

Btbd9 Cas9-CKO Strategy

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Design Date:

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Project Overview

Project Name

Btbd9

Project type

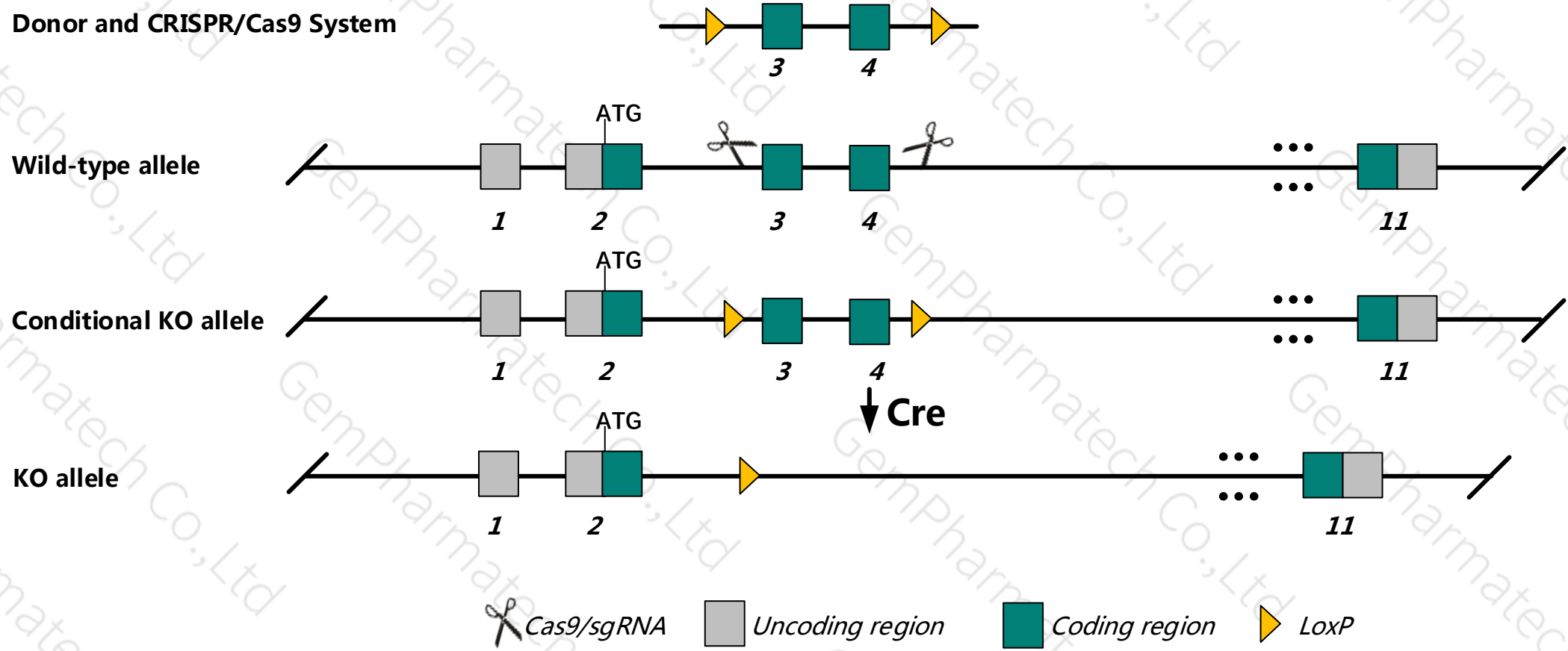
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Btbd9* gene. The schematic diagram is as follows:



- The *Btbd9* gene has 25 transcripts. According to the structure of *Btbd9* gene, exon3-exon4 of *Btbd9*-202 (ENSMUST00000168787.7) transcript is recommended as the knockout region. The region contains 629bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Btbd9* 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 or cell types.

- According to the existing MGI data , Mice homozygous for a gene trapped allele show hyperactivity, unidirectional circling, sleep disturbances, thermal sensory alterations, increased serum iron levels, altered serotonin metabolism, enhanced long-term potentiation and paired-pulse ratios, and enhanced cued and contextual fear memory.
- The KO region contains functional region of the *Gm50252* gene. Knockout the region may affect the function of *Gm50252* gene.
- The *Btbd9* gene is located on the Chr17. 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)

Btbd9 BTB (POZ) domain containing 9 [*Mus musculus* (house mouse)]

