Htr3a Cas9-CKO Strategy

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Design Date: 2019-11-20
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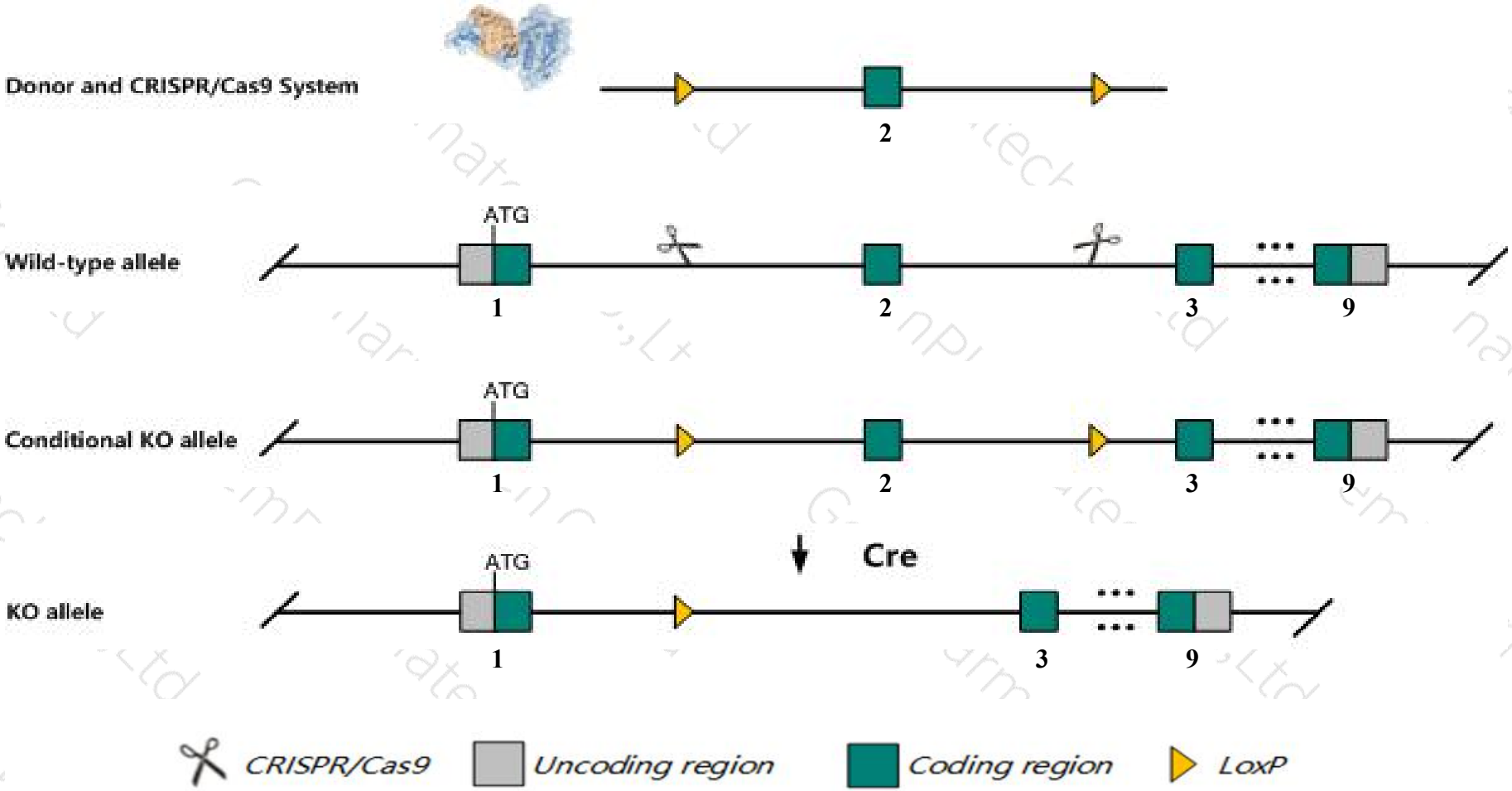
Project Overview

Project Name	***Htr3a***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Htr3a* gene. The schematic diagram is as follows:



Technical routes

- The *Htr3a* gene has 2 transcripts. According to the structure of *Htr3a* gene, exon2 of *Htr3a-201* (ENSMUST00000003826.7) transcript is recommended as the knockout region.The region contains 167bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Htr3a* 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,Homozygous mice display a decreased lifespan, cachexia, increased blood urea nitrogen, proteinuria, kidney inflammation, and a hyperdistended and neurogenic urinary bladder. Mice homozygous for a second null mutation display reduced chemical pain persistence responses but are otherwise healthy.

