Sept5 Cas9-CKO Strategy

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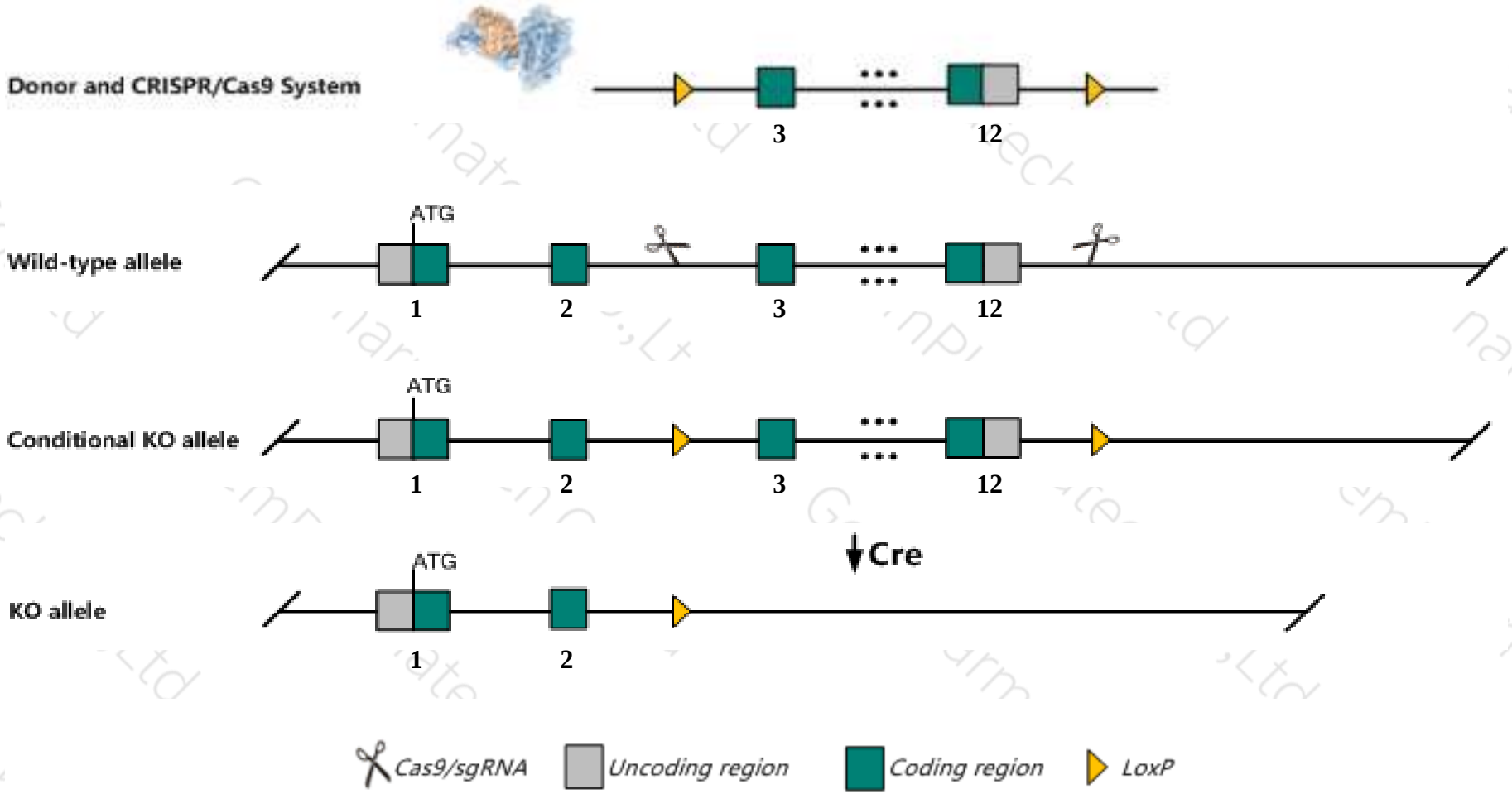
Project Overview

Project Name	***Sept5***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sept5* gene. The schematic diagram is as follows:



Technical routes

- The *Sept5* gene has 5 transcripts. According to the structure of *Sept5* gene, exon3-exon12 of *Sept5-201* (ENSMUST00000096987.6) transcript is recommended as the knockout region.The region contains most of the coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Sept5* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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 disruptions in this gene show no gross phenotypic changes. Partial defects in synaptic transmission is reported for one allele, and platelet secretion and modest behavioral defects reported for a different allele.