Gene ID: 224671, updated on 13-Mar-2020

Summary

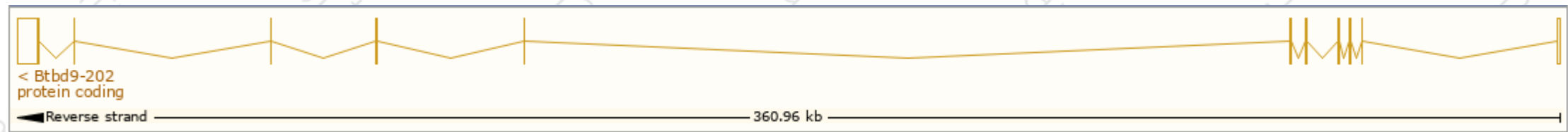
Official Symbol	Btbd9 provided by MGI
Official Full Name	BTB (POZ) domain containing 9 provided by MGI
Primary source	MGI:MGI:1916625
See related	Ensembl:ENSMUSG00000062202
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<i>Mus musculus</i>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI464421; 4930402L05; 1700023F20Rik
Expression	Ubiquitous expression in testis adult (RPKM 5.8), whole brain E14.5 (RPKM 4.8) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

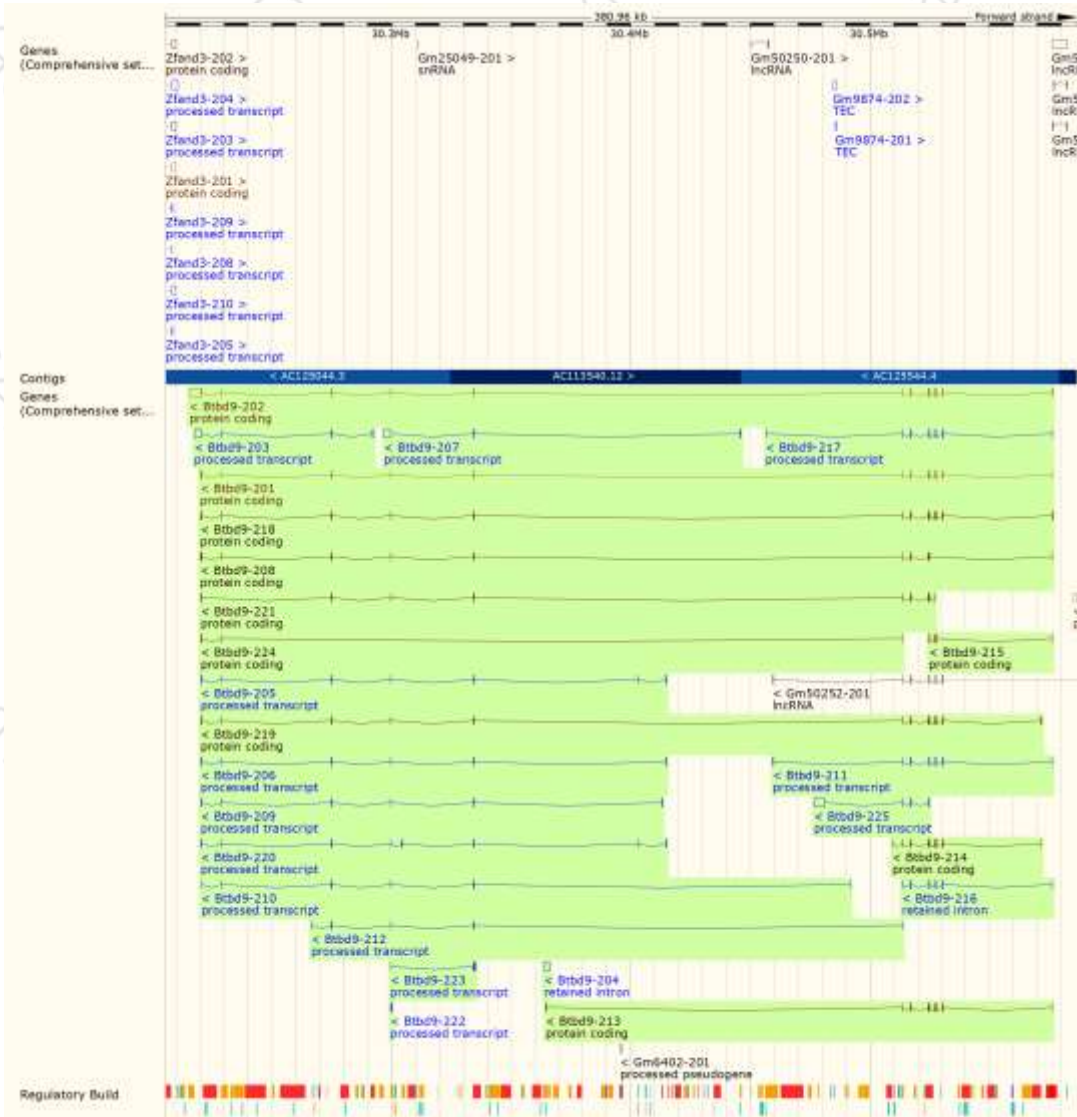
The gene has 25 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Btbd9-202	ENSMUST00000168787.7	7058	612aa	Protein coding	CCDS28599.6	Q8C726.9	TSL:1 GENCODE basic APPRIS P1
Btbd9-201	ENSMUST00000079924.7	2007	612aa	Protein coding	CCDS20599.6	Q8C726.9	TSL:1 GENCODE basic APPRIS P1
Btbd9-219	ENSMUST00000237048.1	1944	612aa	Protein coding	CCDS28599.6	Q8C726.9	GENCODE basic APPRIS P1
Btbd9-218	ENSMUST00000237037.1	2079	582aa	Protein coding	-	A0A49489M3.9	CDS 3' incomplete
Btbd9-208	ENSMUST00000235587.1	1680	391aa	Protein coding	-	A0A49489R7.9	GENCODE basic
Btbd9-213	ENSMUST00000236584.1	1535	435aa	Protein coding	-	A0A49489Q1.6	GENCODE basic
Btbd9-221	ENSMUST00000237164.1	1300	433aa	Protein coding	-	A0A49489J5.9	CDS 5' and 3' incomplete
Btbd9-214	ENSMUST00000236799.1	1272	385aa	Protein coding	-	A0A49489S2.9	GENCODE basic
Btbd9-215	ENSMUST00000236925.1	581	165aa	Protein coding	-	A0A49489P6.9	CDS 2' incomplete
Btbd9-224	ENSMUST00000237732.1	429	132aa	Protein coding	-	A0A49486B6.6	CDS 5' incomplete
Btbd9-225	ENSMUST00000237825.1	4856	No protein	Processed transcript	-	-	-
Btbd9-203	ENSMUST00000235187.1	3480	No protein	Processed transcript	-	-	-
Btbd9-207	ENSMUST00000235555.1	3104	No protein	Processed transcript	-	-	-
Btbd9-211	ENSMUST00000236233.1	1307	No protein	Processed transcript	-	-	-
Btbd9-217	ENSMUST00000237013.1	1287	No protein	Processed transcript	-	-	-
Btbd9-220	ENSMUST00000237078.1	1207	No protein	Processed transcript	-	-	-
Btbd9-205	ENSMUST00000235342.1	1071	No protein	Processed transcript	-	-	-
Btbd9-206	ENSMUST00000235431.1	923	No protein	Processed transcript	-	-	-
Btbd9-209	ENSMUST00000235843.1	863	No protein	Processed transcript	-	-	-
Btbd9-212	ENSMUST00000236430.1	648	No protein	Processed transcript	-	-	-
Btbd9-210	ENSMUST00000236153.1	564	No protein	Processed transcript	-	-	-
Btbd9-223	ENSMUST00000237575.1	357	No protein	Processed transcript	-	-	-
Btbd9-222	ENSMUST00000237635.1	245	No protein	Processed transcript	-	-	-
Btbd9-204	ENSMUST00000235299.1	2266	No protein	Retained intron	-	-	-
Btbd9-216	ENSMUST00000236910.1	1375	No protein	Retained intron	-	-	-

