

***Rassf1* Cas9-CKO Strategy**

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Project Overview

Project Name

Rassf1

Project type

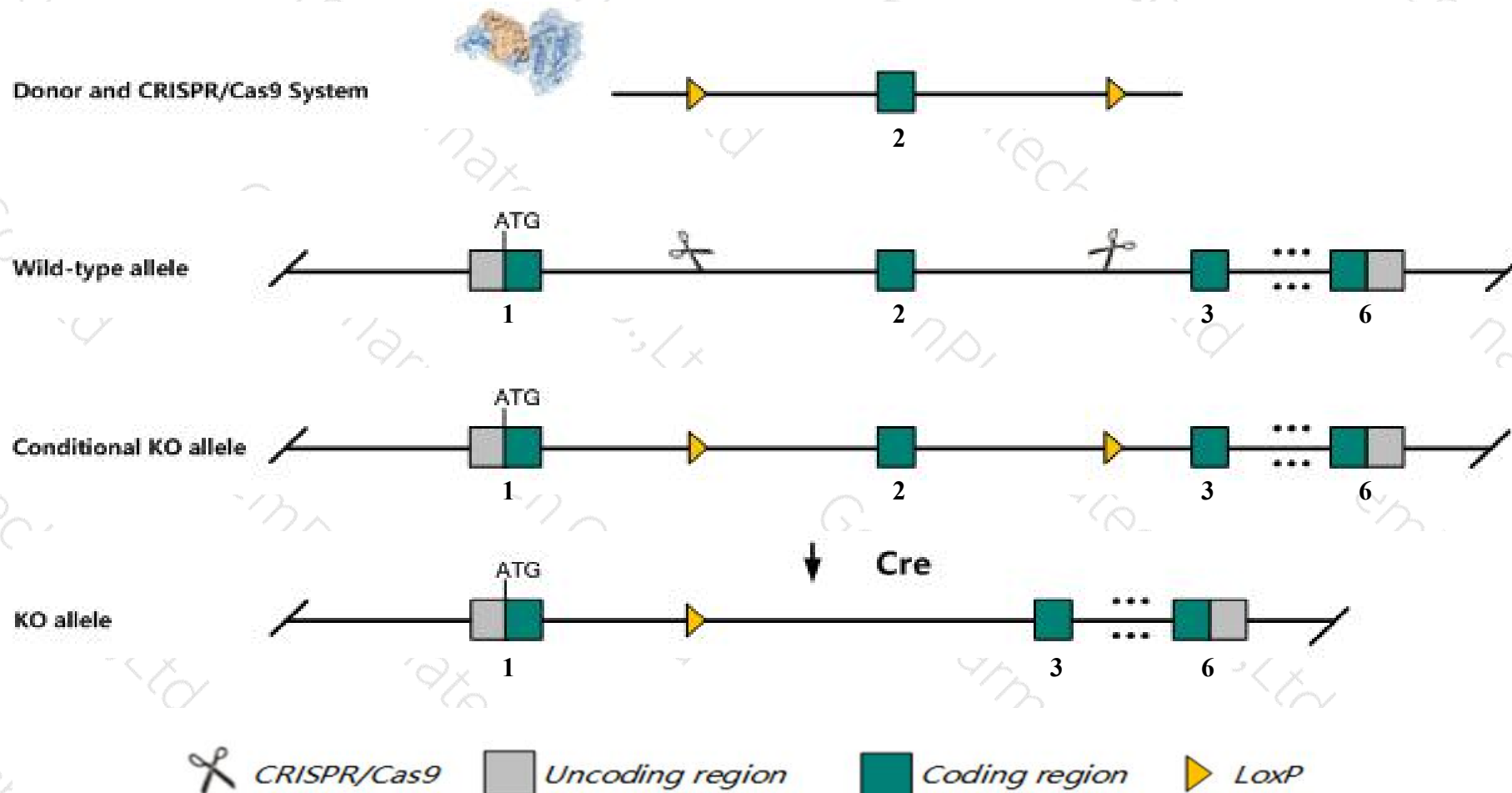
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rassf1* gene. The schematic diagram is as follows:



- The *Rassf1* gene has 10 transcripts. According to the structure of *Rassf1* gene, exon2 of *Rassf1-202* (ENSMUST00000093786.8) transcript is recommended as the knockout region. The region contains 107bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rassf1* 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, Homozygous and heterozygous null mice display increased tumor incidence, especially of lung adenomas and lymphomas, and increased sensitivity to chemically induced tumors.
- Transcript *Rassf1*-203&206&207&208 may not be affected.
- The effect on transcript *Rassf1*-201&209 is unknown.
- The floxed region is near to the N-terminal of *Gm34106* gene and *Tusc2* gene, this strategy may influence the regulatory function of the N-terminal of these genes.
- The *Rassf1* gene is located on the Chr9. 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)

