

Psme4 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Psme4

Project type

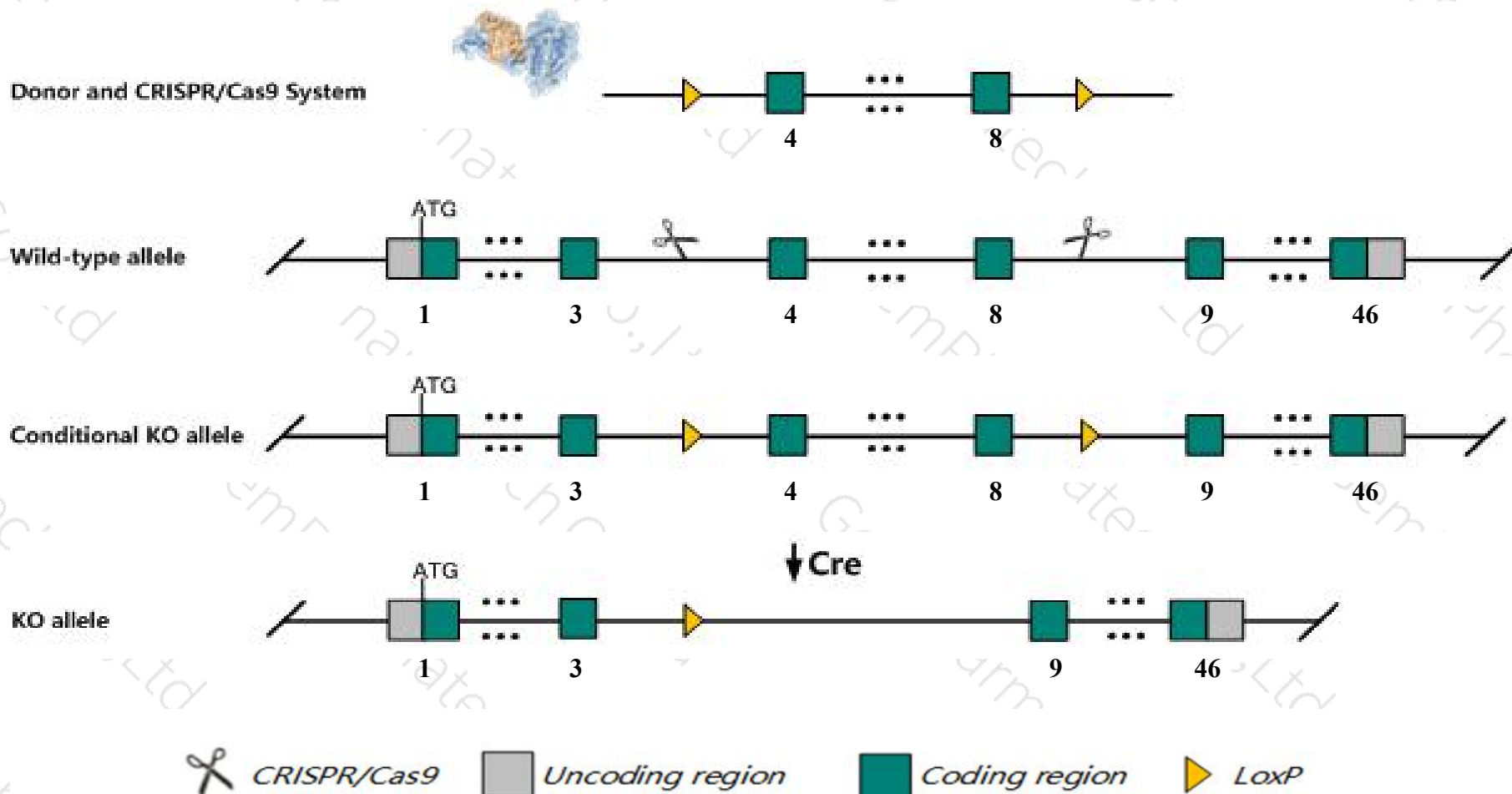
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Psme4* gene. The schematic diagram is as follows:



- The *Psme4* gene has 7 transcripts. According to the structure of *Psme4* gene, exon4-exon8 of *Psme4-201*(ENSMUST00000041231.13) transcript is recommended as the knockout region. The region contains 457bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Psme4* 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-out allele show normal repair of DNA double-strand breaks but exhibit significantly reduced male fertility due to defects in spermatogenesis observed in both meiotic spermatocytes and postmeiotic haploid spermatids.
- Transcript *Psme4-202* may not be affected.
- The *Psme4* gene is located on the Chr11. 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)

Psme4 proteasome (prosome, macropain) activator subunit 4 [*Mus musculus* (house mouse)]

