

Ehd4 Cas9-CKO Strategy

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Project Overview

Project Name

Ehd4

Project type

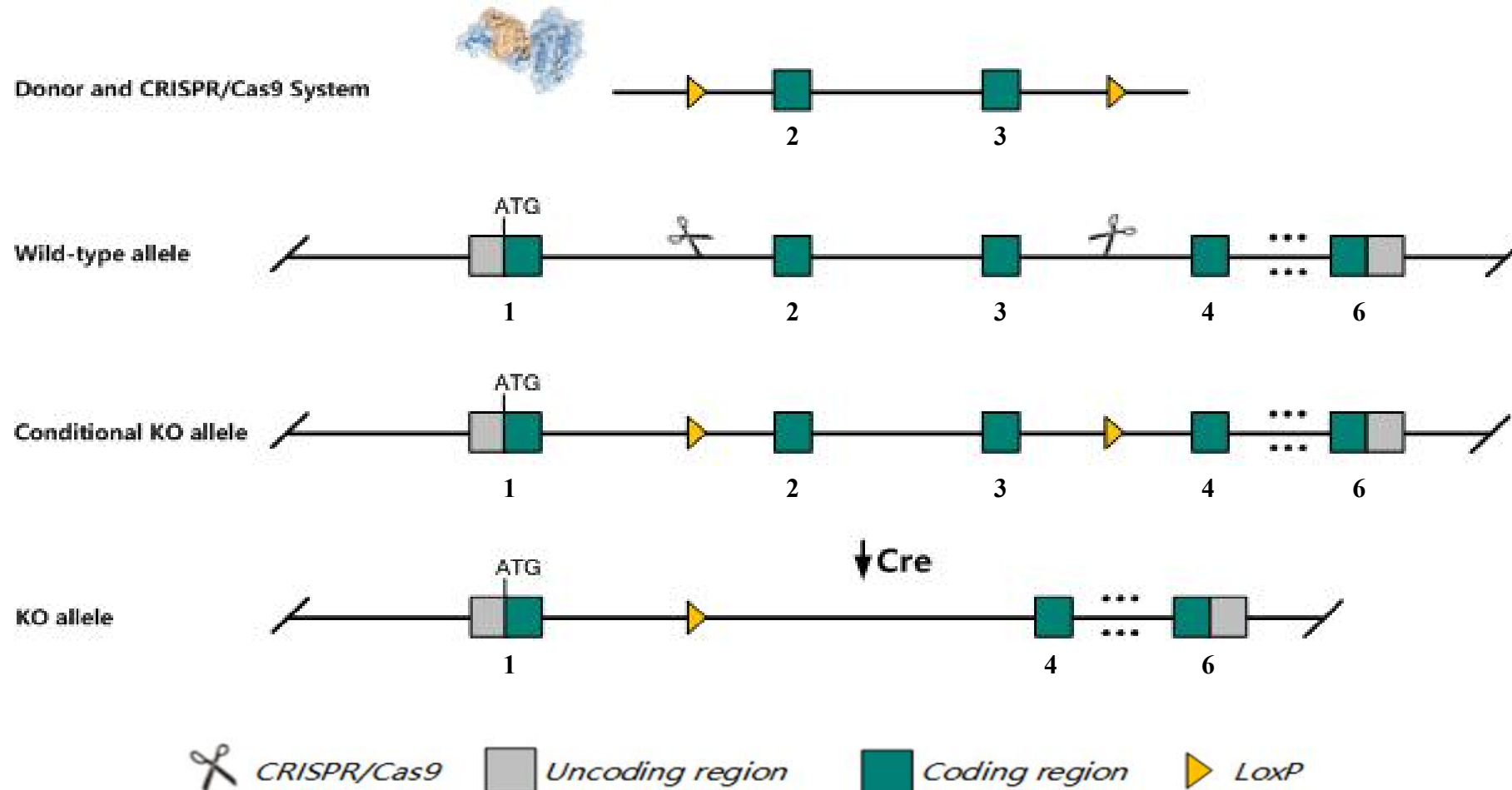
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ehd4* gene. The schematic diagram is as follows:



The *Ehd4* gene has 4 transcripts. According to the structure of *Ehd4* gene, exon2-exon3 of *Ehd4-201*(ENSMUST00000028755.8) transcript is recommended as the knockout region. The region contains 275bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Ehd4* 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 exhibit reduced male fertility.

