

Robo2 Cas9-CKO Strategy

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Project Overview

Project Name

Robo2

Project type

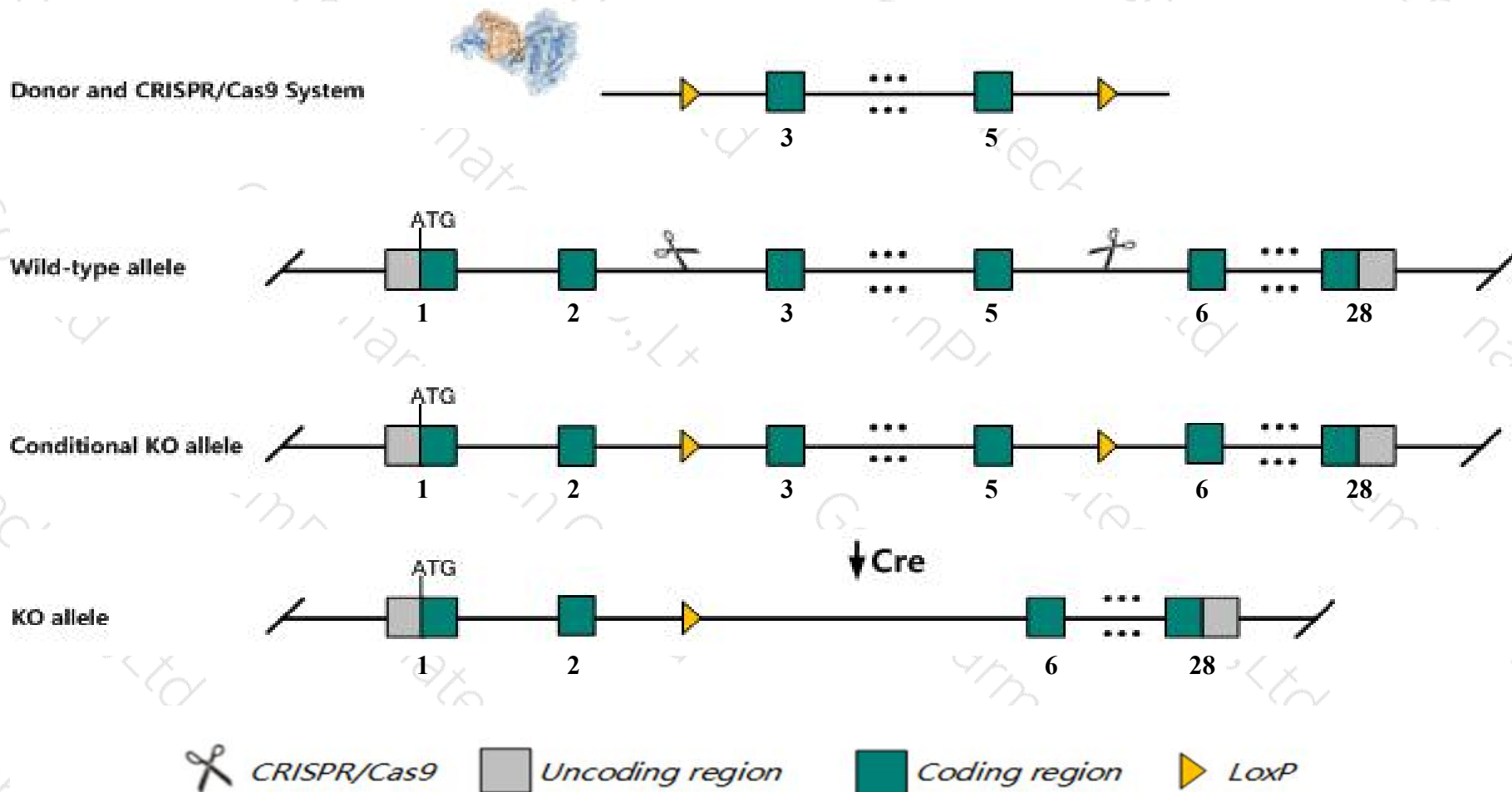
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Robo2* gene. The schematic diagram is as follows:



- The *Robo2* gene has 13 transcripts. According to the structure of *Robo2* gene, exon3-exon5 of *Robo2*-203 (ENSMUST00000117785.8) transcript is recommended as the knockout region. The region contains 418bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Robo2* 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, Homozygous mutants display postnatal lethality, abnormal ureteric bud development, multiple fused kidneys, multiple ureters, and abnormal commissural axon growth.
- Transcripts 207,208,209 may not be affected.
- The effect of transcripts 201,204,205,212,213 is unknown.
- The *Robo2* gene is located on the Chr16. 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)

