

***Comt* Cas9-CKO Strategy**

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Project Overview

Project Name

Comt

Project type

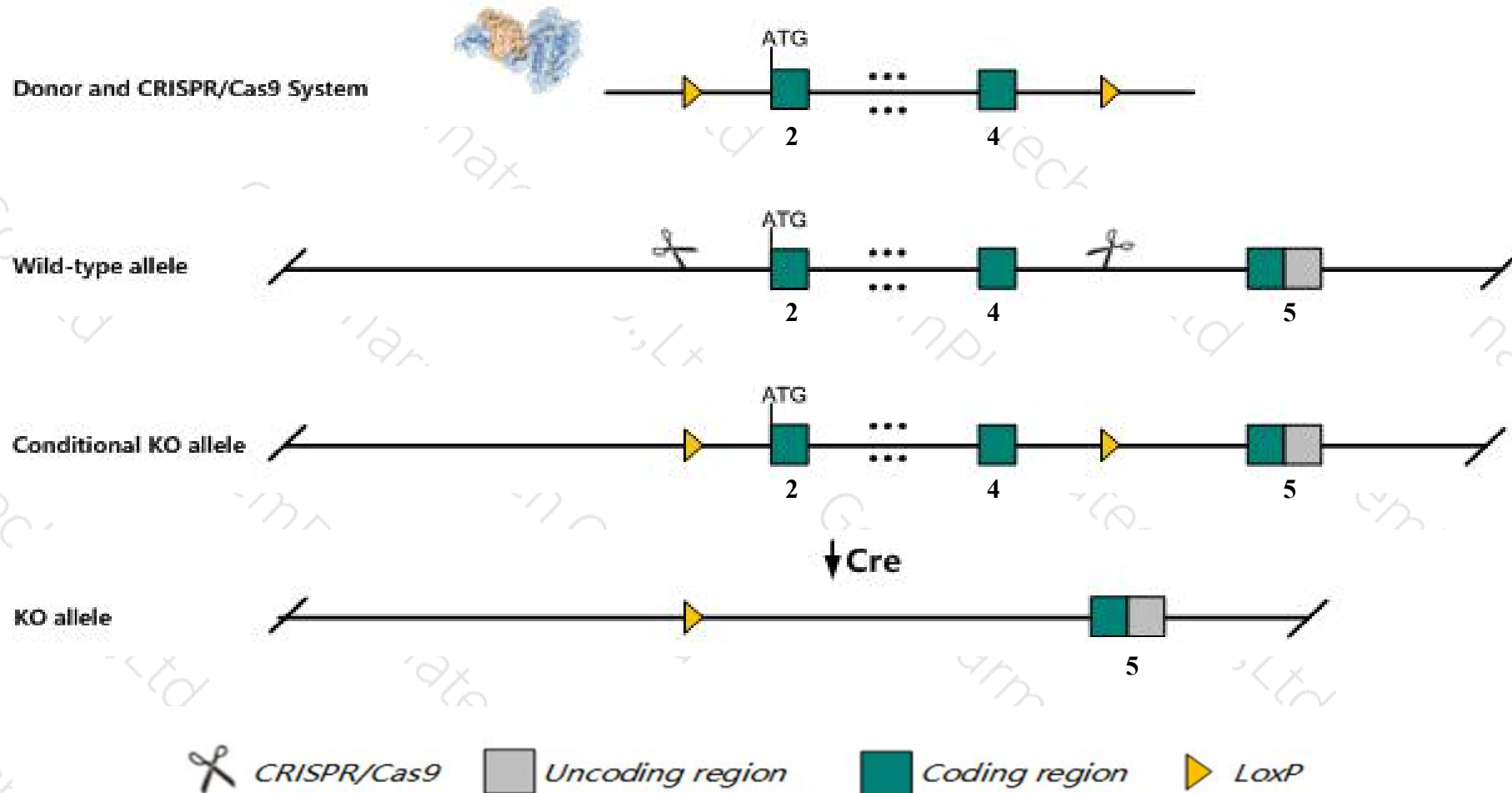
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Comt* gene has 4 transcripts. According to the structure of *Comt* gene, exon2-exon4 of *Comt-204* (ENSMUST00000165430.7) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Comt* 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, Mice homozygous for disruption of this gene are viable, fertile, and show no gross or histological abnormalities. However dopamine levels in the frontal cortex of males are increased. Also, males show increased aggression and females show increased anxiety.
- The floxed region is near to the N-terminal of *Gm15764* gene, this strategy may influence the regulatory function of the N-terminal of *Gm15764* gene.
- The *Comt* 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)