- The *Htr3a* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Htr3a 5-hydroxytryptamine (serotonin) receptor 3A [*Mus musculus* (house mouse)]

Gene ID: 15561, updated on 24-Oct-2019

Summary

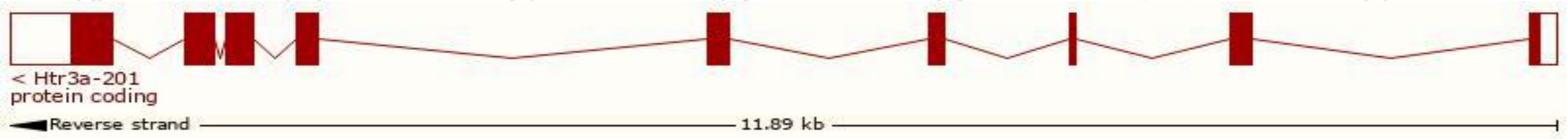
Official Symbol	Htr3a provided by MGI
Official Full Name	5-hydroxytryptamine (serotonin) receptor 3A provided by MGI
Primary source	MGI:MGI:96282
See related	Ensembl:ENSMUSG00000032269
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	5-HT3
Expression	Biased expression in colon adult (RPKM 3.9), adrenal adult (RPKM 3.2) and 14 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

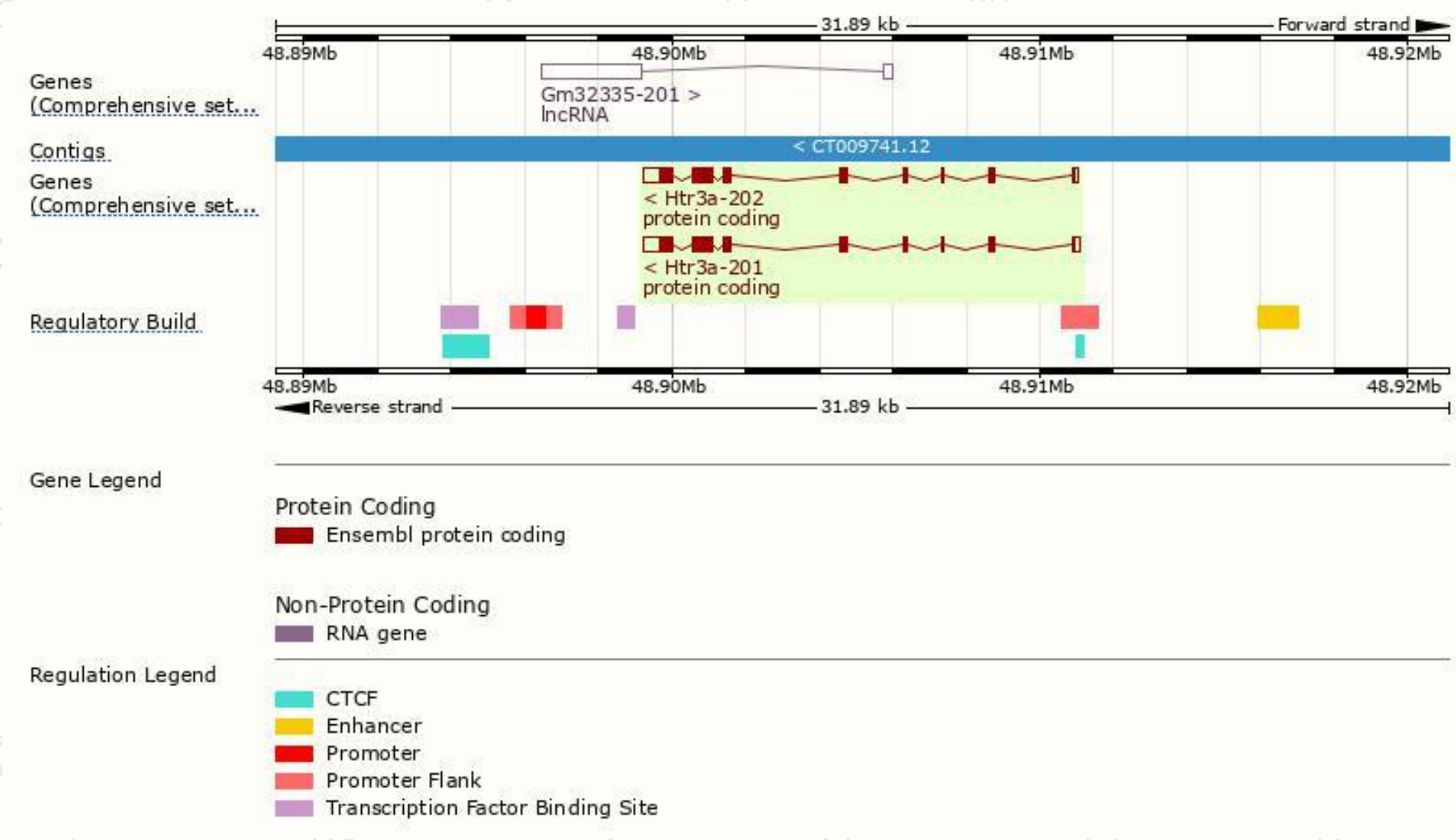
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Htr3a-201	ENSMUST00000003826.7	2082	489aa	Protein coding	CCDS52788	E9QLC0	TSL:1 GENCODE basic APPRIS P2
Htr3a-202	ENSMUST00000217289.1	1987	483aa	Protein coding	-	Q8K1F4	TSL:1 GENCODE basic APPRIS ALT2

