Creb3l4 Cas9-CKO Strategy

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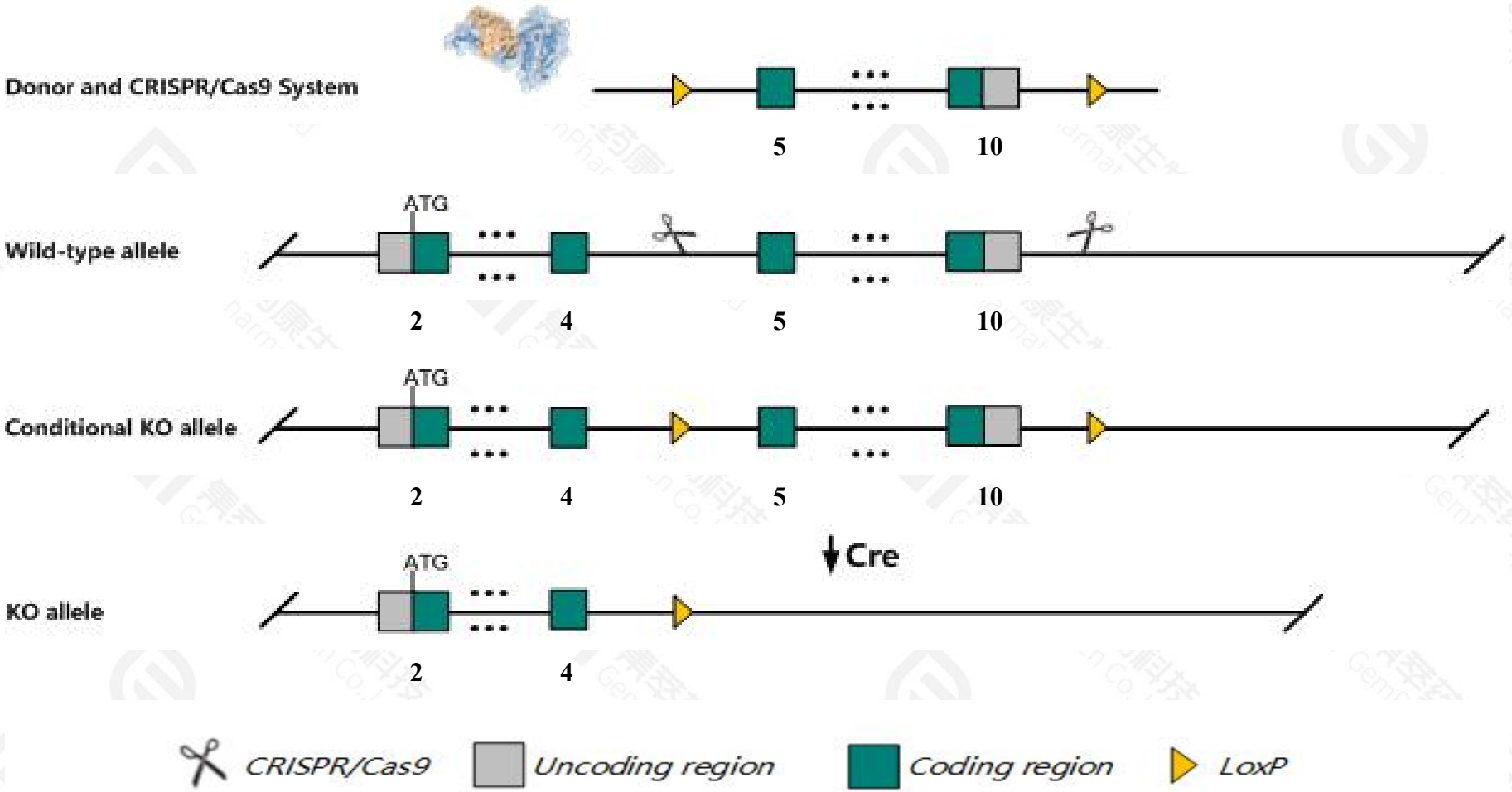
Project Overview

Project Name	***Creb3l4***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Creb3l4* gene. The schematic diagram is as follows:



Technical routes

➢ The *Creb3l4* gene has 2 transcripts. According to the structure of *Creb3l4* gene, exon5-exon10 of *Creb3l4-201*(ENSMUST00000029547.10) transcript is recommended as the knockout region.The region contains translation stop codon sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Creb3l4* 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.

Notice

➢ According to the existing MGI data,homozygous null mice display oligozoospermia but have normal fertility and sperm morphology and motility.