➢ *Gp1bb* and *Gm49601* gene will be deleted together in this strategy.

➢ The *Sept5* 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）

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Sept5 septin 5 [*Mus musculus* (house mouse)]

Gene ID: 18951, updated on 10-Oct-2019

Summary

Official Symbol	Sept5 provided by MGI
Official Full Name	septin 5 provided by MGI
Primary source	MGI:MGI:1195461
See related	Ensembl:ENSMUSG00000072214
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	Pnutl1; Cdcrel1; Septin5; Cdcrel-1
Expression	Broad expression in cortex adult (RPKM 236.3), large intestine adult (RPKM 199.0) and 15 other tissues See more
Orthologs	human all

Genomic context

Location: 16 A3; 16 11.53 cM

See Sept5 in Genome Data Viewer

Exon count: 12

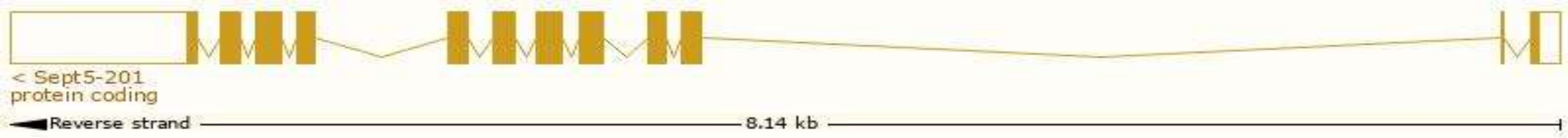
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	16	NC_000082.6 (18621811..18629938, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	16	NC_000082.5 (18621904..18630031, complement)

