B4galt1 Cas9-CKO Strategy

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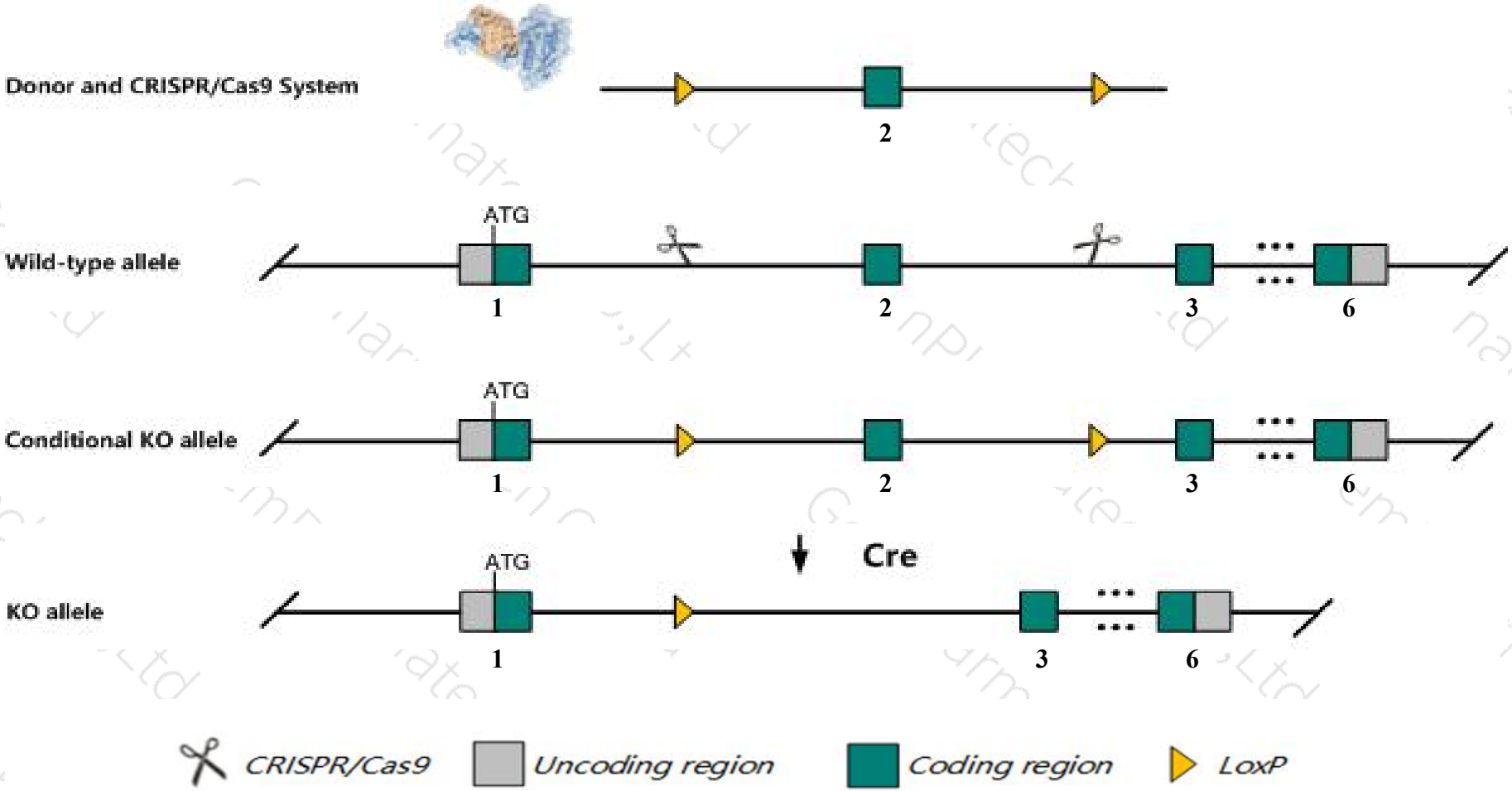
Project Overview

Project Name	***B4galt1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *B4galt1* gene. The schematic diagram is as follows:



Technical routes

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- The *B4galt1* gene has 2 transcripts. According to the structure of *B4galt1* gene, exon2 of *B4galt1-201* (ENSMUST00000030121.12) transcript is recommended as the knockout region.The region contains 236bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *B4galt1* 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,Homozygotes for targeted null mutations exhibit growth retardation, low viability, excessive epithelial cell proliferation of skin and small intestine, sperm with reduced fertilizing capacity, birthing difficulty, and mammary gland defects.