➢ The *Creb3l4* gene is located on the Chr3. 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）

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Creb3l4 cAMP responsive element binding protein 3-like 4 [*Mus musculus* (house mouse)]

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Gene ID: 78284, updated on 16-Oct-2021

Summary

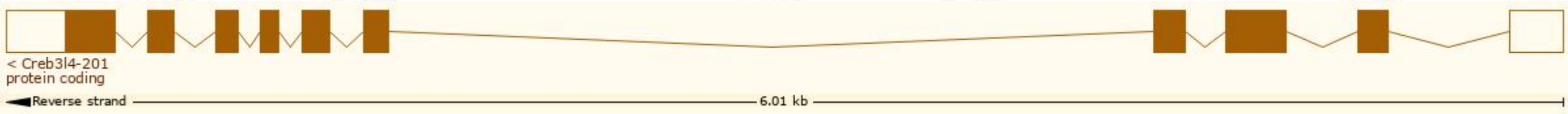
Official Symbol	Creb3l4 provided by MGI
Official Full Name	cAMP responsive element binding protein 3-like 4 provided by MGI
Primary source	MGI:MGI:1916603
See related	Ensembl:ENSMUSG00000027938
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	JAL; CREB4; acre1; AIBZIP; TISP40; AV040530; AV258827; 1700012K17Rik; 5330432F22Rik
Summary	This gene encodes a CREB (cyclic AMP-responsive element-binding) protein with a transmembrane domain which localizes it to the ER membrane. The encoded protein may play a role in adiposity and male germ cell development. Homozygous knockout mice for this gene show increased adipogenesis, elevated testicular germ cell apoptosis and defects in spermatogenesis. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Apr 2015]
Expression	Biased expression in testis adult (RPKM 127.4), colon adult (RPKM 11.4) and 1 other tissue See more
Orthologs	human all

Transcript information（Ensembl）

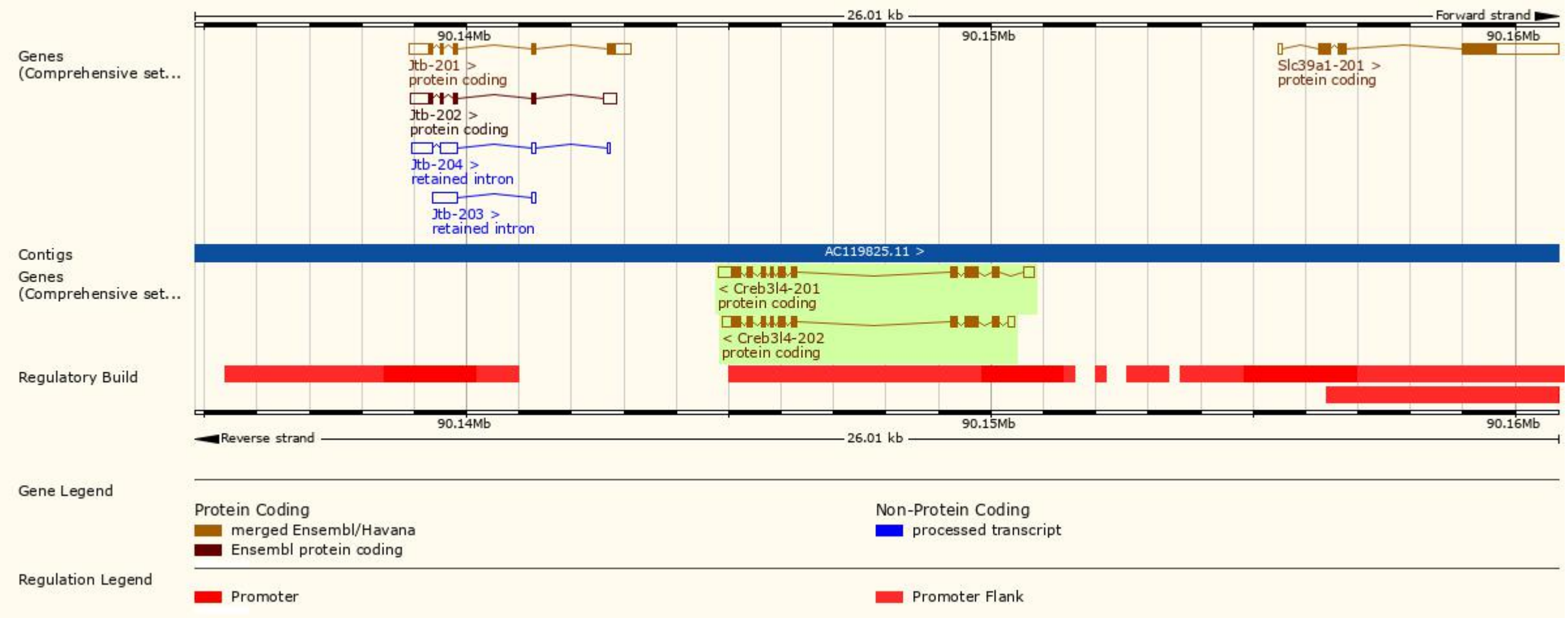
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Creb3l4-201	ENSMUST00000029547.10	1553	370aa	Protein coding	CCDS17524		TSL:1 , GENCODE basic , APPRIS P1 ,
Creb3l4-202	ENSMUST00000107369.2	1404	370aa	Protein coding	CCDS17524		TSL:1 , GENCODE basic , APPRIS P1 ,

