

Cxcr5 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Cxcr5

Project type

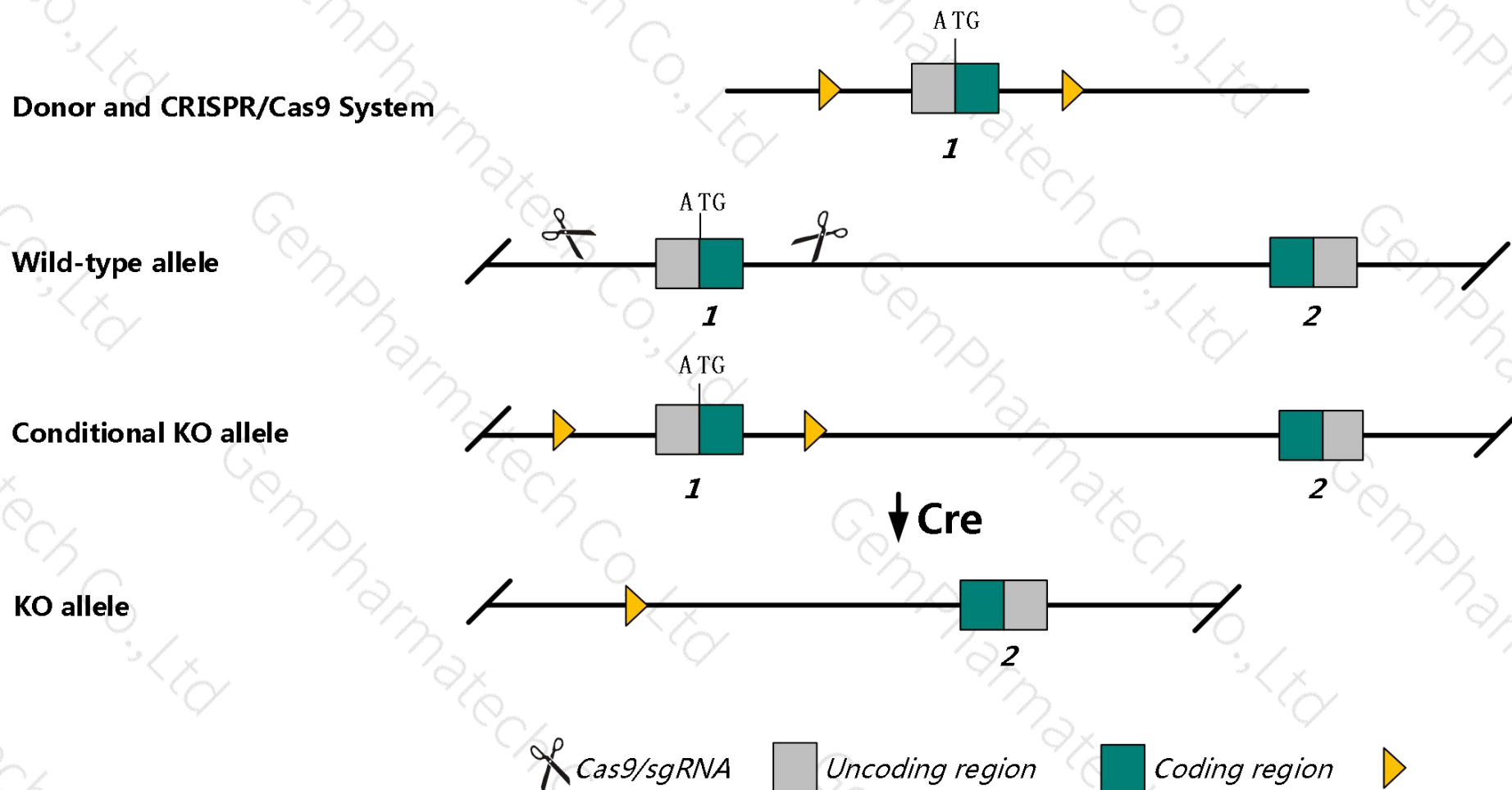
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cxcr5* gene. The schematic diagram is as follows:



- The *Cxcr5* has 5 transcripts. According to the structure of *Cxcr5* gene, exon1 of *Cxcr5*-201 (ENSMUST00000062215.7) transcript is recommended as the knockout region. The region contains 57 bp start codon ATG coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cxcr5* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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 or cell types.