➢ The *B4galt1* gene is located on the Chr4. 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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B4galt1 UDP-Gal:betaGlcNAc beta 1,4- galactosyltransferase, polypeptide 1 [*Mus musculus* (house mouse)]

Gene ID: 14595, updated on 27-Aug-2019

Summary

Official Symbol	B4galt1 provided by MGI
Official Full Name	UDP-Gal:betaGlcNAc beta 1,4- galactosyltransferase, polypeptide 1 provided by MGI
Primary source	MGI:MGI:95705
See related	Ensembl:ENSMUSG00000028413
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	GalT; Ggtb; Ggtb2; b4Gal-T1; B-1,4-GalT; B-1,4-GalT1; beta4Gal-T1; beta-1,4-GalT; beta-1,4-GalT1
Summary	This gene encodes two distinct enzyme isoforms, a long membrane-bound form and a short soluble form. These alternate isoforms are thought to be produced through alternative nested transcription initiation and different in-frame start codon usage. These enzymes catalyze the transfer of galactose to acceptor sugars, such as N-acetylglucosamine and glucose. The long form of this enzyme is localized to the trans-Golgi membrane and is involved in glycoconjugate biosynthesis. The short form functions in lactose biosynthesis though formation of a heterodimer with alpha-lactalbumin. [provided by RefSeq, Nov 2012]
Expression	Ubiquitous expression in adrenal adult (RPKM 28.6), ovary adult (RPKM 25.6) and 25 other tissues See more
Orthologs	human all

Genomic context

Location: 4 A5; 4 20.46 cM

See B4galt1 in Genome Data Viewer

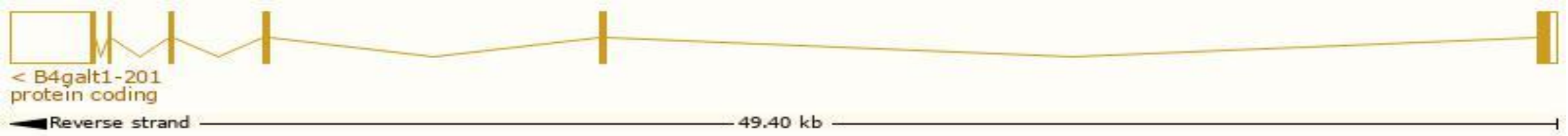
Exon count: 7

Transcript information（Ensembl）

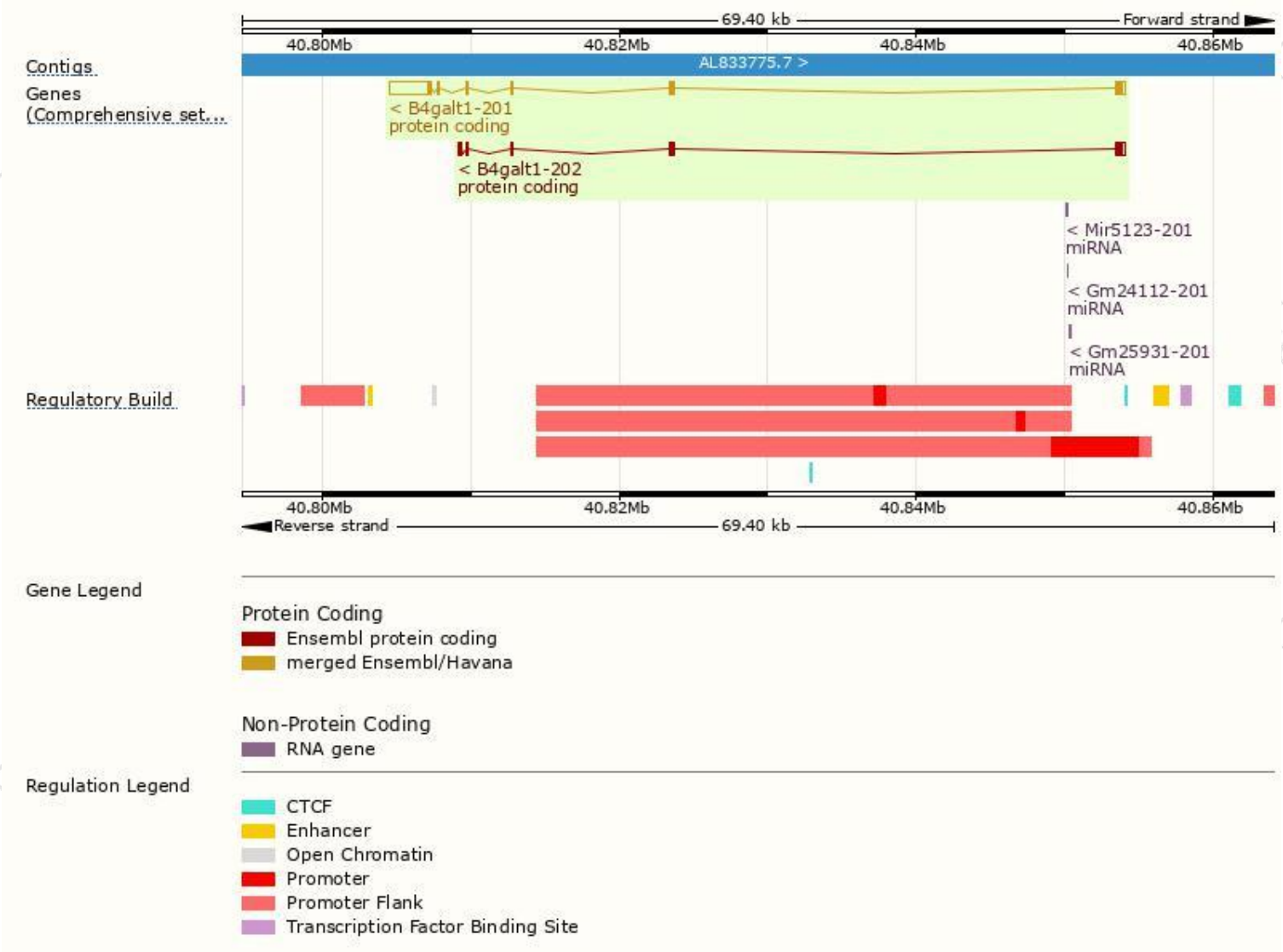
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
B4galt1-201	ENSMUST00000030121.12	3983	399aa	Protein coding	CCDS18051	P15535 Q3U478	TSL:1 GENCODE basic APPRIS P1
B4galt1-202	ENSMUST00000108096.2	1413	346aa	Protein coding	-	B1AXY5	TSL:1 GENCODE basic

