Mettl14 Cas9-CKO Strategy

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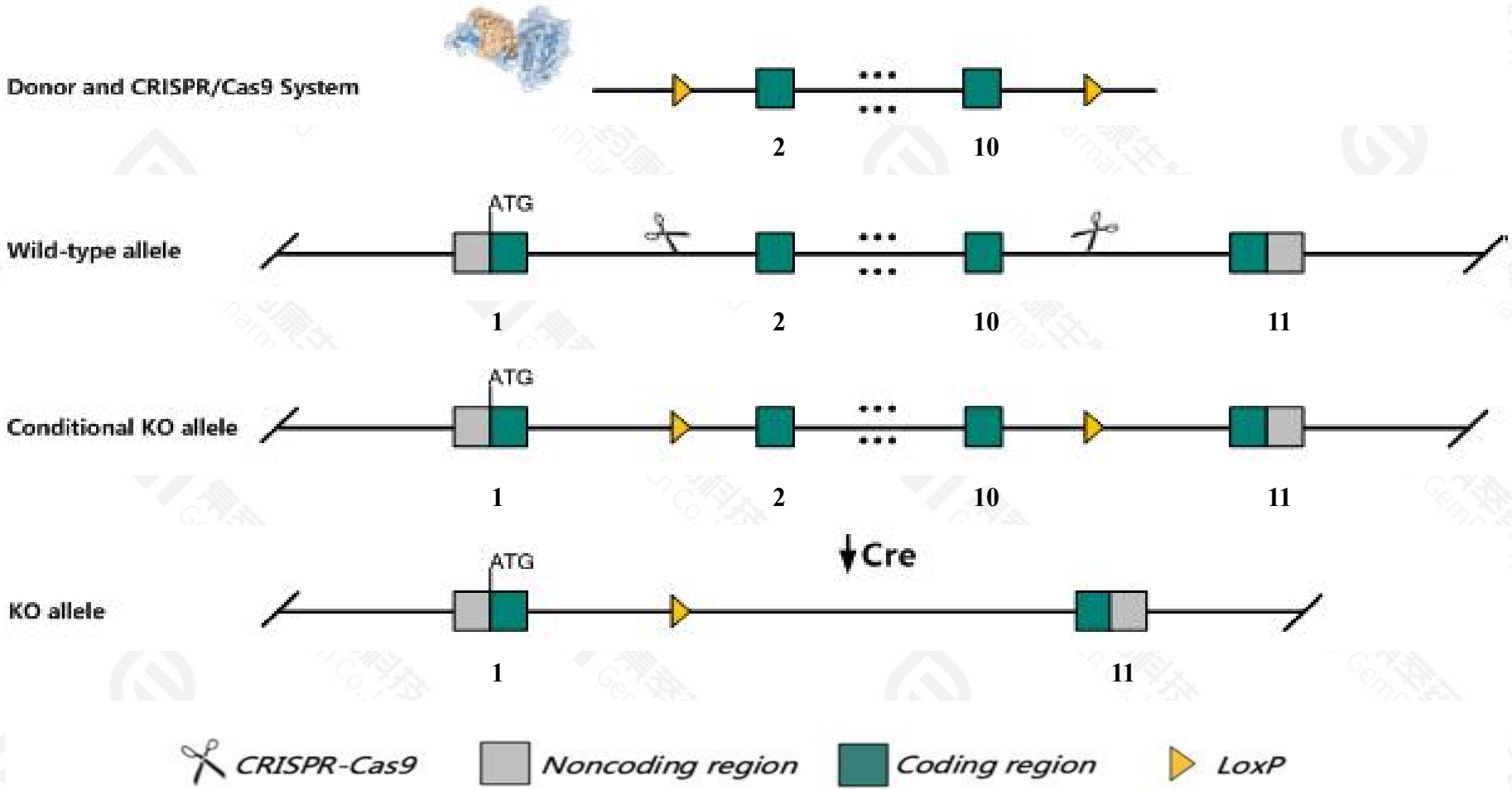
Design Date: 2019-7-18

