

# ***Ksr2* Cas9-CKO Strategy**

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Design Date: 2019-9-11  
Reviewer: Jia Yu

# Project Overview

**Project Name**

***Ksr2***

**Project type**

**Cas9-CKO**

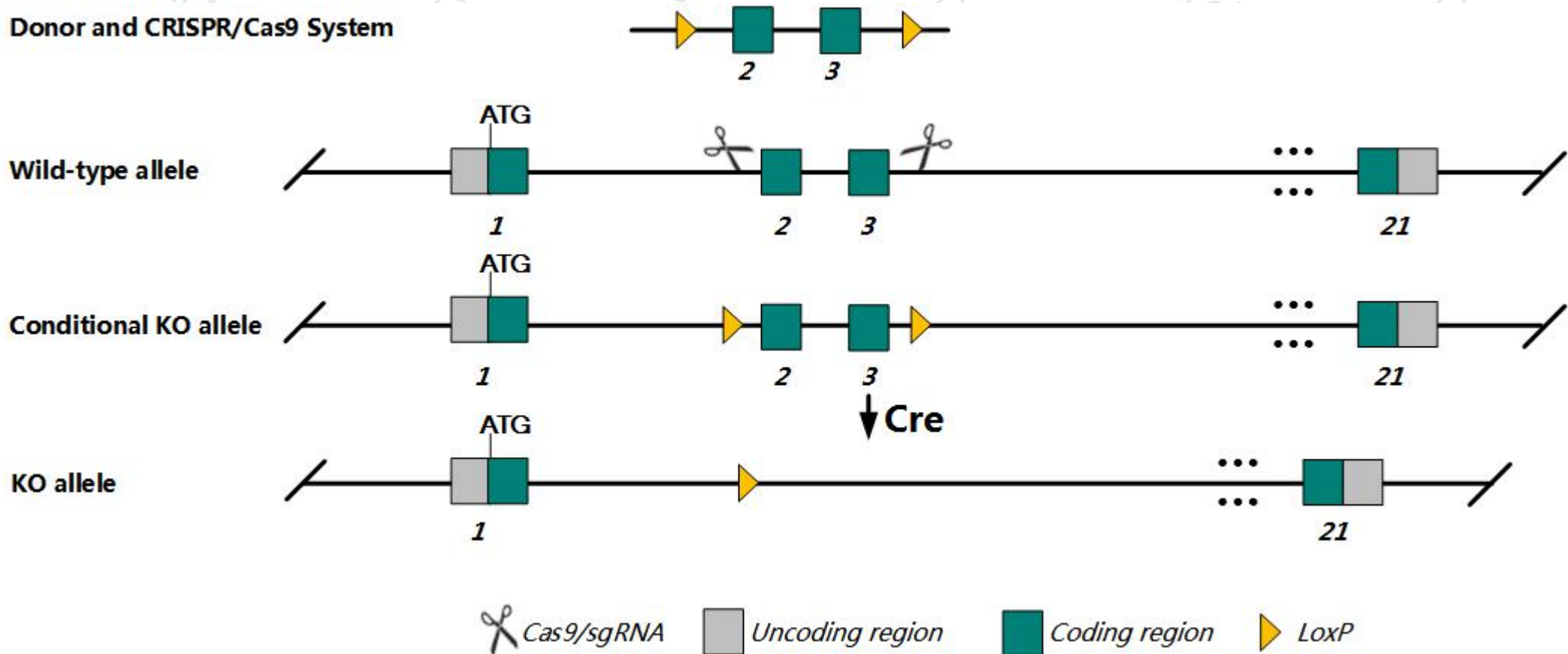
**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ksr2* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System



- The *Ksr2* gene has 2 transcripts. According to the structure of *Ksr2* gene, exon2-3 of *Ksr2-201* (ENSMUST00000180430.1) transcript is recommended as the knockout region. The region contains 292bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ksr2* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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 mice exhibit increased body fat and obesity, resulting from hyperphagia. Mice are also glucose intolerant and have high serum cholesterol, ALT, serum lipids and show hepatic steatosis.
- The *Ksr2* gene is located on the Chr5. 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)