The *Ehd4* gene is located on the Chr2. 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.

Ehd4 EH-domain containing 4 [Mus musculus (house mouse)]

Gene ID: 98878, updated on 17-Dec-2020

Summary



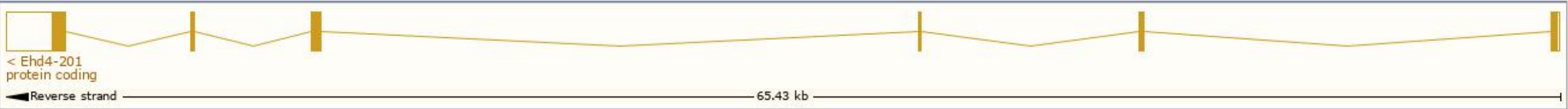
Official Symbol	Ehd4 provided by MGI
Official Full Name	EH-domain containing 4 provided by MGI
Primary source	MGI:MGI:1919619
See related	Ensembl:ENSMUSG00000027293
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2210022F10Rik, AI197390, AI846352, AV006278, Past2
Expression	Broad expression in lung adult (RPKM 115.2), heart adult (RPKM 78.1) and 16 other tissues See more
Orthologs	human all

Transcript information Ensembl

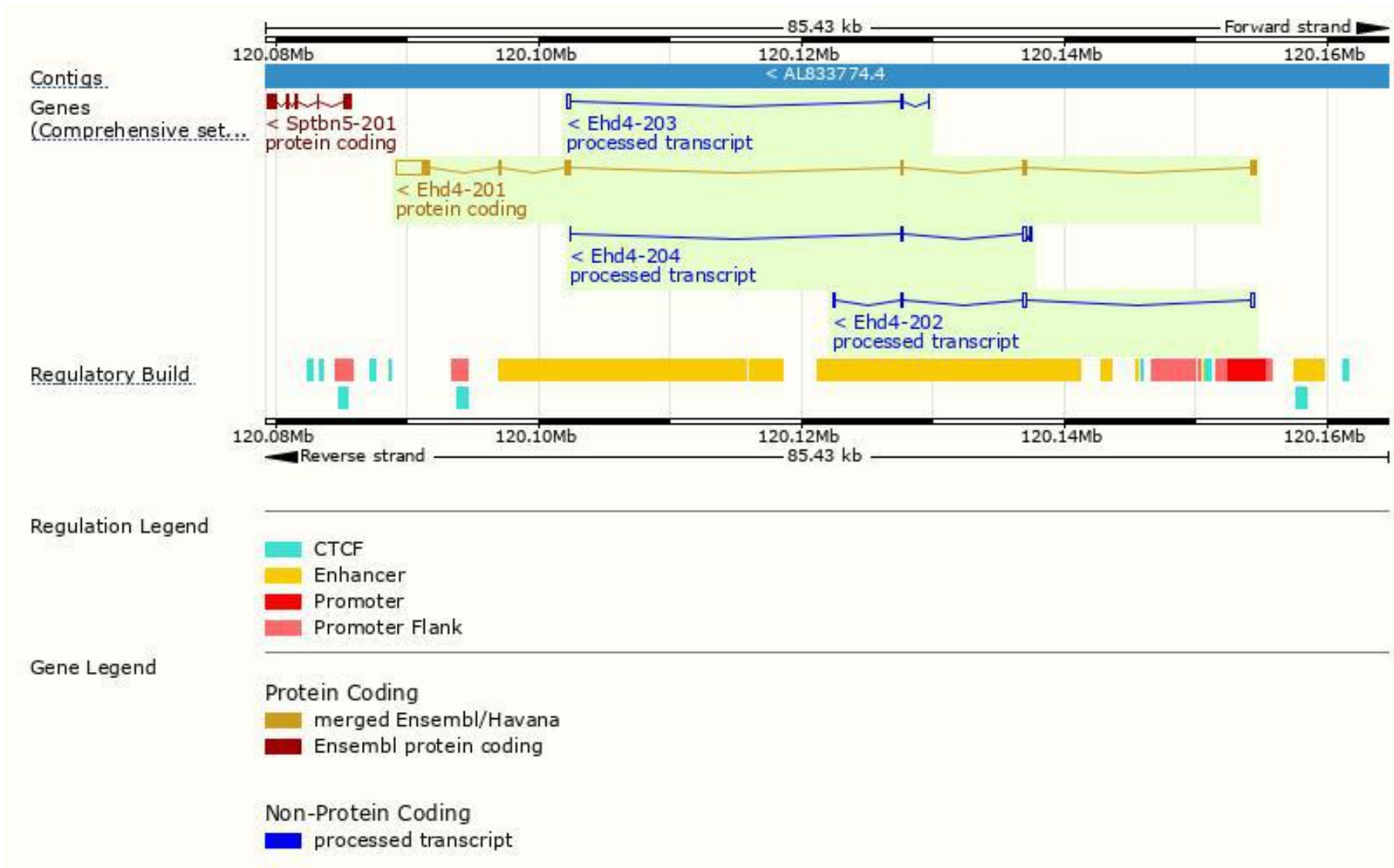
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ehd4-201	ENSMUST00000028755.8	3730	541aa	Protein coding	CCDS16615		TSL:1 , GENCODE basic , APPRIS P1 ,
Ehd4-202	ENSMUST00000148073.2	541	No protein	Processed transcript	-		TSL:3 ,
Ehd4-204	ENSMUST00000152366.8	455	No protein	Processed transcript	-		TSL:5 ,
Ehd4-203	ENSMUST00000150240.8	393	No protein	Processed transcript	-		TSL:3 ,