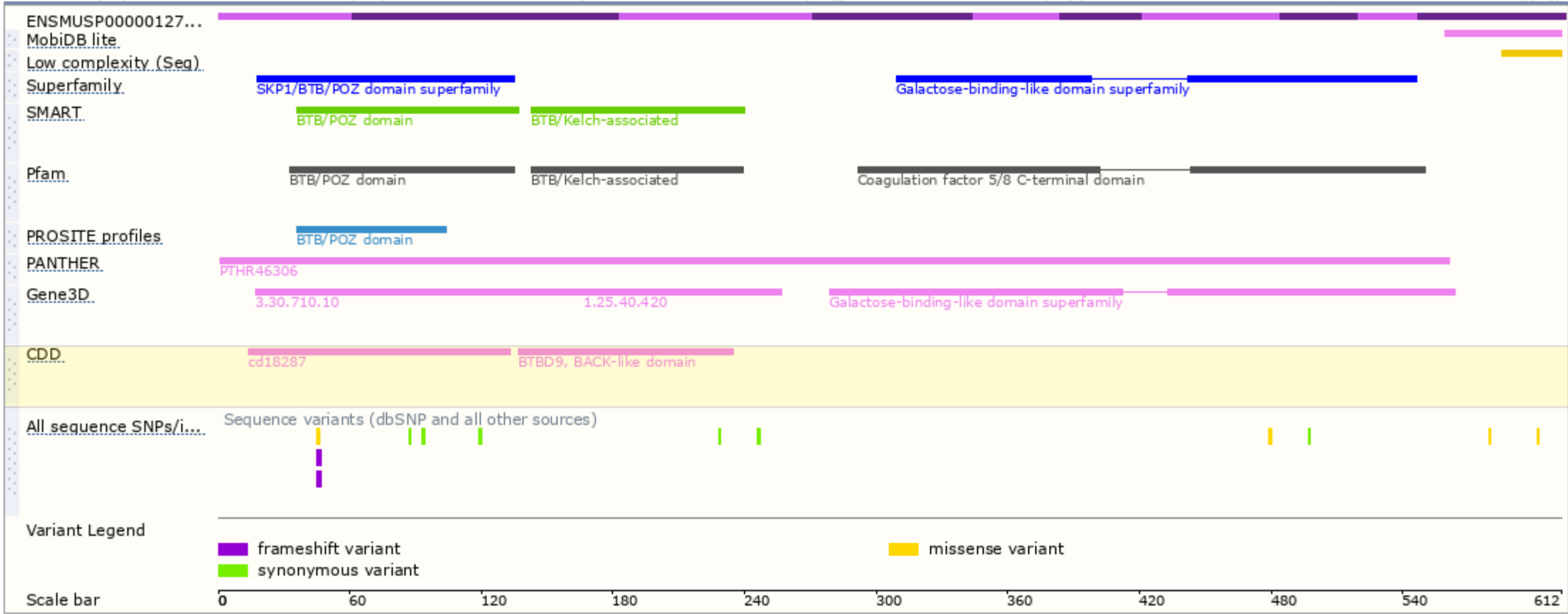
The strategy is based on the design of *Btbd9*-202 transcript, The transcription is shown below