Comt catechol-O-methyltransferase [*Mus musculus* (house mouse)]

Gene ID: 12846, updated on 12-Aug-2019

Summary

Official Symbol Comt provided by [MGI](#)
Official Full Name catechol-O-methyltransferase provided by [MGI](#)
Primary source [MGI:MG1:88470](#)
See related [Ensembl:ENSMUSG00000000326](#)
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 Comt1; D16Wsu103e; D330014B15Rik
Expression Broad expression in liver adult (RPKM 254.4), subcutaneous fat pad adult (RPKM 175.4) and 25 other tissues [See more](#)
Orthologs [human](#) [all](#)

Genomic context

Location: 16 A3; 16 11.4 cM

See Comt in [Genome Data Viewer](#)

Exon count: 7

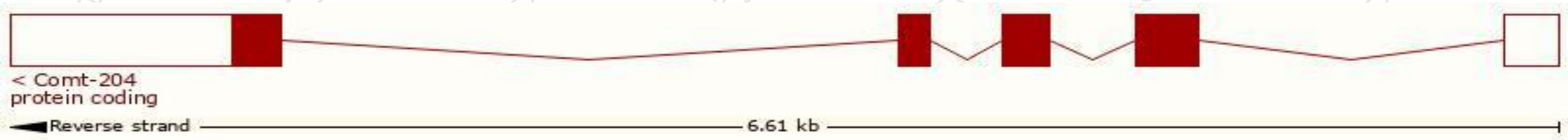
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	16	NC_000082.6 (18406882..18426716, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	16	NC_000082.5 (18406975..18426809, complement)

Transcript information (Ensembl)

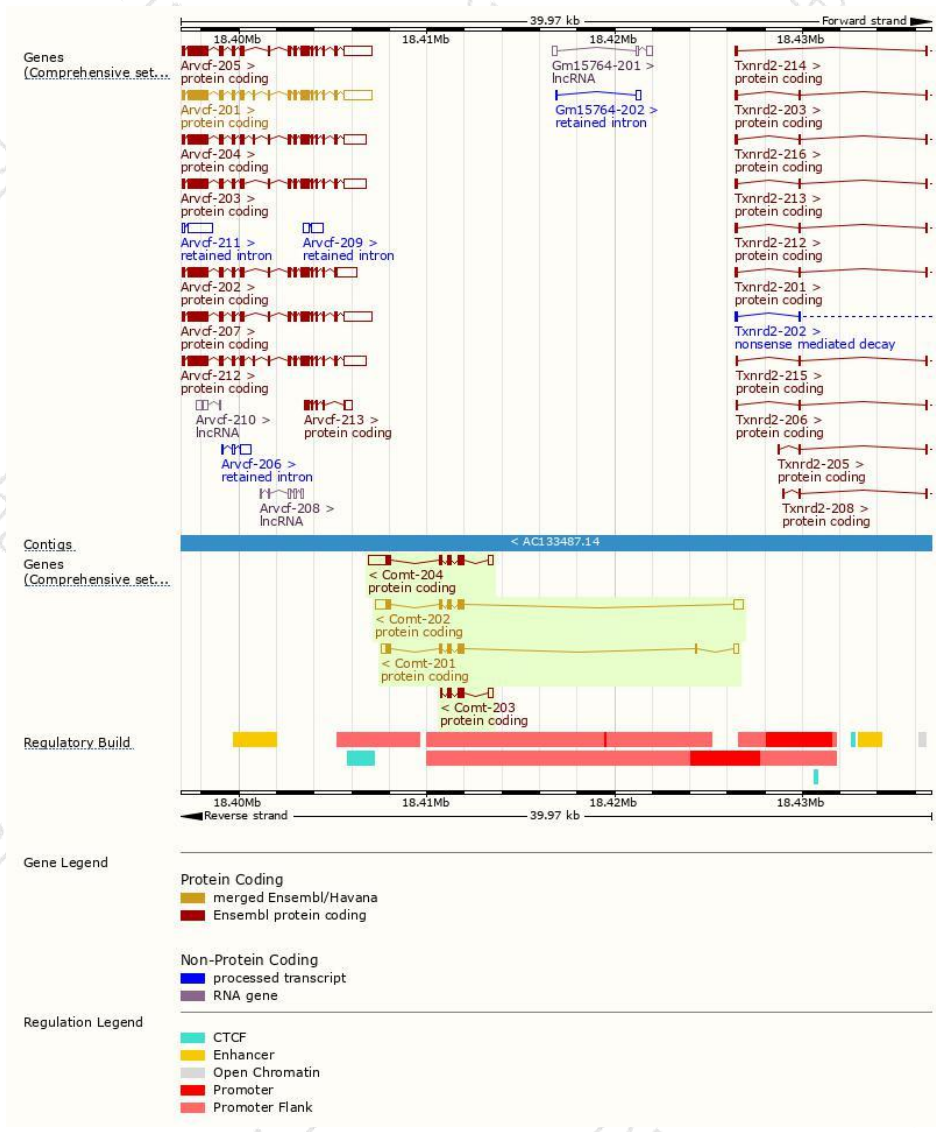
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Comt-204	ENSMUST00000165430.7	1978	265aa	Protein coding	CCDS28021	O88587	TSL:5 GENCODE basic APPRIS P1
Comt-202	ENSMUST00000115609.9	1866	265aa	Protein coding	CCDS28021	O88587	TSL:1 GENCODE basic APPRIS P1
Comt-201	ENSMUST00000000335.11	1397	265aa	Protein coding	CCDS28021	O88587	TSL:1 GENCODE basic APPRIS P1
Comt-203	ENSMUST00000147720.2	743	171aa	Protein coding	-	D3Z227	CDS 3' incomplete TSL:2

