

Arhgef28 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Arhgef28

Project type

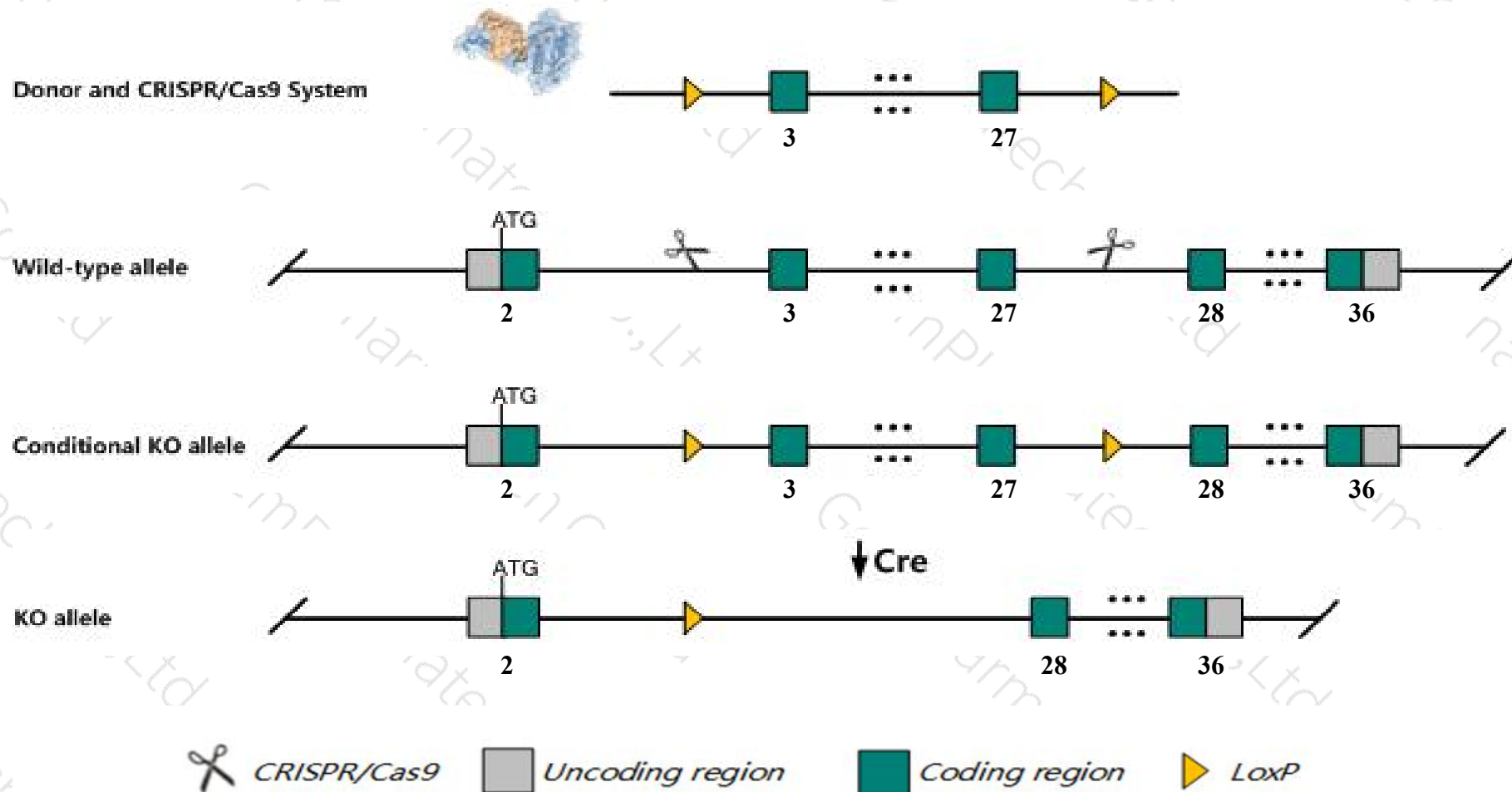
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Arhgef28* gene. The schematic diagram is as follows:



Technical routes

- The *Arhgef28* gene has 7 transcripts. According to the structure of *Arhgef28* gene, exon3-exon27 of *Arhgef28-201* (ENSMUST00000109426.2) transcript is recommended as the knockout region. The region contains 3524bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Arhgef28* 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 are born at lower than expected Mendelian ratios and exhibit a reduction in overall size that becomes negligible by 8 weeks of age. Mouse embryonic fibroblasts display defects in cell migration and focal adhesion formation.
- Transcript 202,203 CDS 3' incomplete the influences is unknown.
- The *Arhgef28* gene is located on the Chr13. 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)

Arhgef28 Rho guanine nucleotide exchange factor (GEF) 28 [*Mus musculus* (house mouse)]

Gene ID: 110596, updated on 13-Mar-2020

Summary

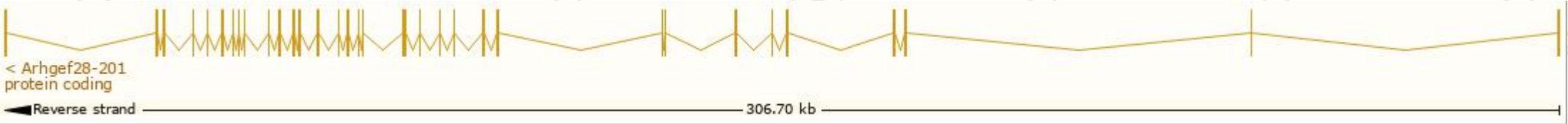
Official Symbol	Arhgef28 provided by MGI
Official Full Name	Rho guanine nucleotide exchange factor (GEF) 28 provided by MGI
Primary source	MGI:MGI:1346016
See related	Ensembl:ENSMUSG000000021662
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	RIP2; Rgnef; RhoGEF; Rhoip2; AI323540; p190RhoGEF; D13Bwg1089e; 9230110L08Rik
Expression	Broad expression in kidney adult (RPKM 5.2), ovary adult (RPKM 5.2) and 22 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

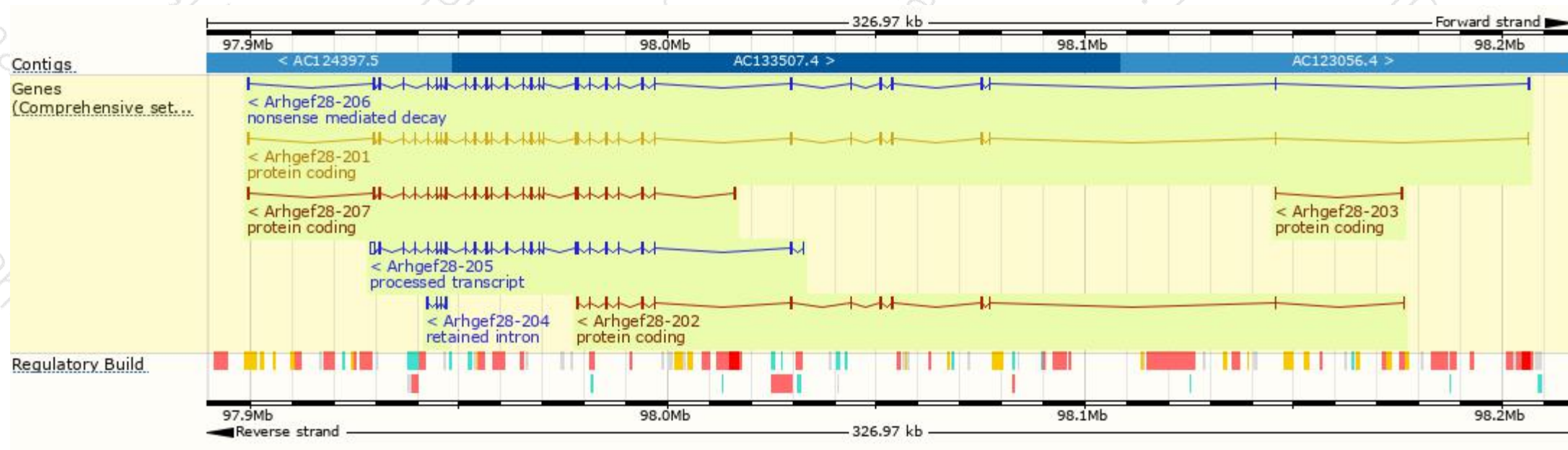
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Arhgef28-201	ENSMUST00000109426.2	5387	1700aa	Protein coding	CCDS36758	G5E8P2	TSL:1 GENCODE basic APPRIS P2
Arhgef28-207	ENSMUST00000238746.1	4660	1387aa	Protein coding	-	-	APPRIS ALT2
Arhgef28-202	ENSMUST00000223849.1	2001	643aa	Protein coding	-	A0A286YCY0	CDS 3' incomplete
Arhgef28-203	ENSMUST00000224866.1	203	11aa	Protein coding	-	A0A286YD71	CDS 3' incomplete
Arhgef28-206	ENSMUST00000225884.1	5307	1324aa	Nonsense mediated decay	-	A0A286YDI4	-
Arhgef28-205	ENSMUST00000225663.1	4977	No protein	Processed transcript	-	-	-
Arhgef28-204	ENSMUST00000225269.1	783	No protein	Retained intron	-	-	-