## Ksr2 kinase suppressor of ras 2 [ *Mus musculus* (house mouse) ]

Gene ID: 333050, updated on 12-Aug-2019

### Summary

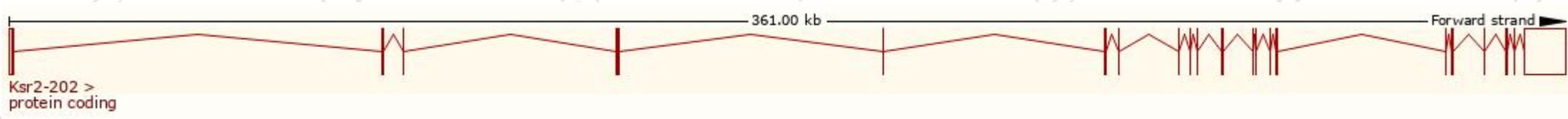
Official Symbol	Ksr2 provided by <a href="#">MGI</a>
Official Full Name	kinase suppressor of ras 2 provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:3610315</a>
See related	<a href="#">Ensembl:ENSMUSG000000061578</a>
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Biased expression in cerebellum adult (RPKM 2.9), frontal lobe adult (RPKM 2.1) and 10 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

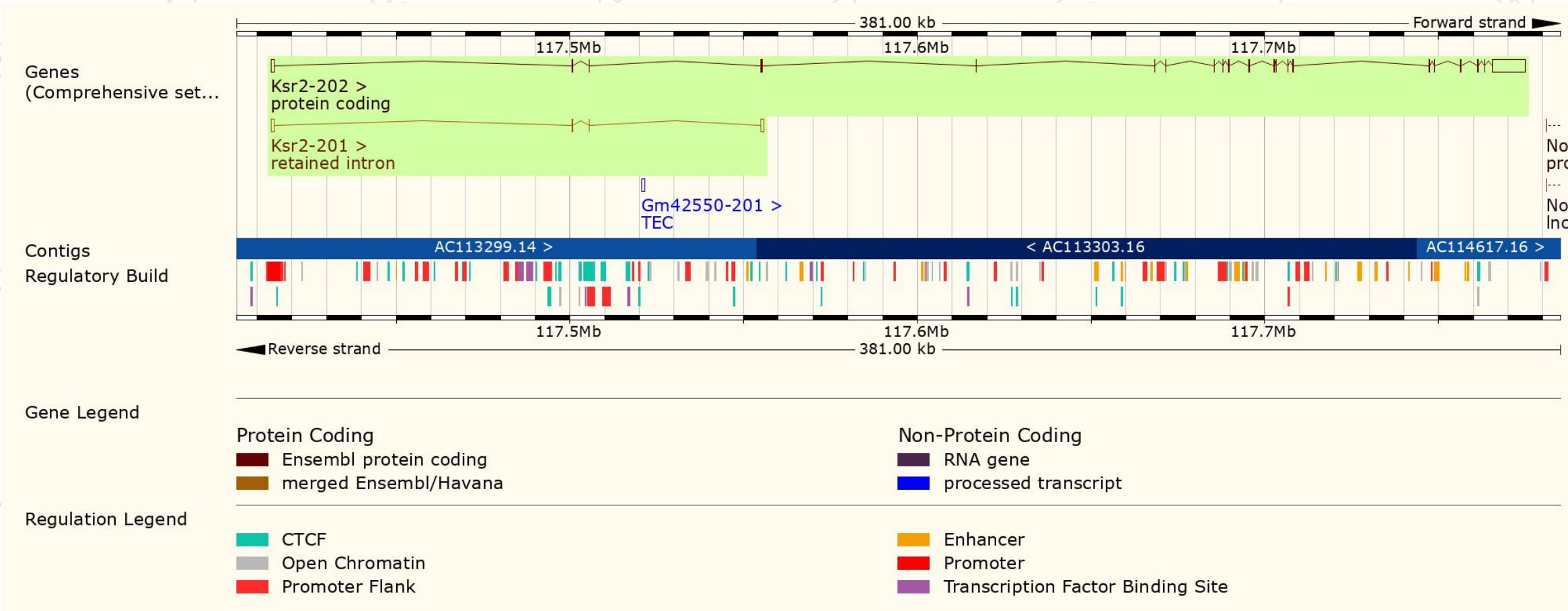
The gene has 2 transcripts,all transcripts are shown below:

Ksr2-201	<a href="#">ENSMUST00000073347.4</a>	2283	No protein	Retained intron	-	-	TSL:1
Ksr2-202	<a href="#">ENSMUST00000180430.1</a>	13125	<a href="#">951aa</a>	Protein coding	-	<a href="#">MQQW59</a>	TSL:5 GENCODE basic APPRIS P1

The strategy is based on the design of *Ksr2-201* transcript,The transcription is shown below

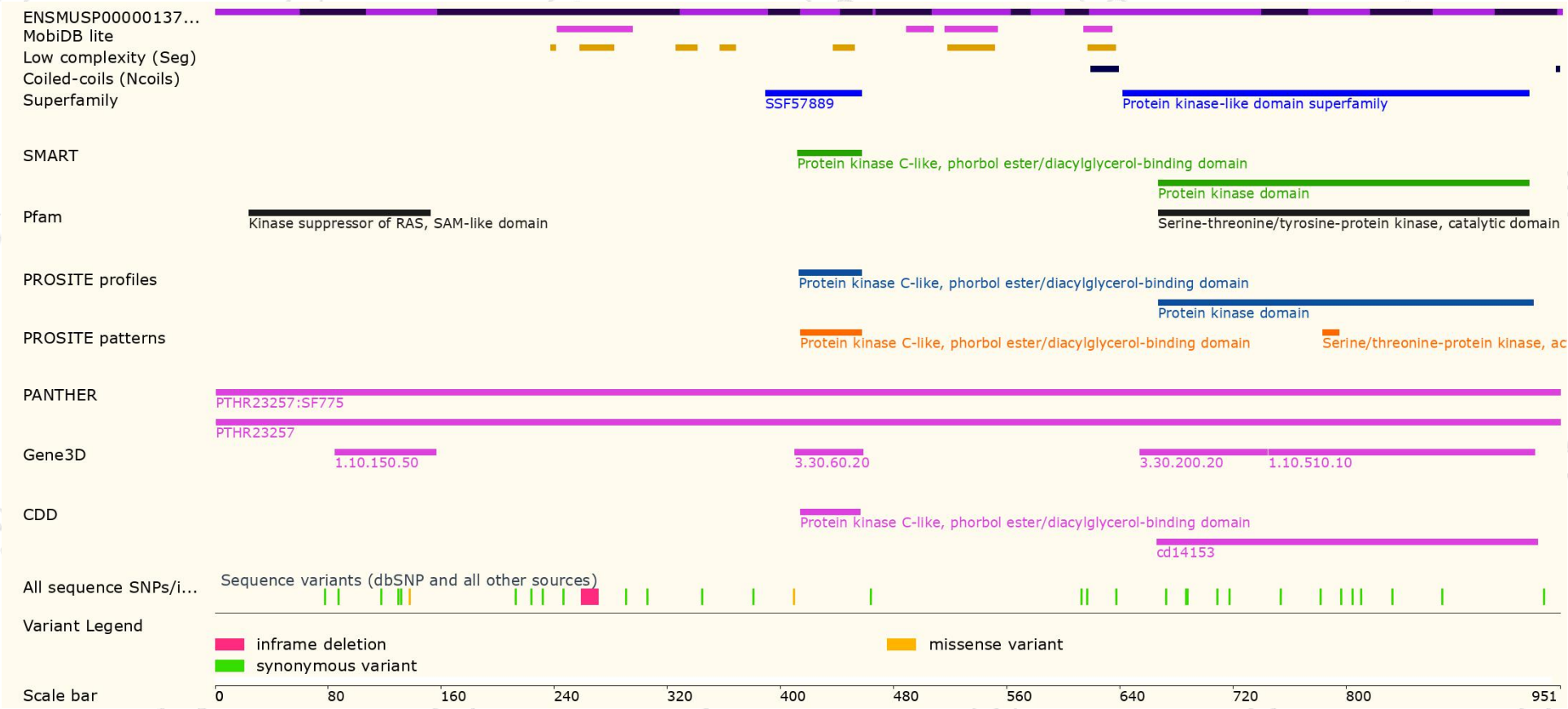


# Genomic location distribution

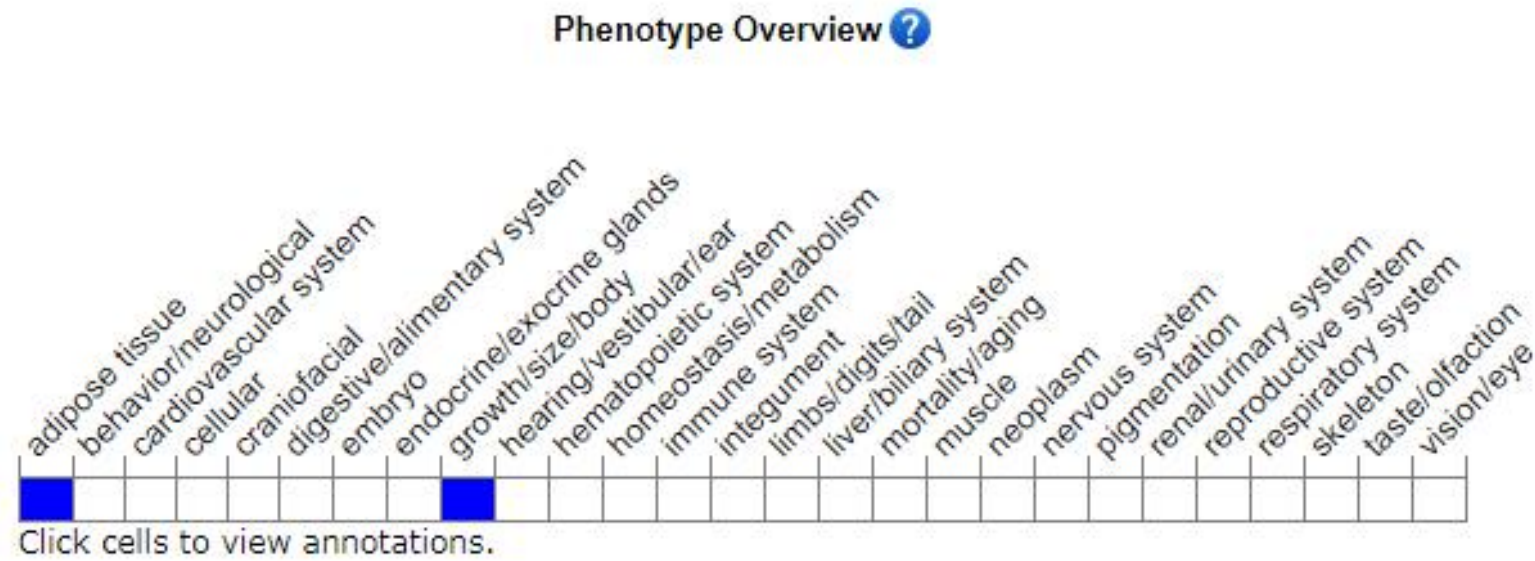




# Protein domain



# Mouse phenotype description(MGI)



*Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).*

Homozygous mice exhibit increased body fat and obesity, resulting from hyperphagia. Mice are also glucose intolerant and have high serum cholesterol, ALT, serum lipids and show hepatic steatosis.

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

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