Rassf1 Ras association (RalGDS/AF-6) domain family member 1 [*Mus musculus* (house mouse)]

Gene ID: 56289, updated on 12-Nov-2019

Summary

Official Symbol Rassf1 provided by [MGI](#)
Official Full Name Ras association (RalGDS/AF-6) domain family member 1 provided by [MGI](#)
Primary source [MGI:MGI:1928386](#)
See related [Ensembl:ENSMUSG00000010067](#)
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 123F2; RDA32; NORE2A; REH3P21; Rassf1A; Rassf1B; Rassf1C; AA536941; AU044980
Expression Ubiquitous expression in lung adult (RPKM 22.7), testis adult (RPKM 20.7) and 28 other tissues [See more](#)
Orthologs [human](#) [all](#)

Genomic context

Location: 9; 9 F1

See Rassf1 in [Genome Data Viewer](#)

Exon count: 7

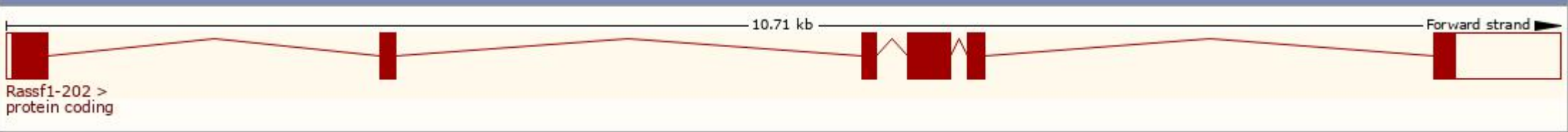
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	9	NC_000075.6 (107551434..107562267)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	9	NC_000075.5 (107456987..107464591)

Transcript information (Ensembl)