Transcript information（Ensembl）

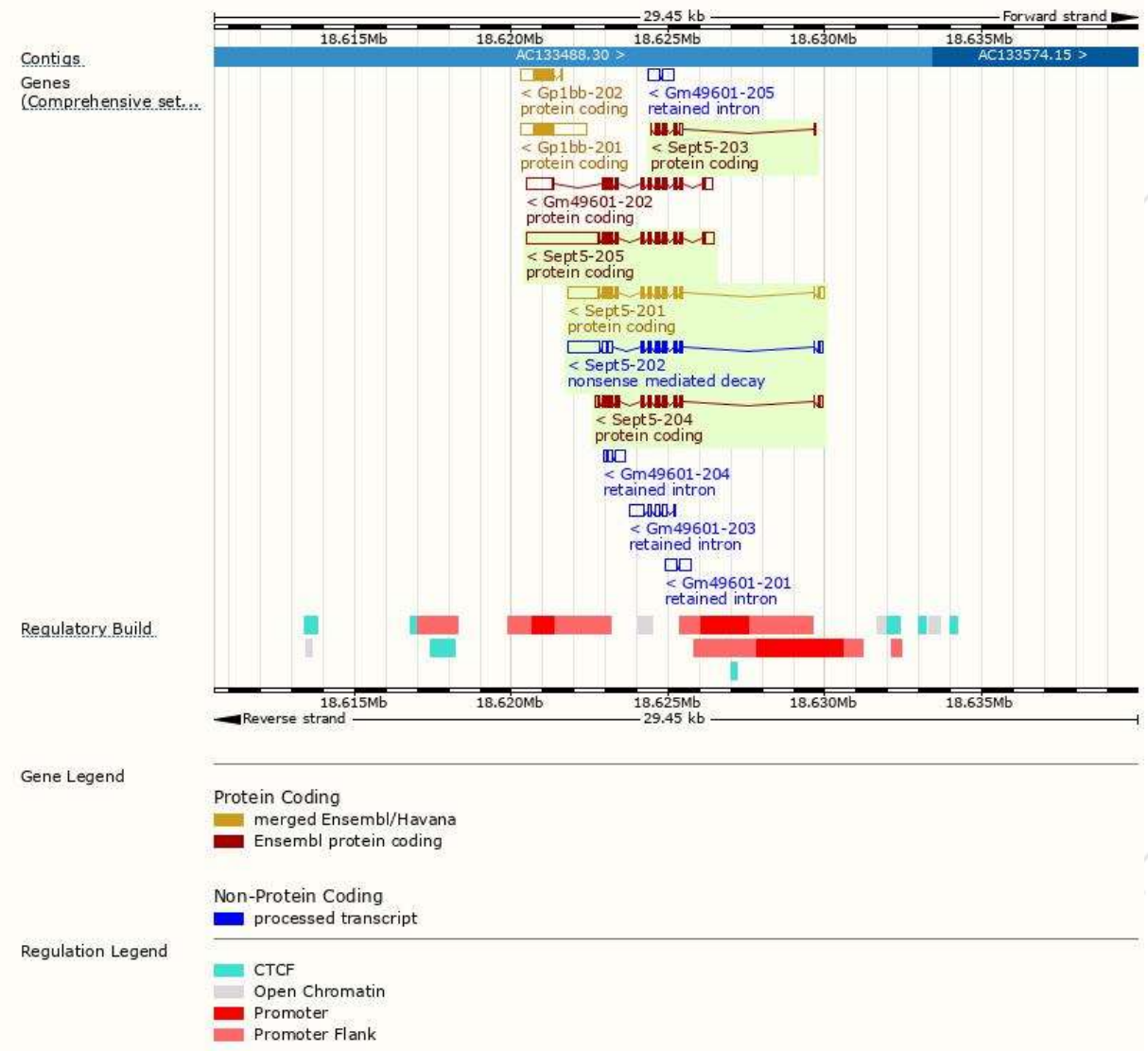
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sept5-201	ENSMUST00000096987.6	2143	369aa	Protein coding	CCDS57021	Q9Z2Q6	TSL:1 GENCODE basic APPRIS P2
Sept5-205	ENSMUST00000232653.1	3678	378aa	Protein coding	-	A0A338P769	GENCODE basic APPRIS ALT1
Sept5-204	ENSMUST00000231956.1	1314	381aa	Protein coding	-	B7ZNM7	GENCODE basic
Sept5-203	ENSMUST00000231622.1	521	126aa	Protein coding	-	A0A338P755	CDS 3' incomplete
Sept5-202	ENSMUST00000231244.1	2003	248aa	Nonsense mediated decay	-	A0A338P729	

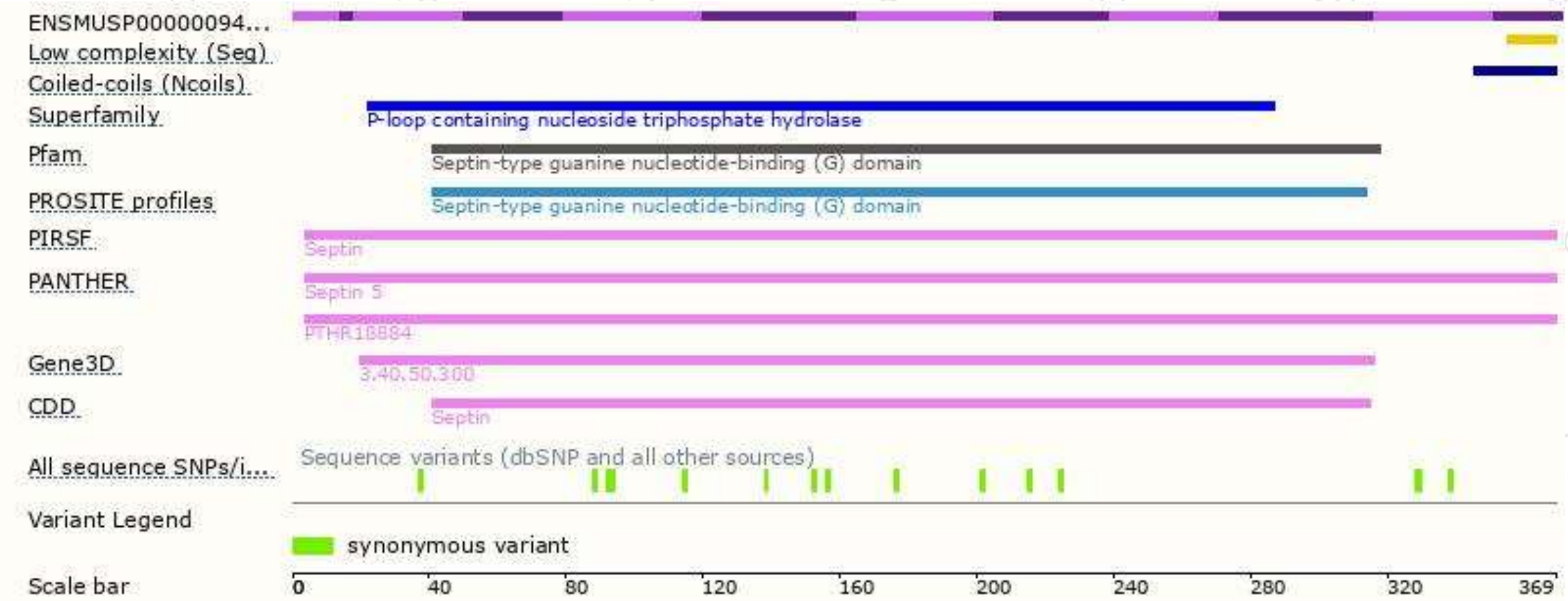
The strategy is based on the design of *Sept5-201* transcript,The transcription is shown below