Project Overview

Project Name	***Mettl14***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR-Cas9 technology to edit the *Mettl14* gene. The schematic diagram is as follows:



Technical routes

➢ The *Mettl14* gene has 5 transcripts. According to the structure of *Mettl14* gene, exon2-exon10 of *Mettl14-201*(ENSMUST00000029759.16) transcript is recommended as the knockout region.The region contains 1000bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR-Cas9 technology to modify *Mettl14* 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,mice homozygous for a knock-out allele exhibit embryonic lethality and decreased histone acetylation. Mice homozygous for a conditional allele activated in neuronal stem cells exhibit decreased NSC proliferation and premature differentiation and decreased number of late-born neurons.

➢ The *Mettl14* 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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Mettl14 methyltransferase like 14 [Mus musculus (house mouse)]

Gene ID: 210529, updated on 12-Jul-2022

Summary

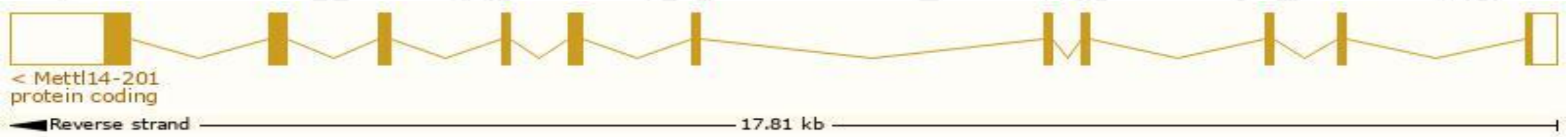
Official Symbol	Mettl14 provided by MGI
Official Full Name	methyltransferase like 14 provided byMGI
Primary source	MGI:MGI:2442926
See related	Ensembl:ENSMUSG00000028114
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	G430022H21Rik, mKIAA1627
Expression	Ubiquitous expression in CNS E11.5 (RPKM 10.1), CNS E14 (RPKM 6.0) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