- According to the existing MGI data , homozygous null mutants lack inguinal lymph nodes, have a few abnormal or no Peyer's patches, morphologically altered primary lymphoid follicles and no functional germinal centers in their spleen.
- The *Cxcr5* gene is located on the Chr9. 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Cxcr5 chemokine (C-X-C motif) receptor 5 [*Mus musculus* (house mouse)]







Gene ID: 12145, updated on 19-Nov-2019

Summary

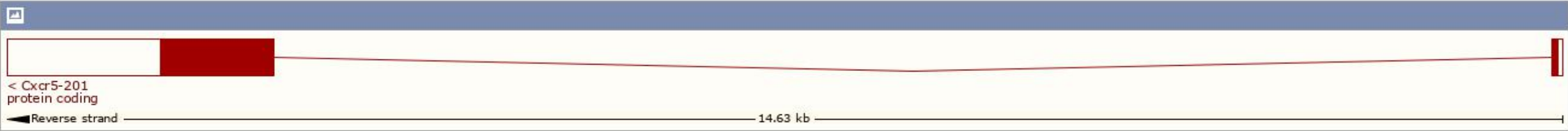
Official Symbol	Cxcr5 provided by MGI
Official Full Name	chemokine (C-X-C motif) receptor 5 provided by MGI
Primary source	MGI:MGI:103567
See related	Ensembl:ENSMUSG000000047880
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Blr1; Gpcr6; MDR15; CXCR-R5; CXCR-5
Expression	Biased expression in spleen adult (RPKM 61.9), mammary gland adult (RPKM 12.5) and 1 other tissue See more
Orthologs	human all

Transcript information (Ensembl)

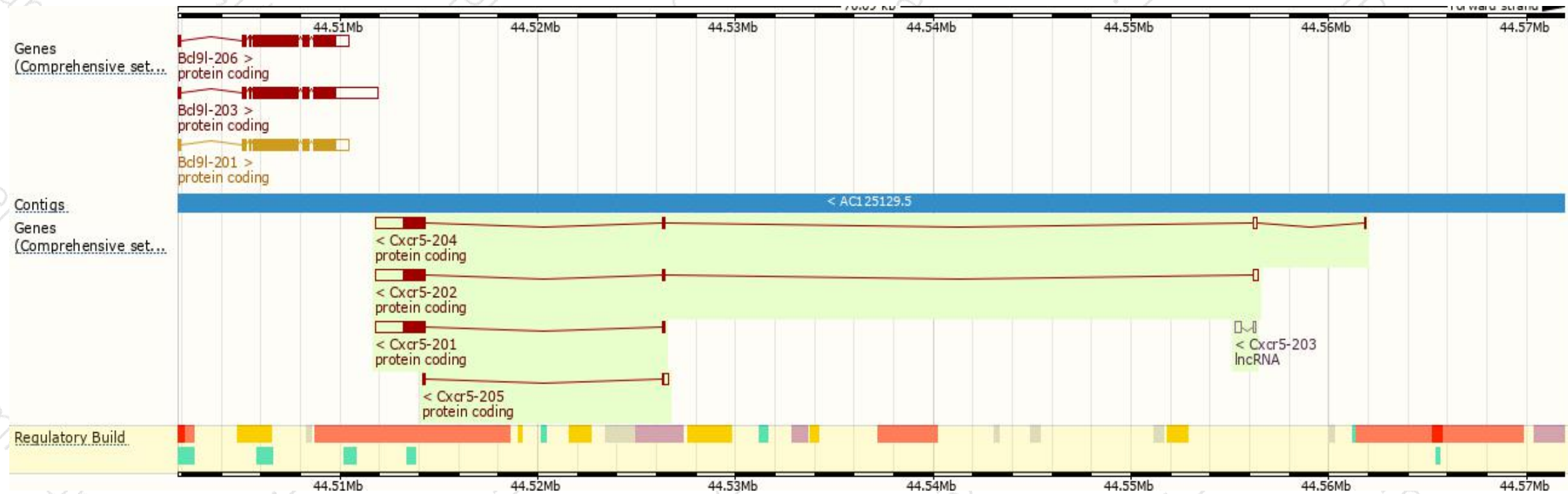
The gene has 5 transcripts, and all transcripts are shown below:

Show/hide columns (1 hidden)							Filter	
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
Cxcr5-204	ENSMUST00000215293.1	2846	374aa	 Protein coding	CCDS23115	Q04683	TSL:1	GENCODE basic APPRIS P1
Cxcr5-202	ENSMUST00000179828.7	2824	374aa	 Protein coding	CCDS23115	Q04683	TSL:5	GENCODE basic APPRIS P1
Cxcr5-201	ENSMUST00000062215.7	2614	374aa	 Protein coding	CCDS23115	Q04683	TSL:1	GENCODE basic APPRIS P1
Cxcr5-205	ENSMUST00000215661.1	358	51aa	 Protein coding	-	A0A1L1SRC9	CDS 3' incomplete TSL:1	
Cxcr5-203	ENSMUST00000213357.1	377	No protein	 lncRNA	-	-	TSL:3	