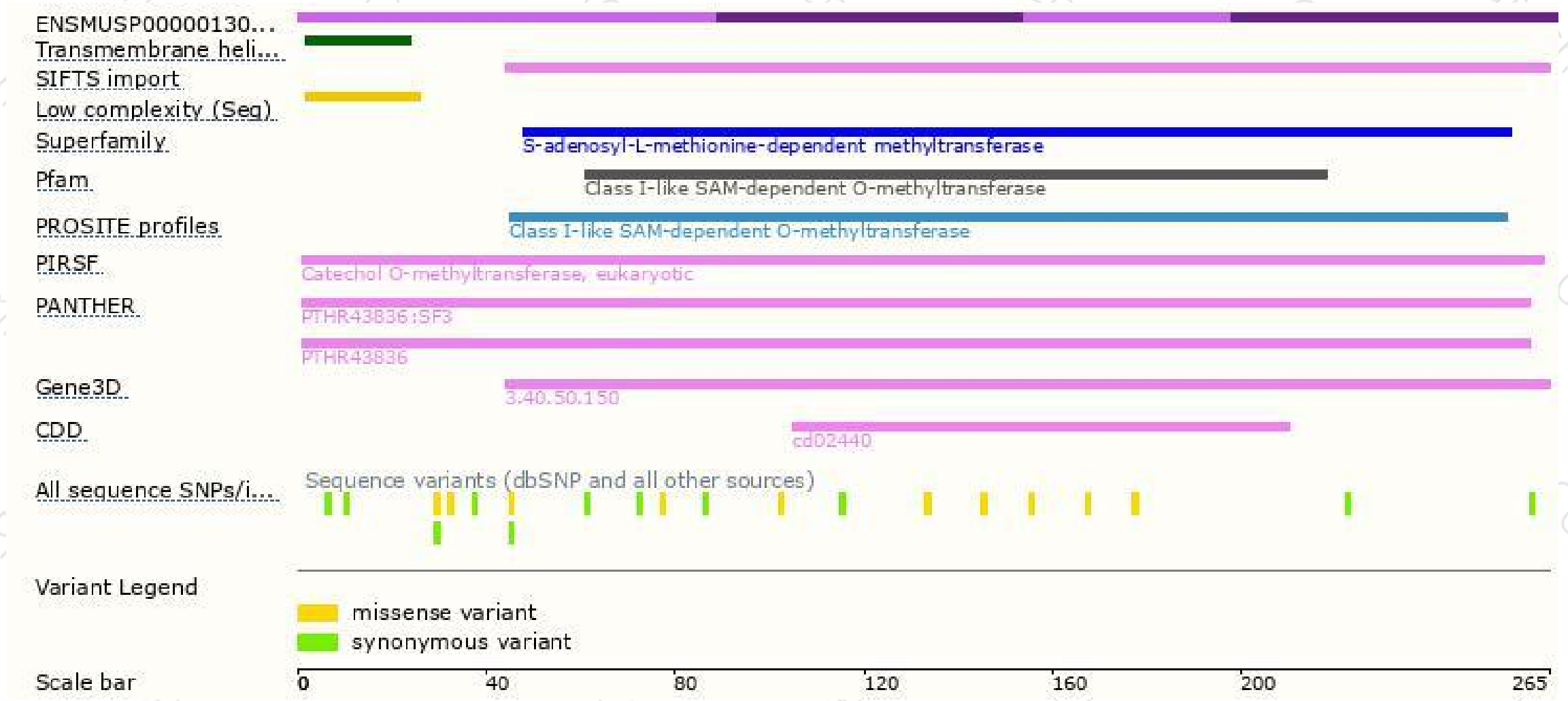
The strategy is based on the design of *Comt-204* transcript,The transcription is shown below