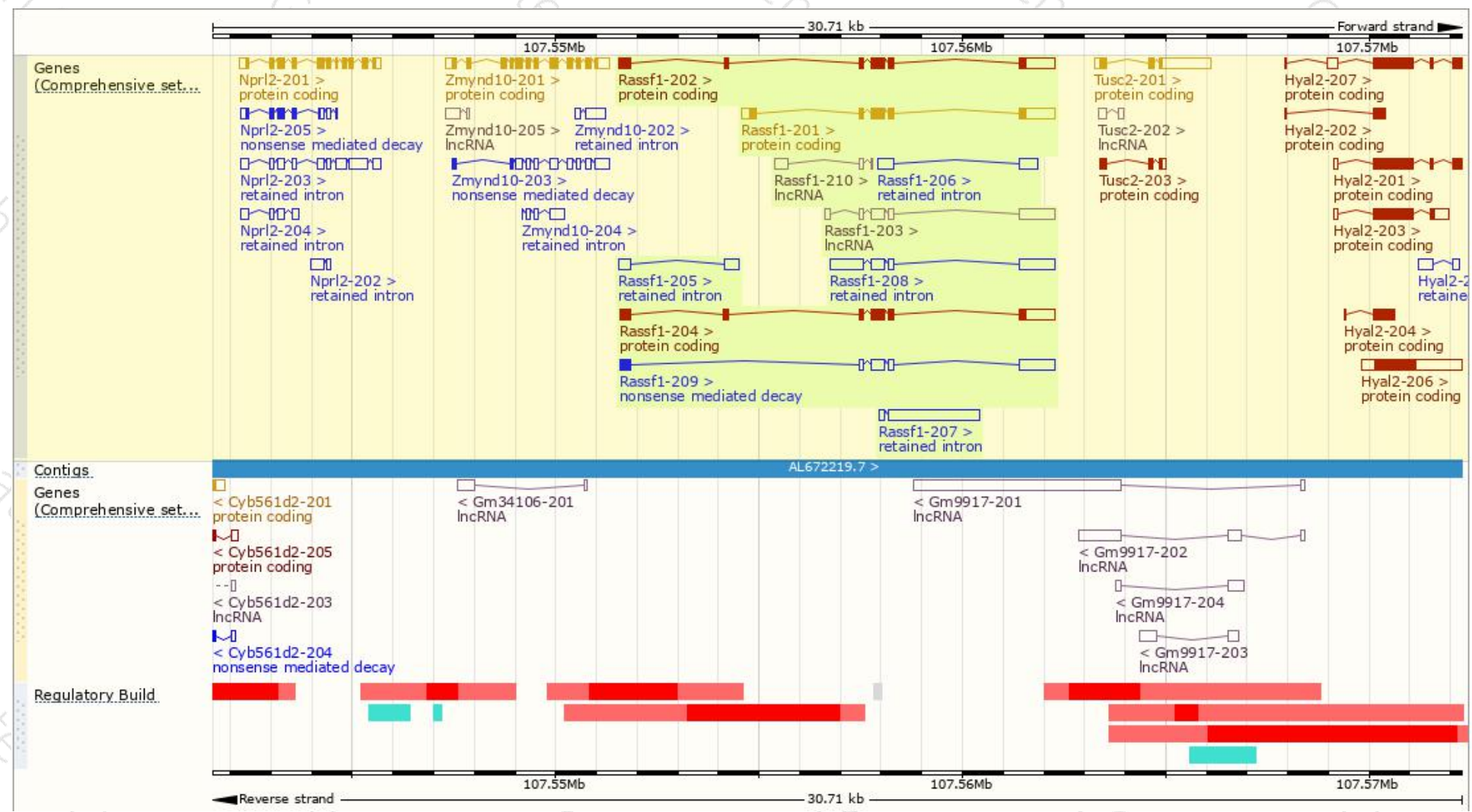
The gene has 10 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rassf1-201	ENSMUST00000010211.6	1727	270aa	Protein coding	CCDS23494	Q99MK9	TSL:1 GENCODE basic
Rassf1-202	ENSMUST00000093786.8	1787	340aa	Protein coding	CCDS57699	A9YZW8 Q99MK9	TSL:1 GENCODE basic APPRIS P1
Rassf1-203	ENSMUST000000121635.6	1525	No protein	lncRNA	-	-	TSL:1
Rassf1-204	ENSMUST000000122225.7	1783	344aa	Protein coding	-	B0QZV7	TSL:5 GENCODE basic
Rassf1-205	ENSMUST000000125080.1	658	No protein	Retained intron	-	-	TSL:3
Rassf1-206	ENSMUST000000125386.1	808	No protein	Retained intron	-	-	TSL:2
Rassf1-207	ENSMUST000000129320.1	2375	No protein	Retained intron	-	-	TSL:2
Rassf1-208	ENSMUST000000144129.1	2094	No protein	Retained intron	-	-	TSL:2
Rassf1-209	ENSMUST000000156198.7	1641	84aa	Nonsense mediated decay	-	D6REH3	TSL:5
Rassf1-210	ENSMUST000000191832.5	479	No protein	lncRNA	-	-	TSL:3