Robo2 roundabout guidance receptor 2 [*Mus musculus* (house mouse)]

Gene ID: 268902, updated on 28-Jan-2020

Summary



Official Symbol Robo2 provided by [MGI](#)

Official Full Name roundabout guidance receptor 2 provided by [MGI](#)

Primary source [MGI:MGI:1890110](#)

See related [Ensembl:ENSMUSG00000052516](#)

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 BB097918; mKIAA1568; 2600013A04Rik; 9430089E08Rik; D230004I22Rik

Expression Biased expression in CNS E18 (RPKM 11.3), whole brain E14.5 (RPKM 10.4) and 12 other tissues [See more](#)

Orthologs [human](#) [all](#)

Genomic context



Location: 16; 16 C3.1

Exon count: 34

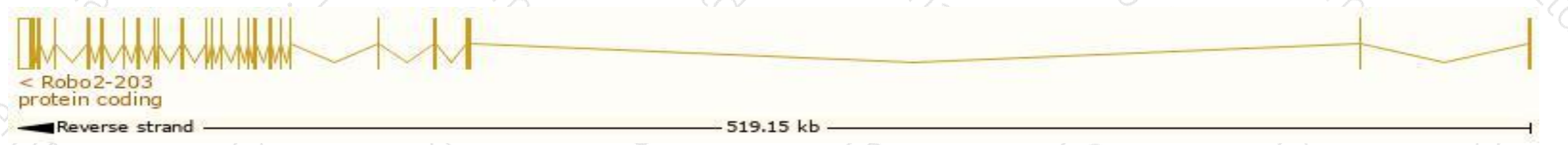
See Robo2 in [Genome Data Viewer](#)

Transcript information (Ensembl)

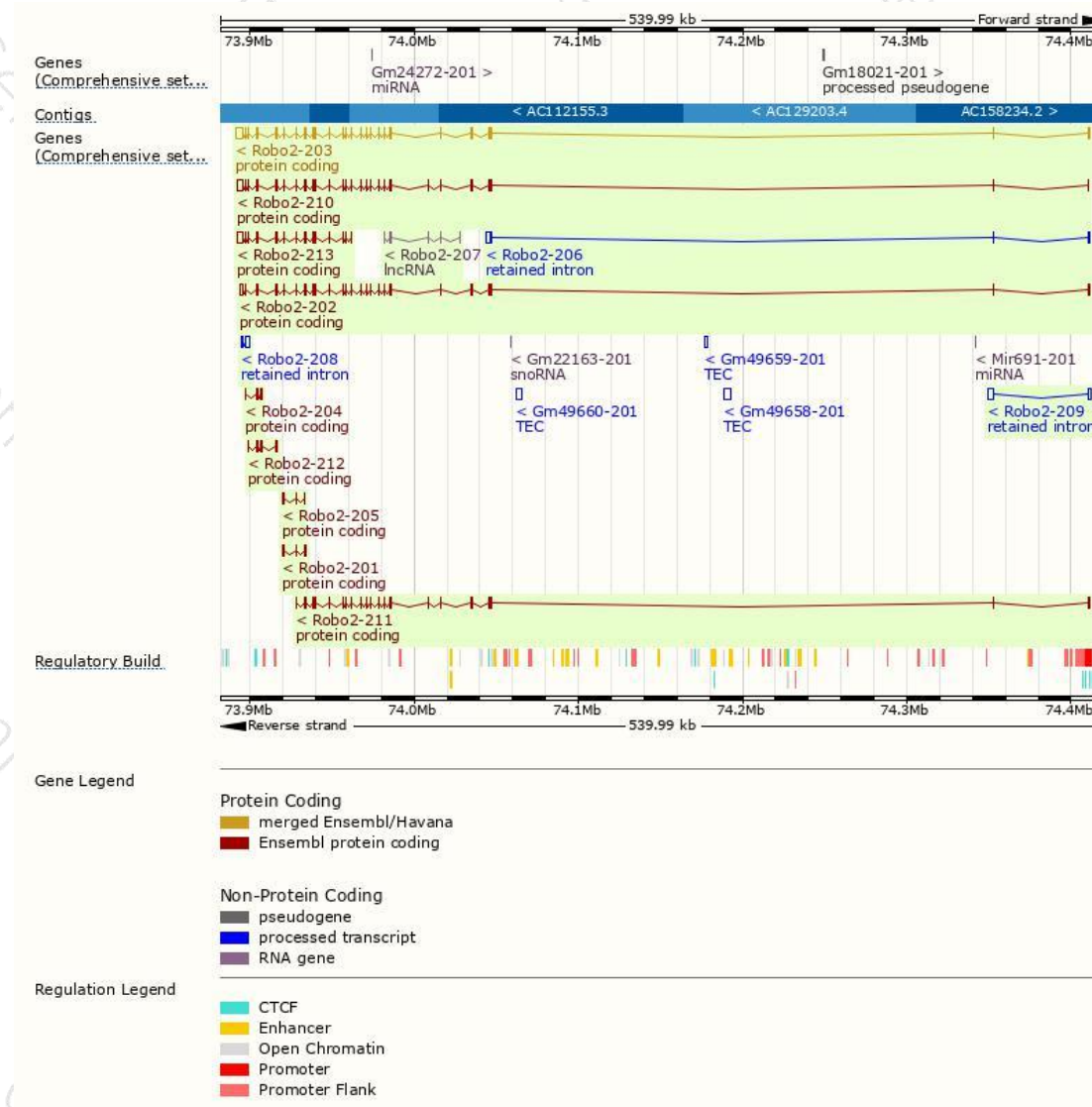
The gene has 13 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Robo2-203	ENSMUST00000117785.8	8591	1508aa	Protein coding	CCDS49886	Q19AB2	TSL:1 GENCODE basic
Robo2-210	ENSMUST00000226478.1	8059	1512aa	Protein coding	-	A0A2I3BR72	GENCODE basic
Robo2-202	ENSMUST00000117200.7	5988	1377aa	Protein coding	-	E9Q6A0	TSL:5 GENCODE basic APPRIS P1
Robo2-213	ENSMUST00000231889.1	5900	793aa	Protein coding	-	A0A338P6D2	CDS 5' incomplete
Robo2-211	ENSMUST00000227347.1	3818	1065aa	Protein coding	-	A0A2I3BQB9	CDS 3' incomplete
Robo2-201	ENSMUST00000116586.3	886	207aa	Protein coding	-	E9PYK2	CDS 5' incomplete TSL:5
Robo2-205	ENSMUST00000138852.2	774	149aa	Protein coding	-	A0A338P7C8	CDS 5' incomplete TSL:3
Robo2-212	ENSMUST00000231426.1	657	219aa	Protein coding	-	A0A338P6W2	CDS 5' and 3' incomplete
Robo2-204	ENSMUST00000137420.1	535	178aa	Protein coding	-	A0A338P6R3	CDS 5' and 3' incomplete TSL:2
Robo2-207	ENSMUST00000147408.2	430	No protein	Processed transcript	-	-	TSL:5
Robo2-209	ENSMUST00000156010.1	4057	No protein	Retained intron	-	-	TSL:1
Robo2-208	ENSMUST00000149114.1	4046	No protein	Retained intron	-	-	TSL:1
Robo2-206	ENSMUST00000140062.1	2942	No protein	Retained intron	-	-	TSL:1