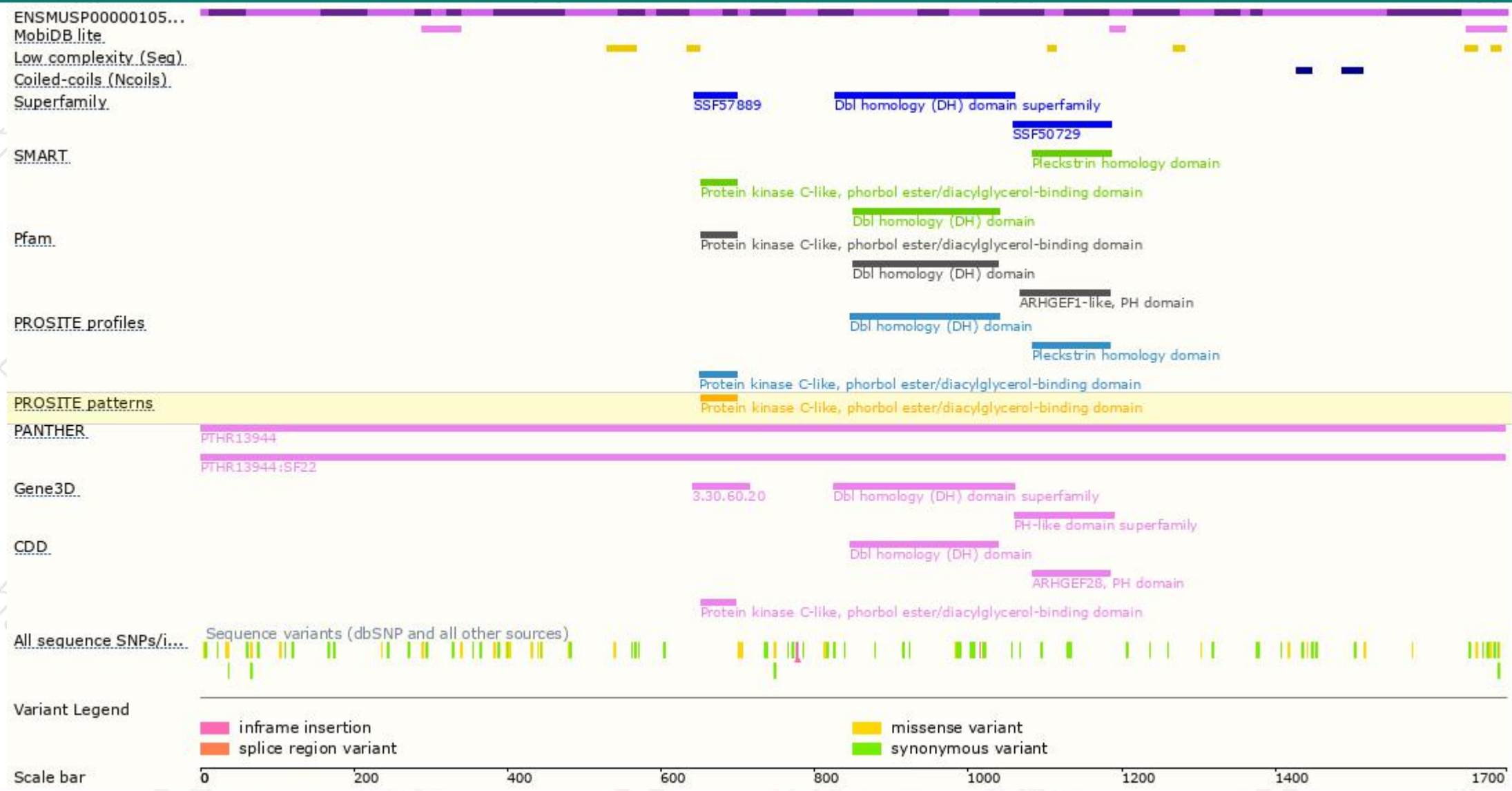
The strategy is based on the design of *Arhgef28-201* transcript,The transcription is shown below