Gene ID: 103554, updated on 26-Jun-2020

Summary

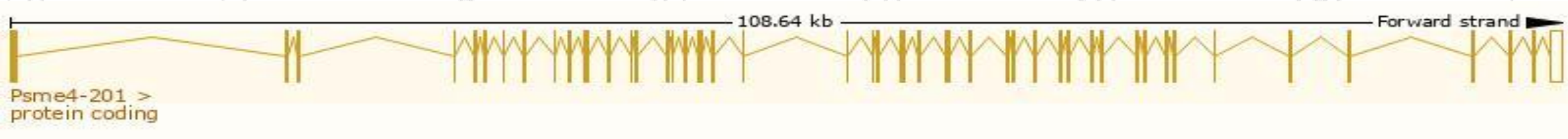
Official Symbol	Psme4 provided by MGI
Official Full Name	proteasome (prosome, macropain) activator subunit 4 provided by MGI
Primary source	MGI:MGI:2143994
See related	Ensembl:ENSMUSG000000040850
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	TEMO; AA409398; AU041366; mKIAA0077
Expression	Ubiquitous expression in testis adult (RPKM 36.4), heart adult (RPKM 19.6) and 23 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

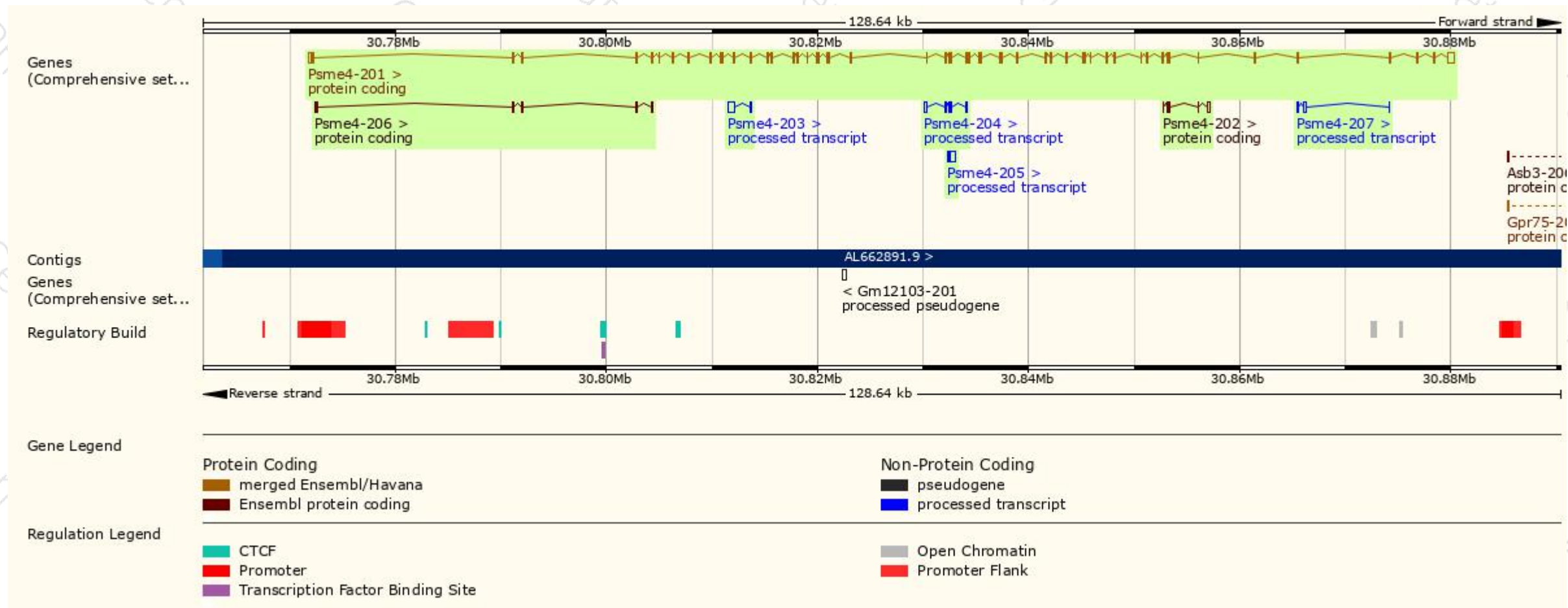
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Psme4-201	ENSMUST00000041231.13	6491	1843aa	Protein coding	CCDS36124	Q5SSW2	TSL:1 Gencode basic APPRIS P1
Psme4-206	ENSMUST00000154757.1	657	219aa	Protein coding	-	F6R7K9	CDS 5' and 3' incomplete TSL:3
Psme4-202	ENSMUST00000129824.1	576	107aa	Protein coding	-	Q5SSW0	CDS 5' incomplete TSL:3
Psme4-203	ENSMUST00000133430.1	695	No protein	Processed transcript	-	-	TSL:3
Psme4-205	ENSMUST00000150219.1	582	No protein	Processed transcript	-	-	TSL:2
Psme4-204	ENSMUST00000142217.1	579	No protein	Processed transcript	-	-	TSL:3
Psme4-207	ENSMUST00000155719.1	452	No protein	Processed transcript	-	-	TSL:5