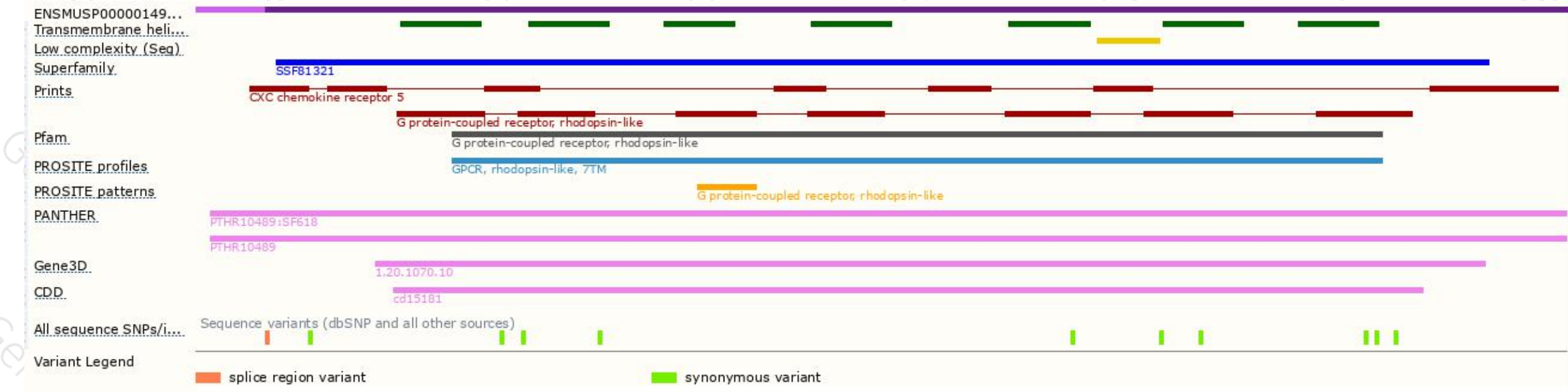
The strategy is based on the design of Cxcr5-201 transcript, The transcription is shown below:



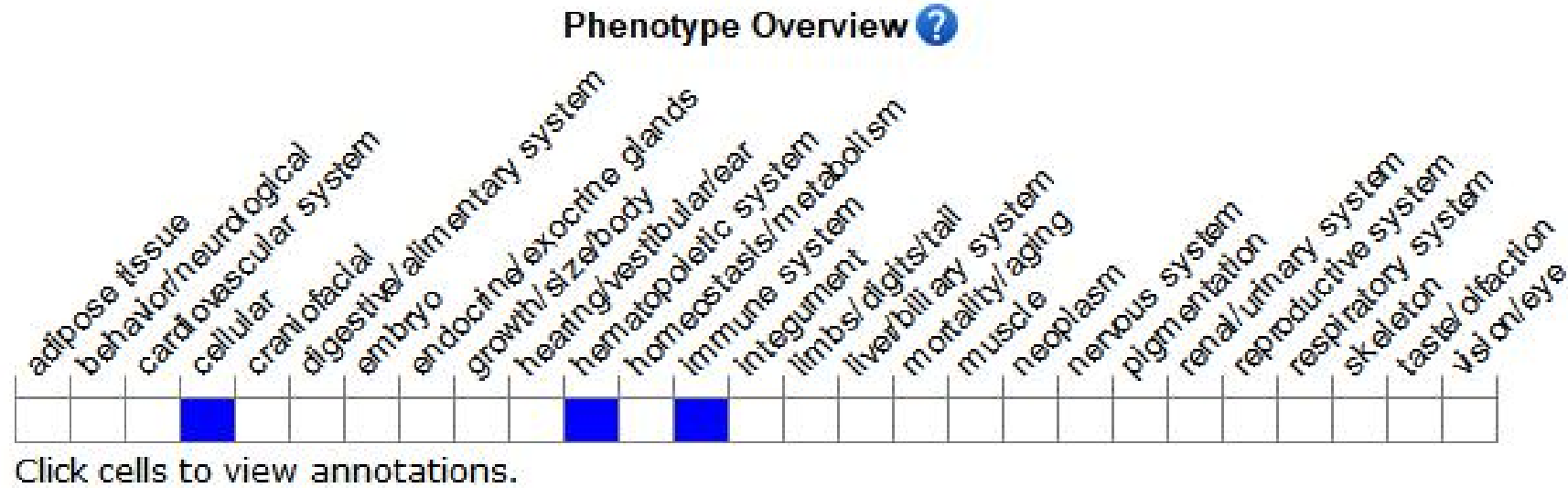
Genomic location distribution



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 mutants lack inguinal lymph nodes, have a few abnormal or no Peyer's patches, morphologically altered primary lymphoid follicles and no functional germinal centers in their spleen.

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