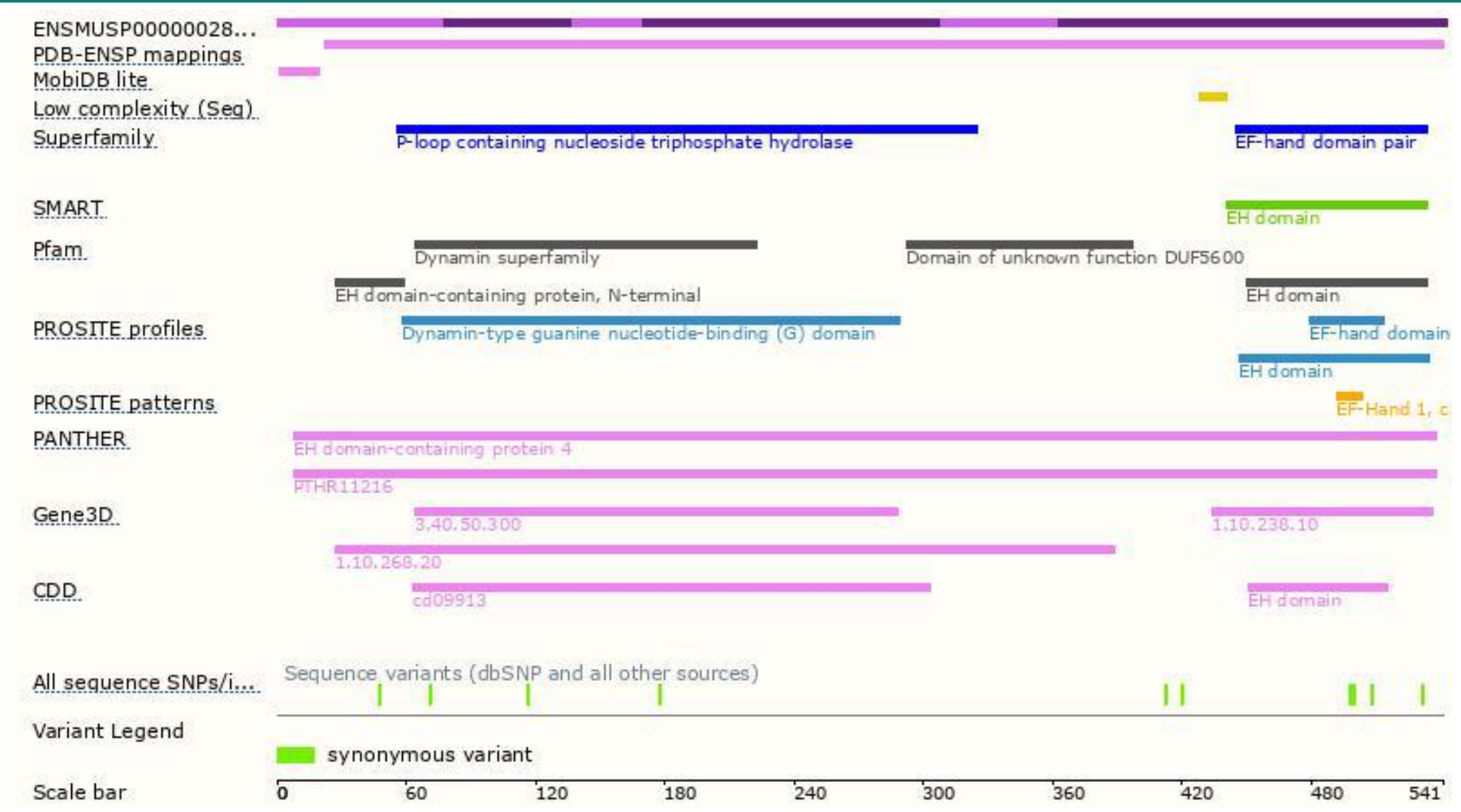
The strategy is based on the design of *Ehd4-201* transcript,the transcription is shown below:



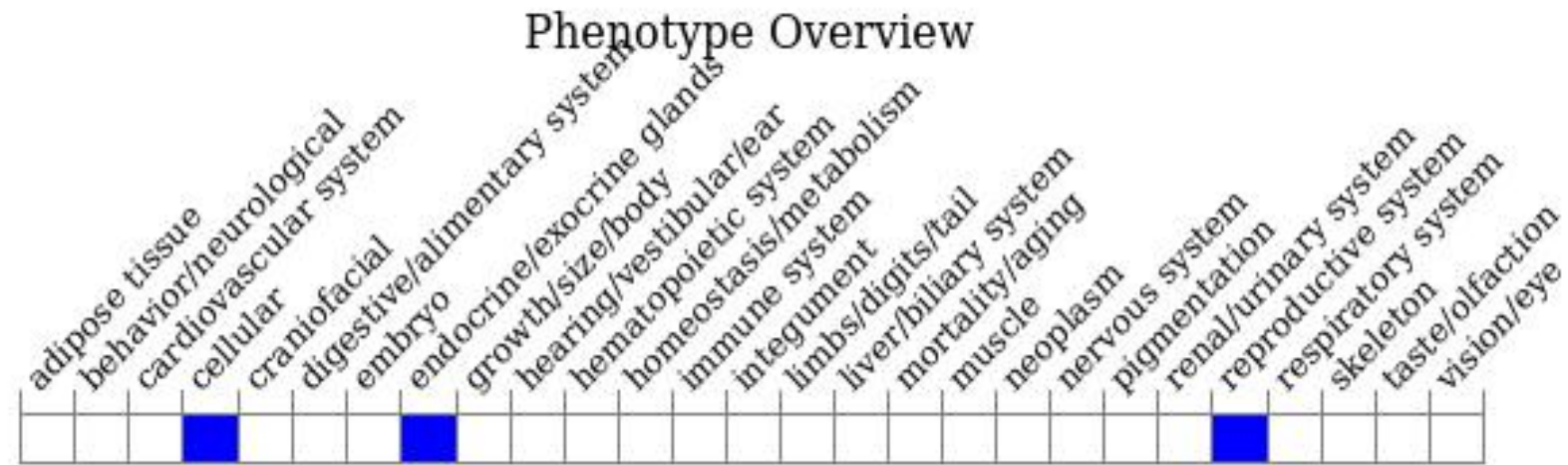
Genomic location distribution



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 exhibit reduced male fertility.

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