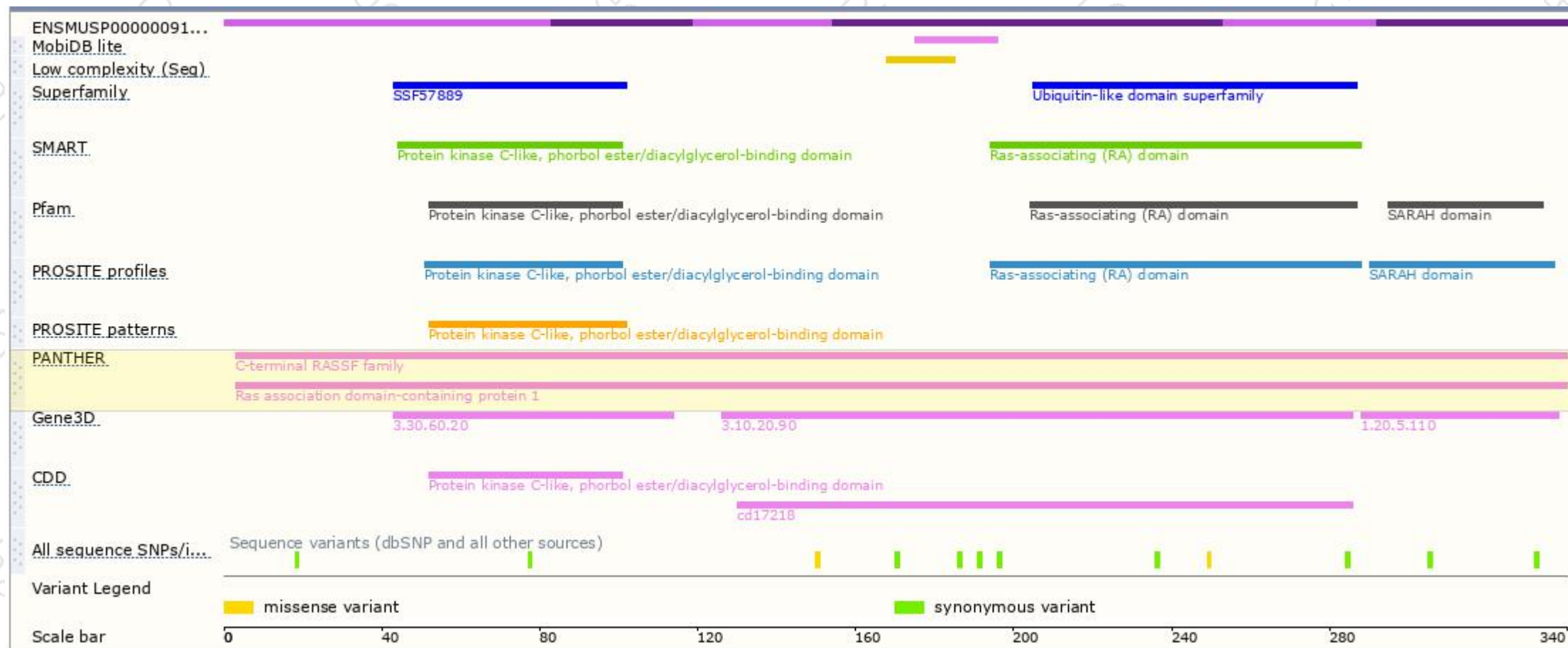
The strategy is based on the design of *Rassf1-202* transcript,The transcription is shown below



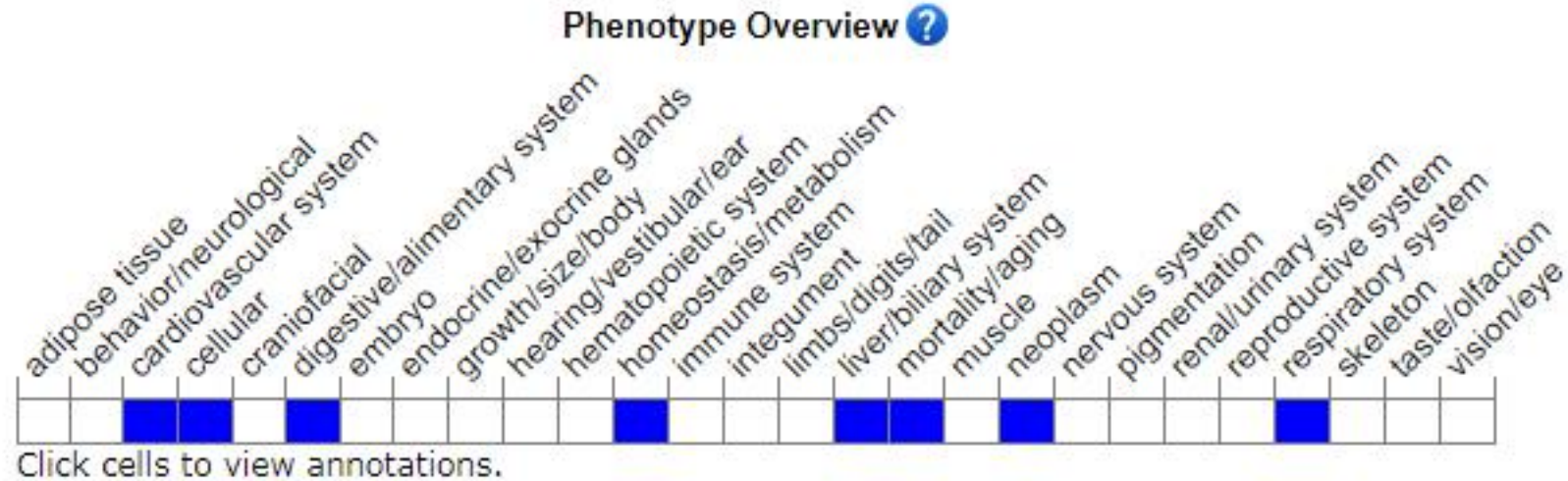
Genomic location distribution



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, Homozygous and heterozygous null mice display increased tumor incidence, especially of lung adenomas and lymphomas, and increased sensitivity to chemically induced tumors.

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

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