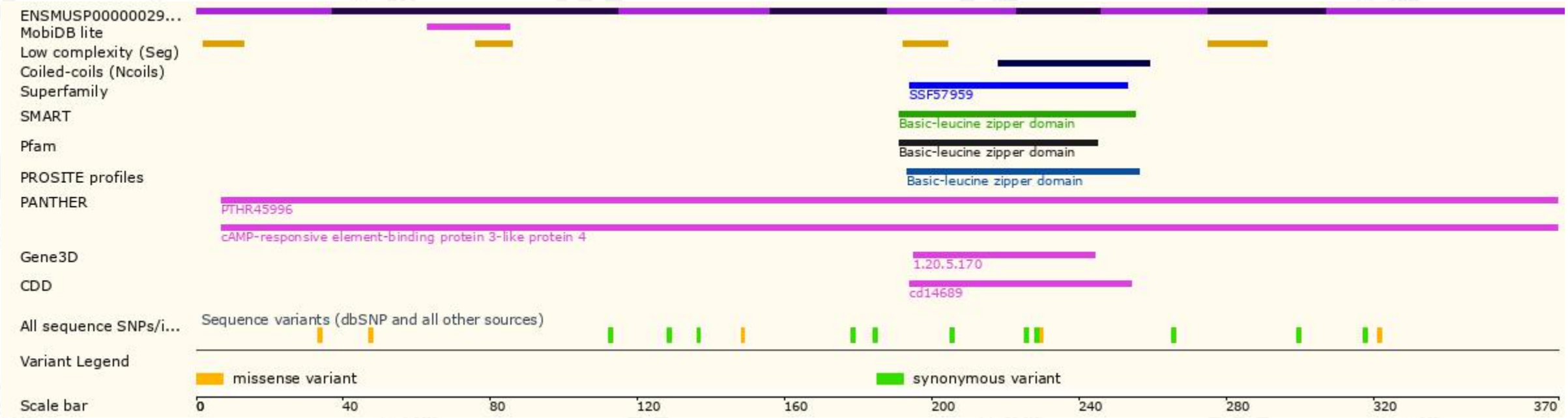
The strategy is based on the design of *Creb3l4-201* transcript,the transcription is shown below:



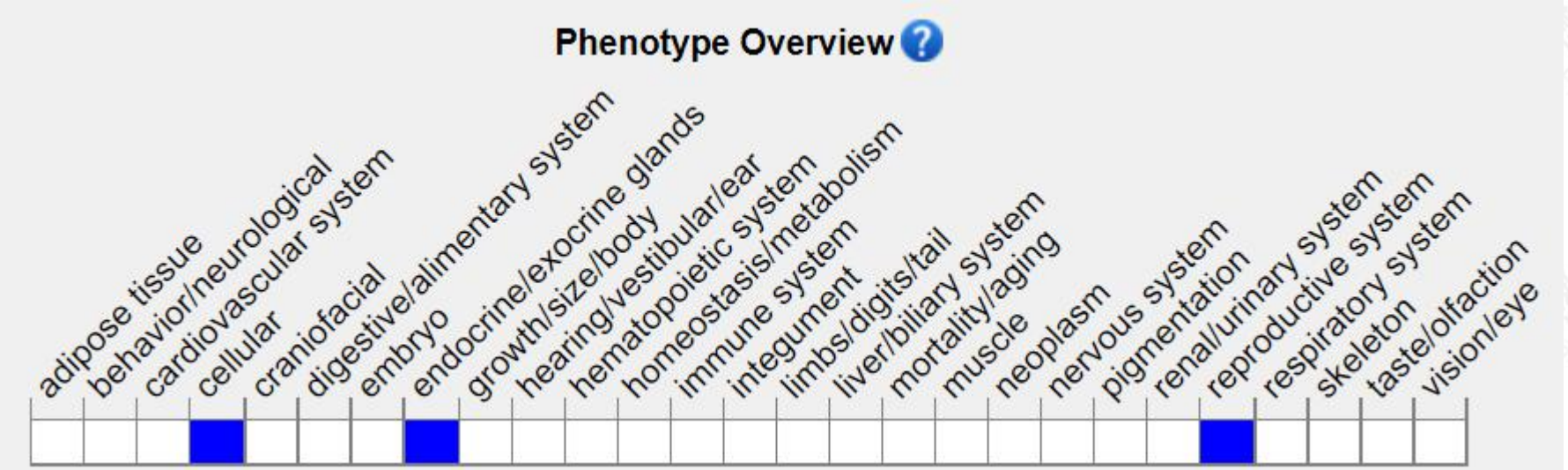
Genomic location distribution



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,homozygous null mice display oligozoospermia but have normal fertility and sperm morphology and motility.

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