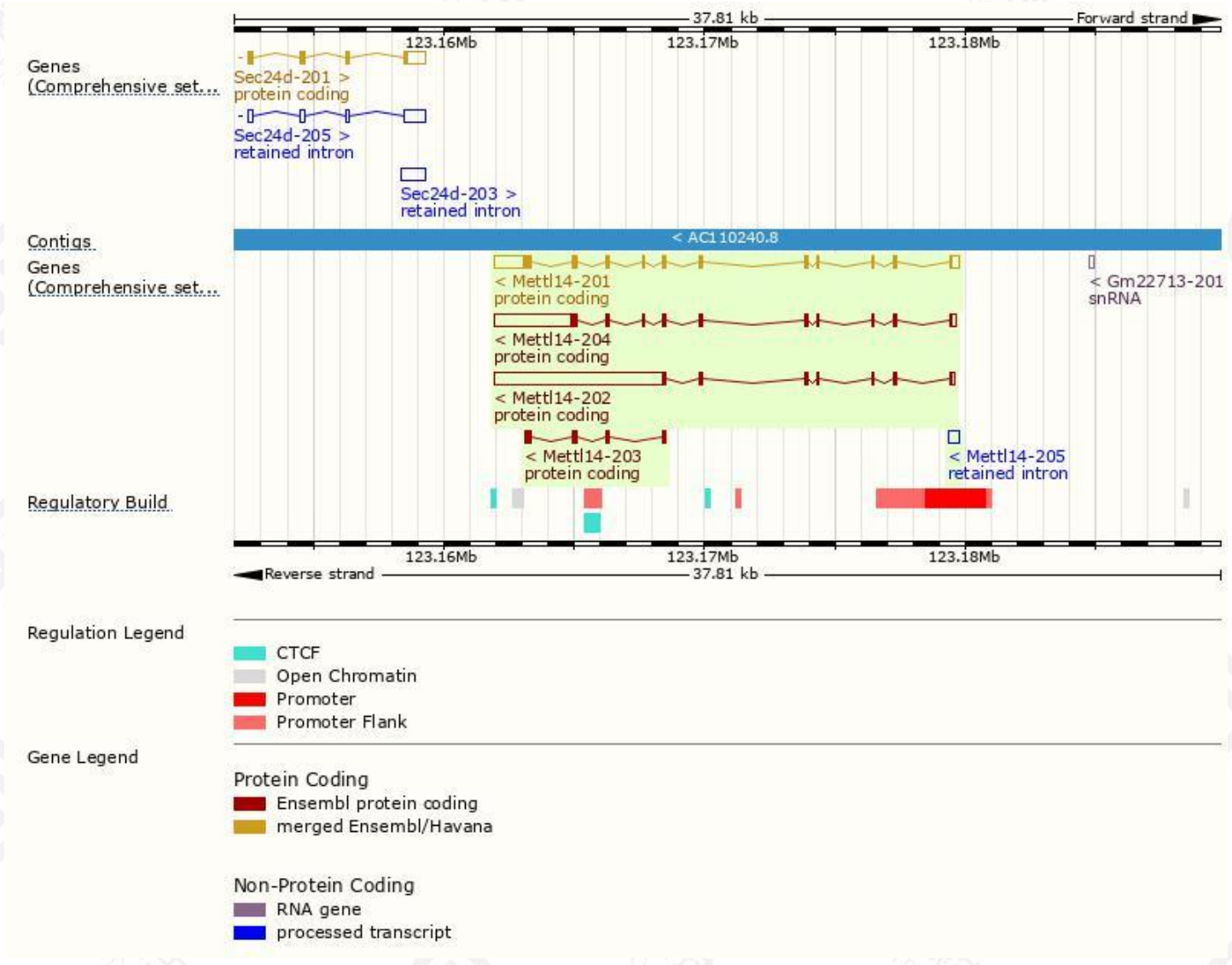
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mettl14-201	ENSMUST00000029759.16	2741	456aa	Protein coding	CCDS38622		TSL:1 , GENCODE basic , APPRIS P1 ,
Mettl14-202	ENSMUST00000090371.14	7197	217aa	Protein coding	-		TSL:1 , GENCODE basic ,
Mettl14-204	ENSMUST00000174323.6	4202	377aa	Protein coding	-		TSL:1 , GENCODE basic ,
Mettl14-203	ENSMUST00000174006.5	634	211aa	Protein coding	-		CDS 5' and 3' incomplete , TSL:3 ,
Mettl14-205	ENSMUST00000197628.2	382	No protein	Retained intron	-		TSL:NA ,

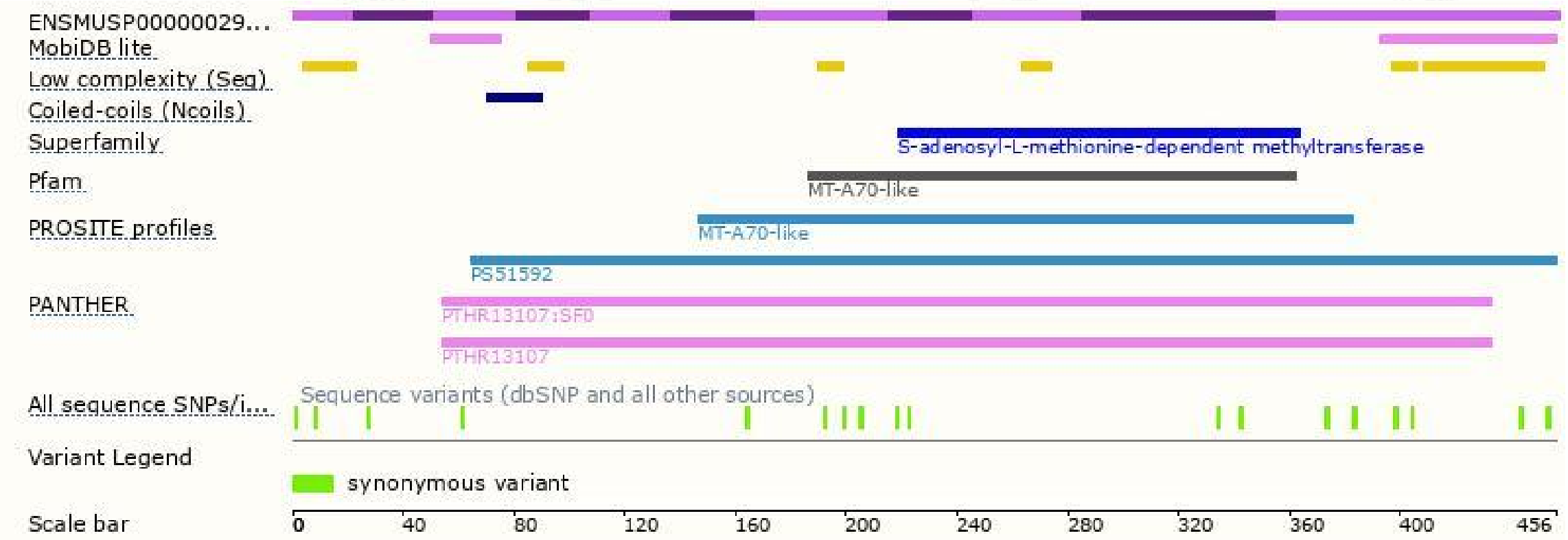
The strategy is based on the design of *Mettl14-201* transcript,the transcription is shown below:



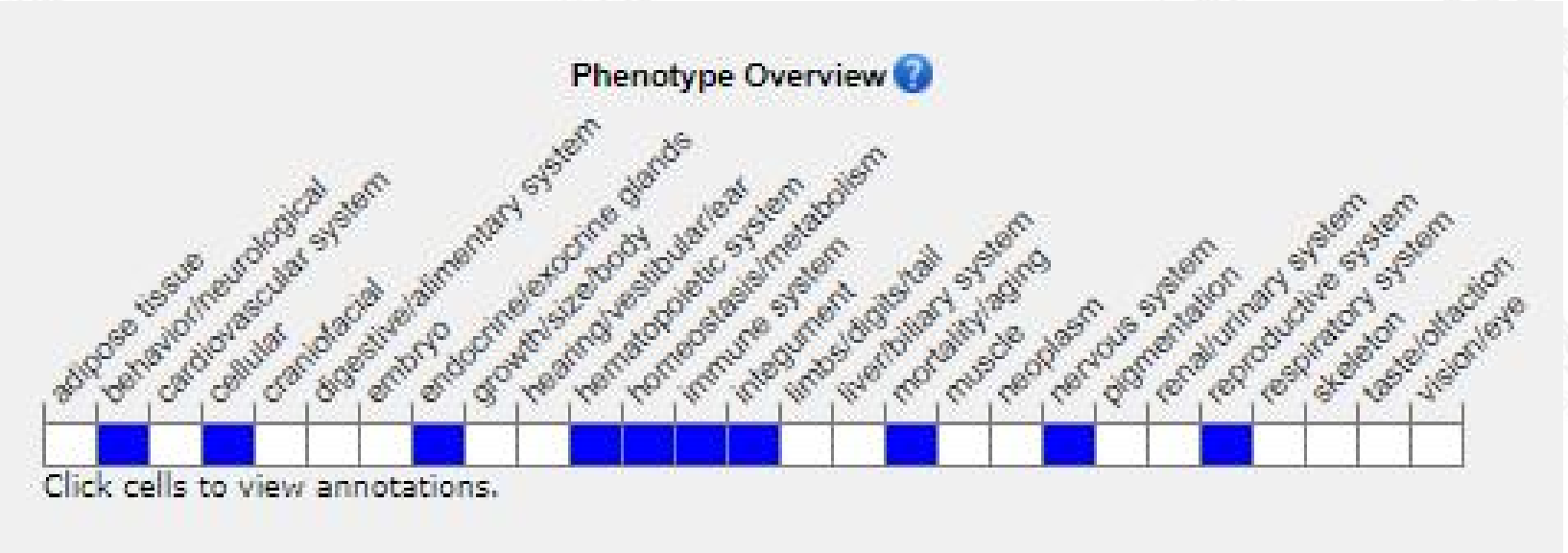
Genomic location distribution



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/).

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