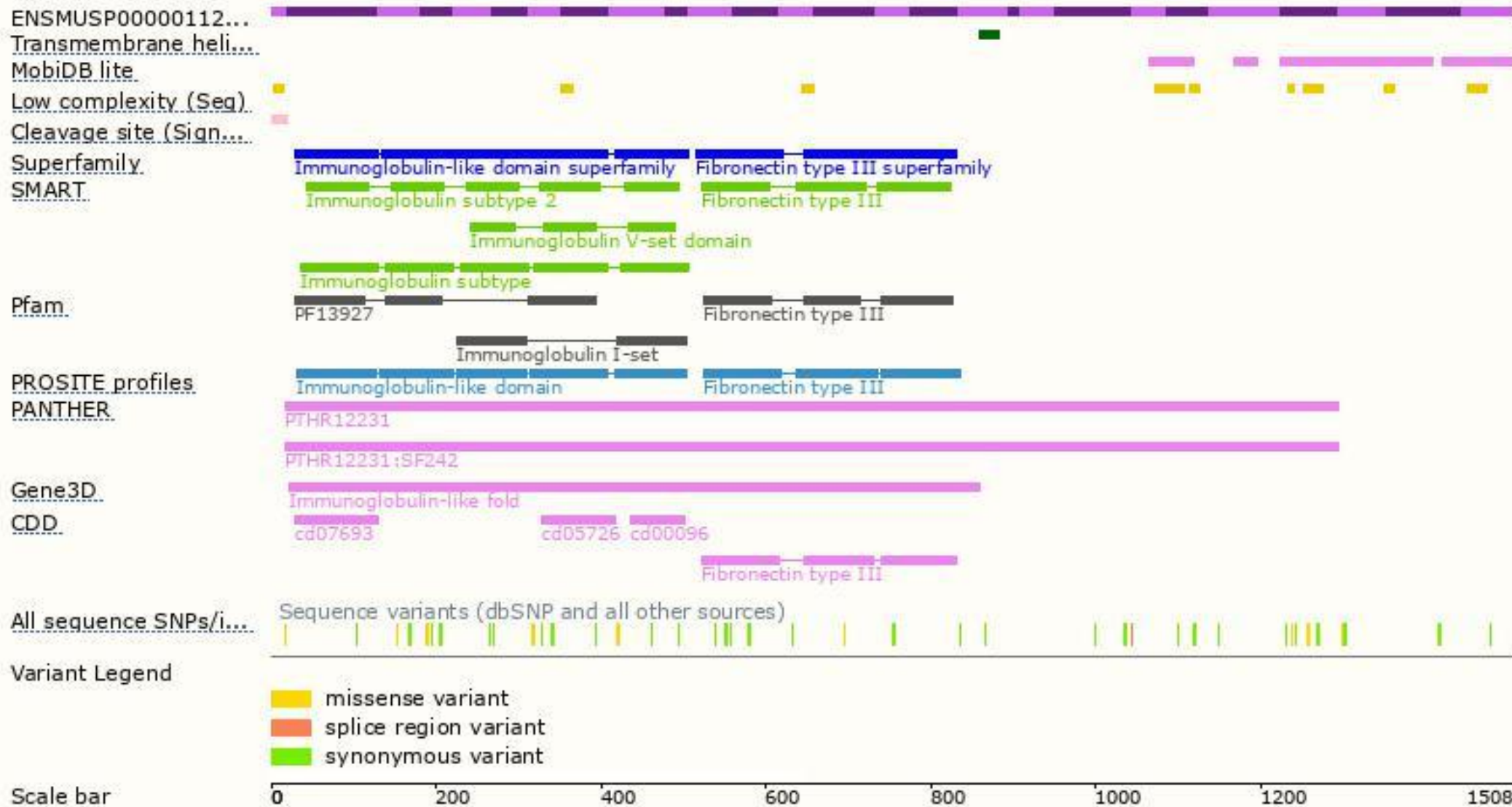
The strategy is based on the design of *Robo2-203* transcript,The transcription is shown below