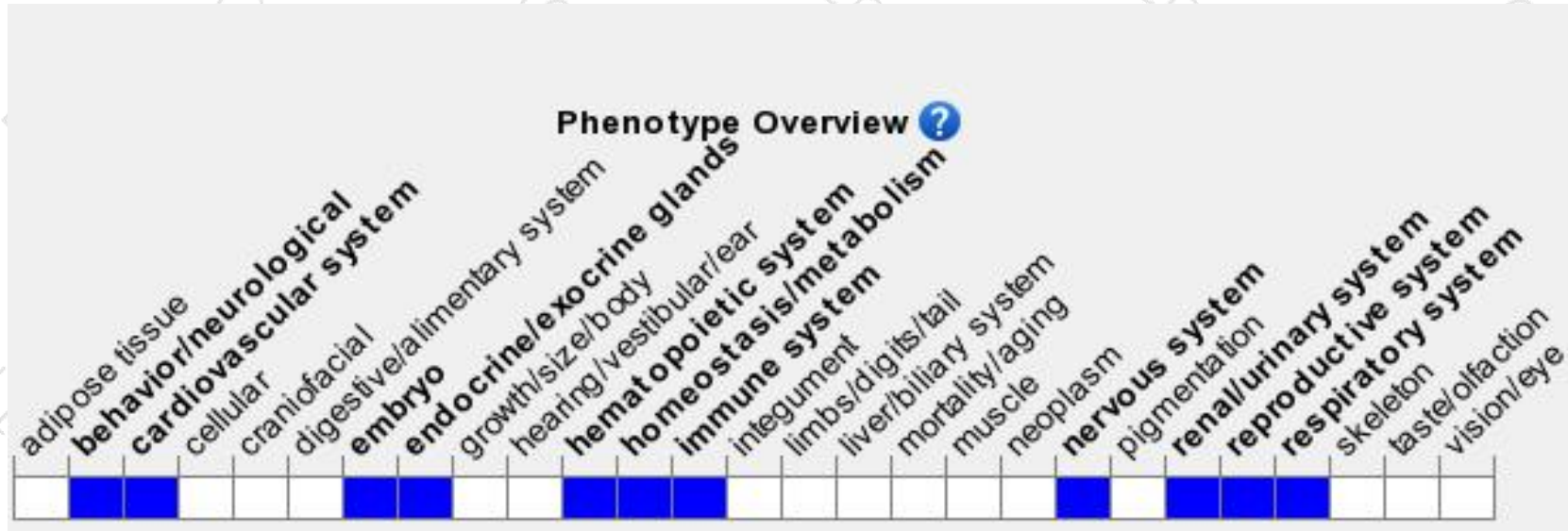
Genomic location distribution



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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