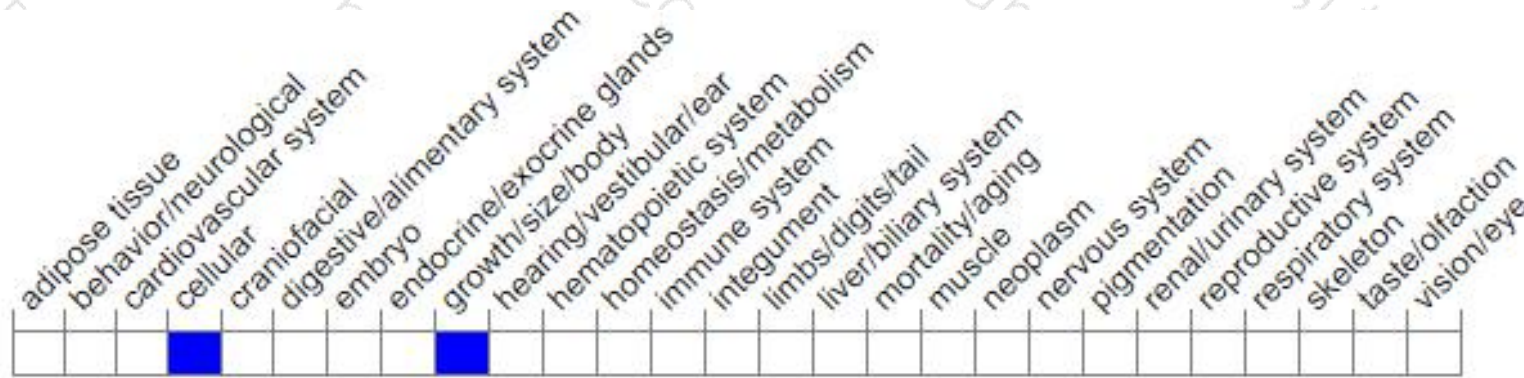
Genomic location distribution



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 are born at lower than expected Mendelian ratios and exhibit a reduction in overall size that becomes negligible by 8 weeks of age. Mouse embryonic fibroblasts display defects in cell migration and focal adhesion formation.

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

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