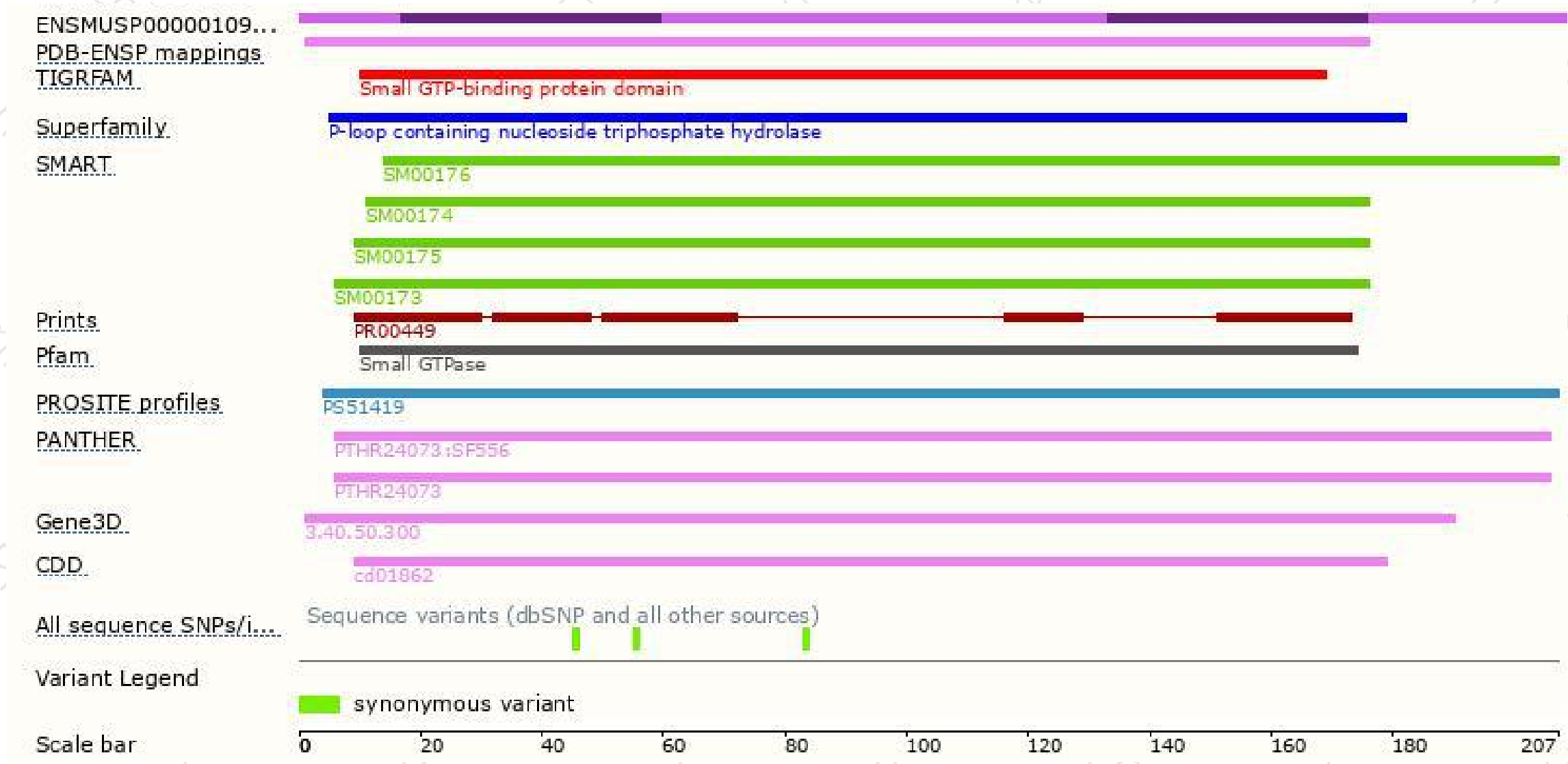
The strategy is based on the design of *Rab7-205* transcript,the transcription is shown below:



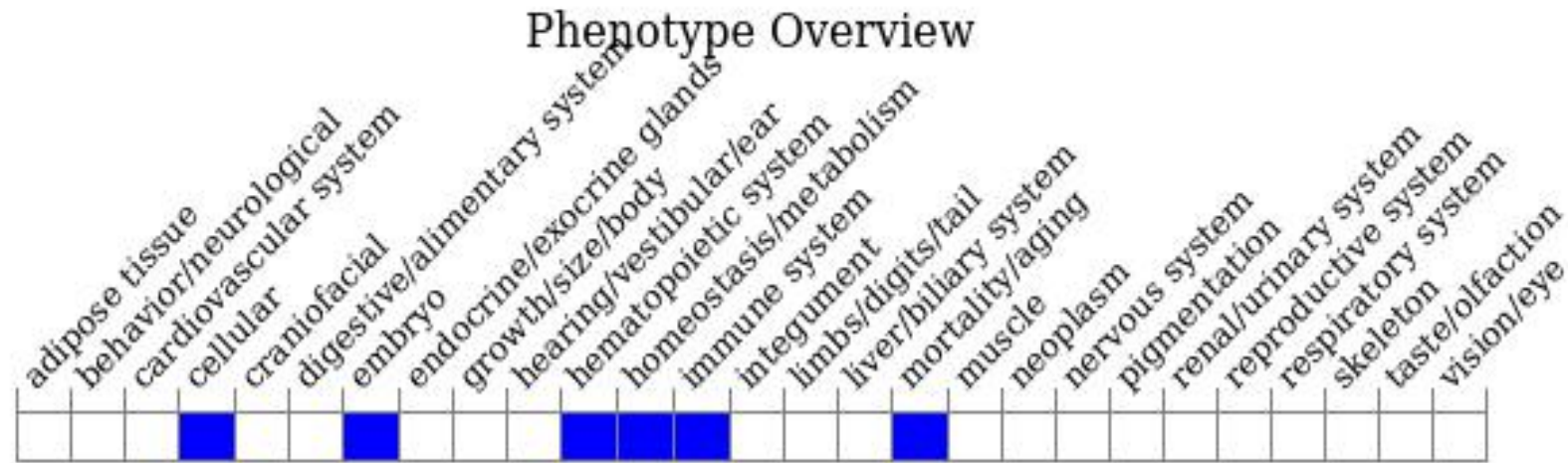
Genomic location distribution



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 targeted alleles exhibit abnormal endocytosis within the visceral endoderm, failure of elongation along the primitive streak and incomplete gastrulation.

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

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