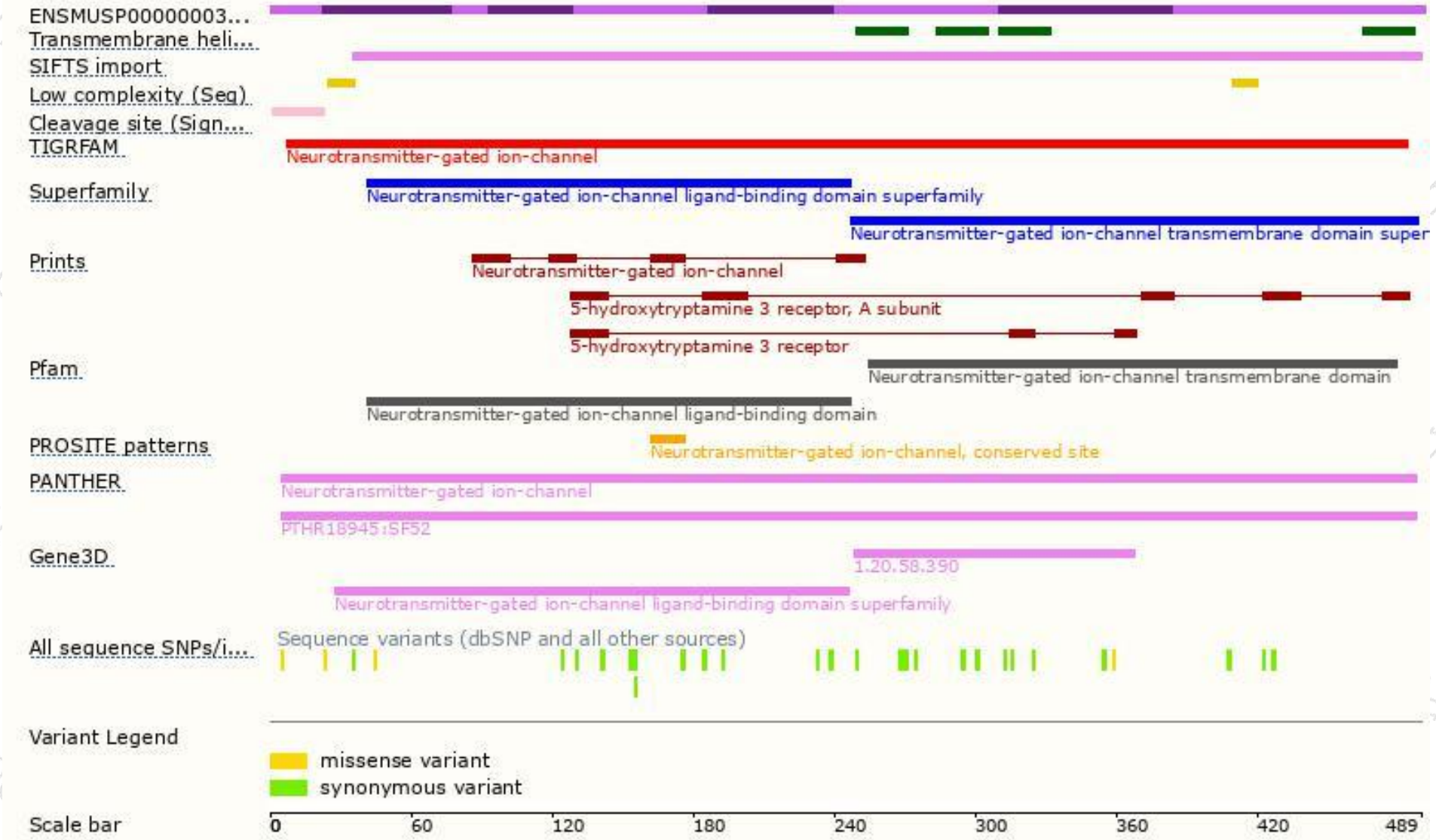
The strategy is based on the design of *Htr3a-201* transcript,The transcription is shown below