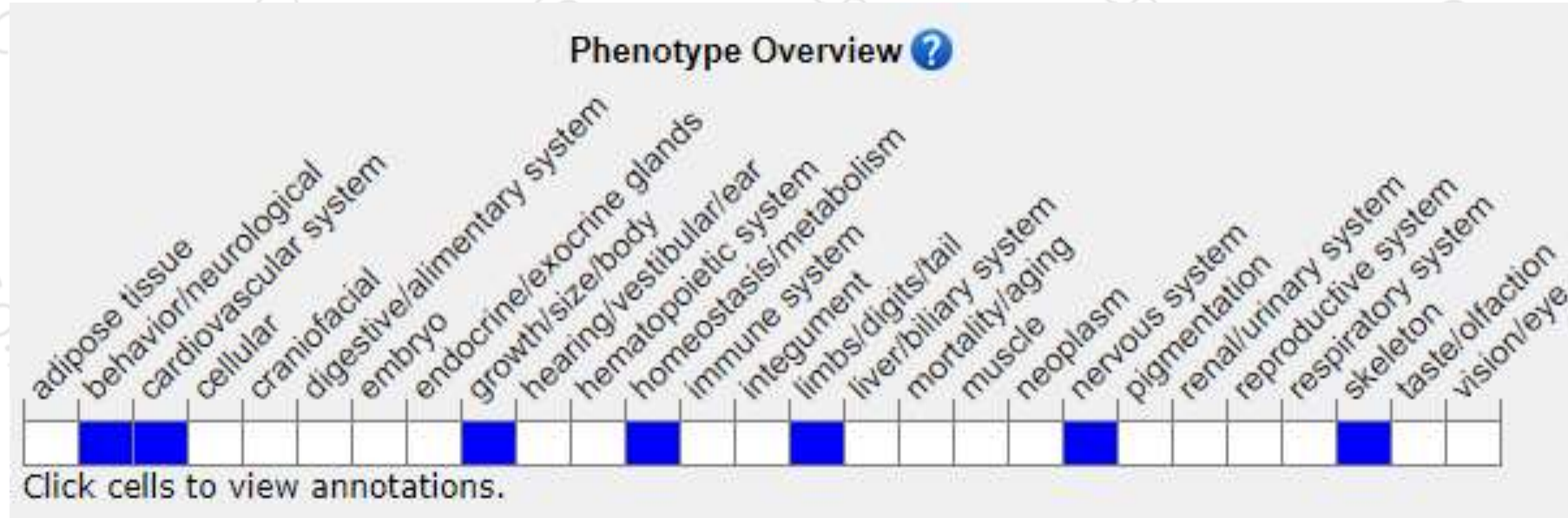
Genomic location distribution



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, Mice homozygous for a gene trapped allele show hyperactivity, unidirectional circling, sleep disturbances, thermal sensory alterations, increased serum iron levels, altered serotonin metabolism, enhanced long-term potentiation and paired-pulse ratios, and enhanced cued and contextual fear memory.

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