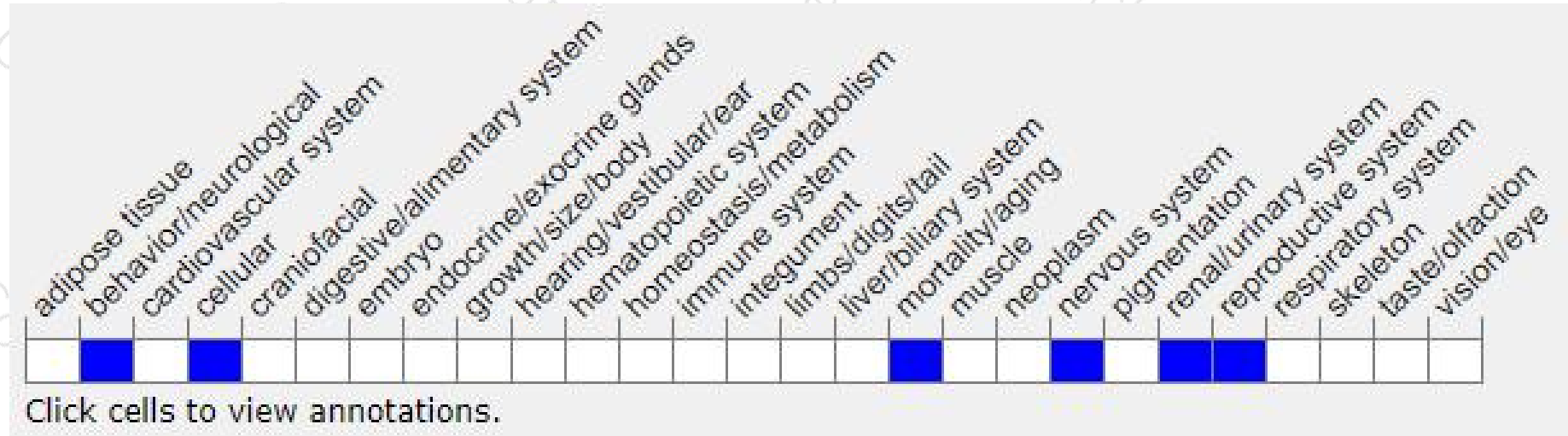
Genomic location distribution



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 mutants display postnatal lethality, abnormal ureteric bud development, multiple fused kidneys, multiple ureters, and abnormal commissural axon growth.

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

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