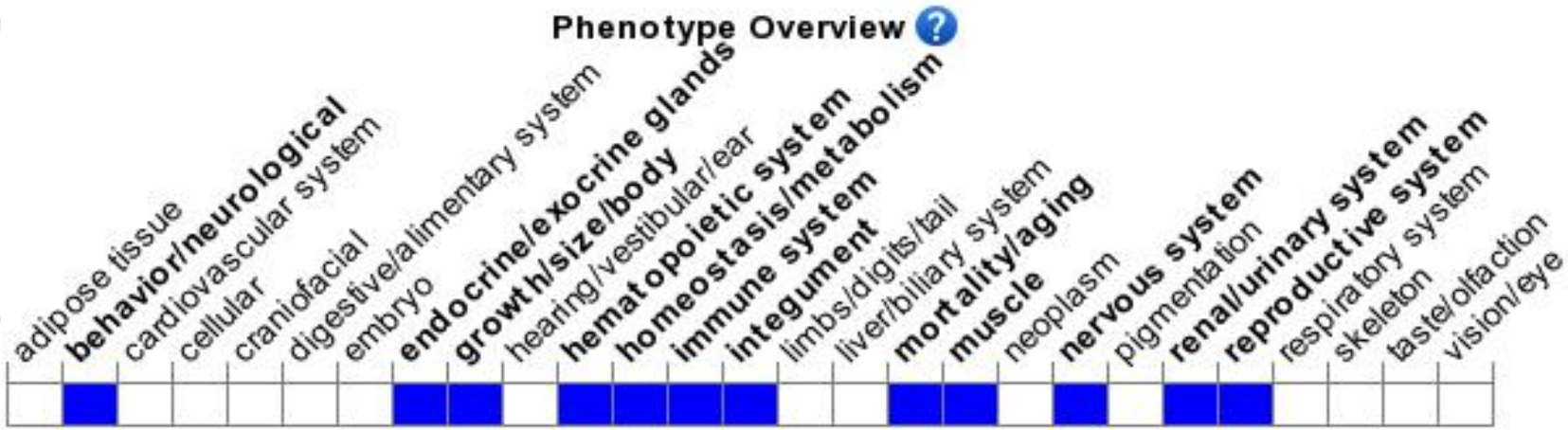
Genomic location distribution



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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