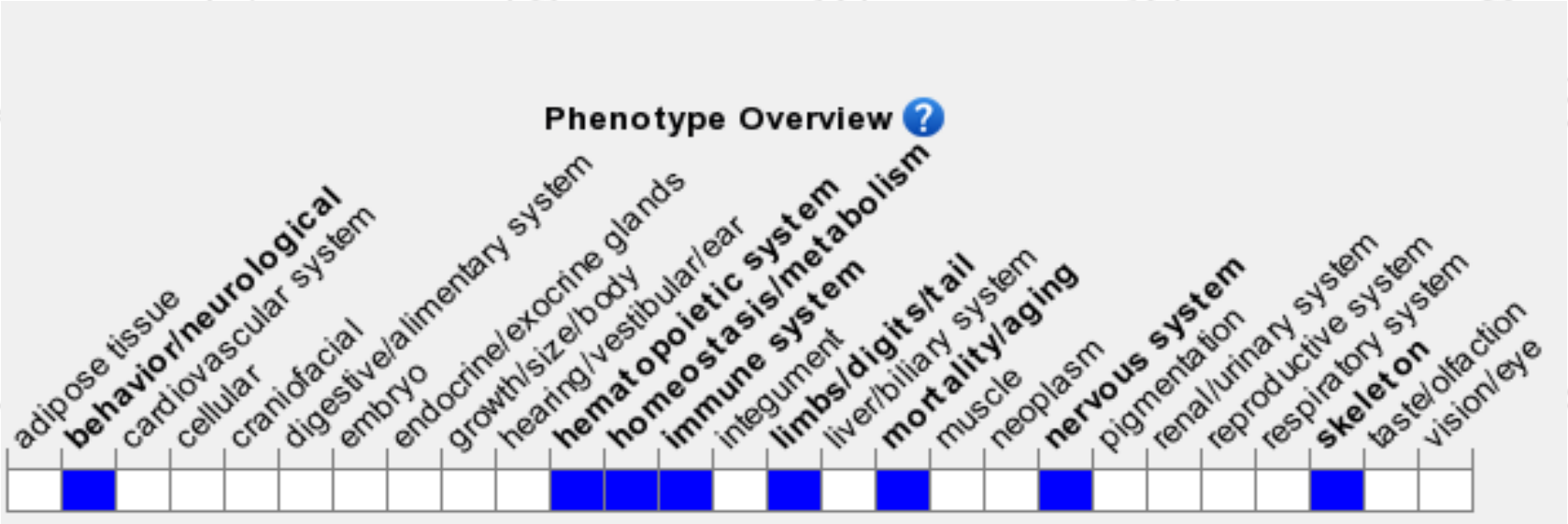
Genomic location distribution



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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