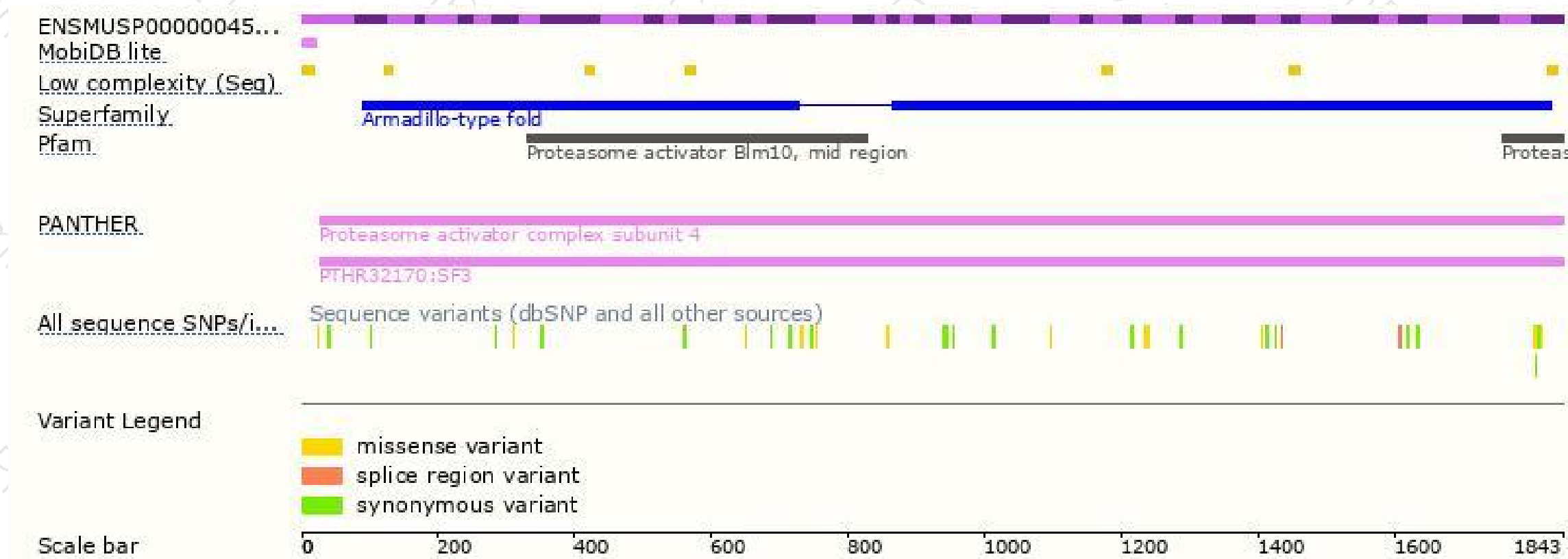
The strategy is based on the design of *Psme4-201* transcript,the transcription is shown below:



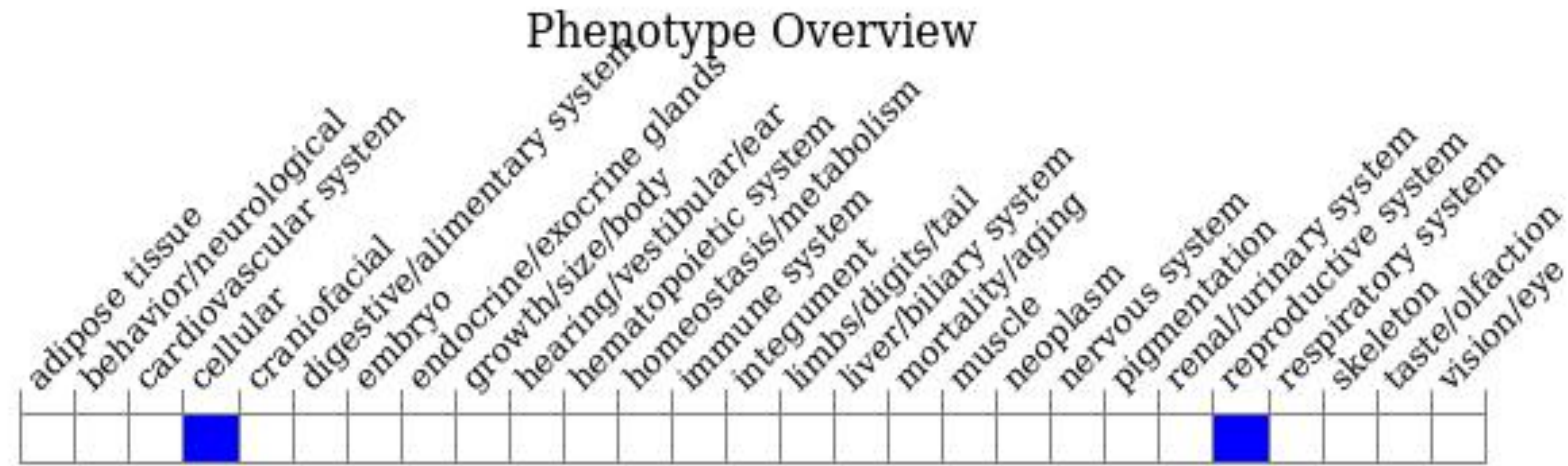
Genomic location distribution



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-out allele show normal repair of DNA double-strand breaks but exhibit significantly reduced male fertility due to defects in spermatogenesis observed in both meiotic spermatocytes and postmeiotic haploid spermatids.

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

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