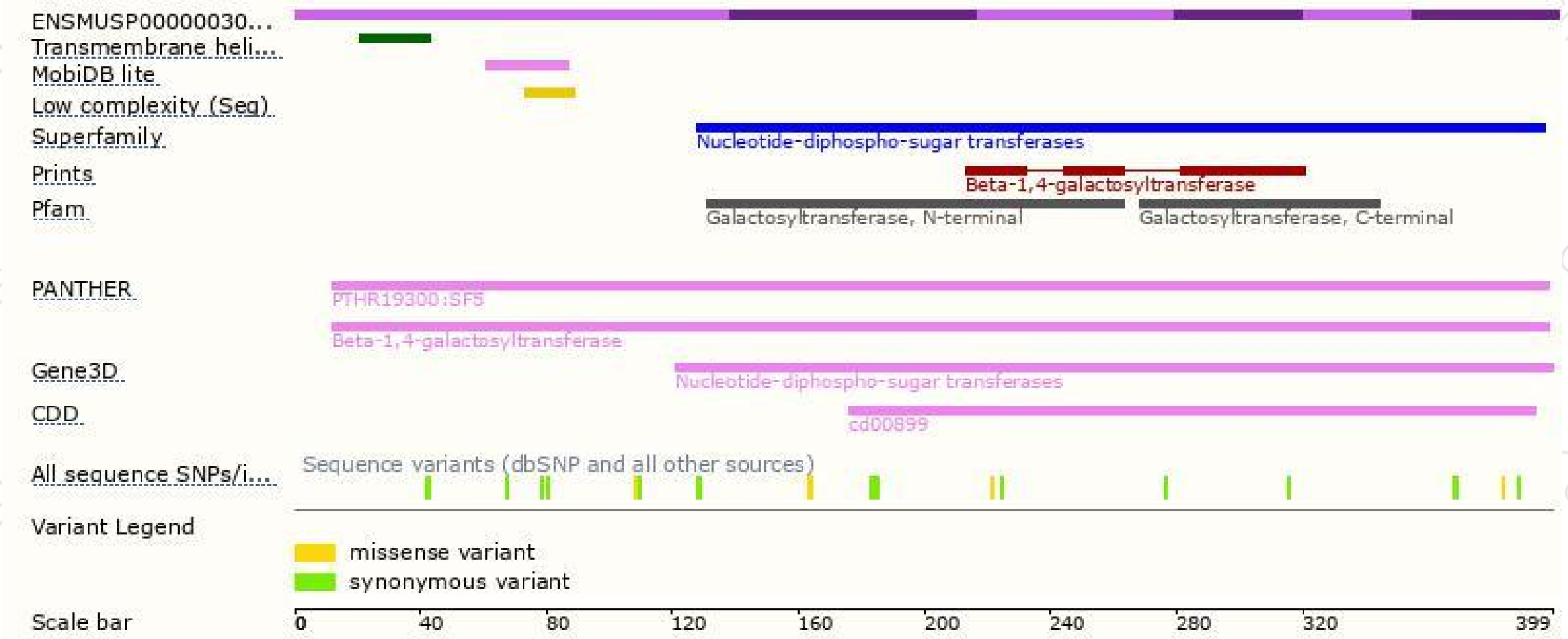
The strategy is based on the design of *B4galt1-201* transcript,The transcription is shown below



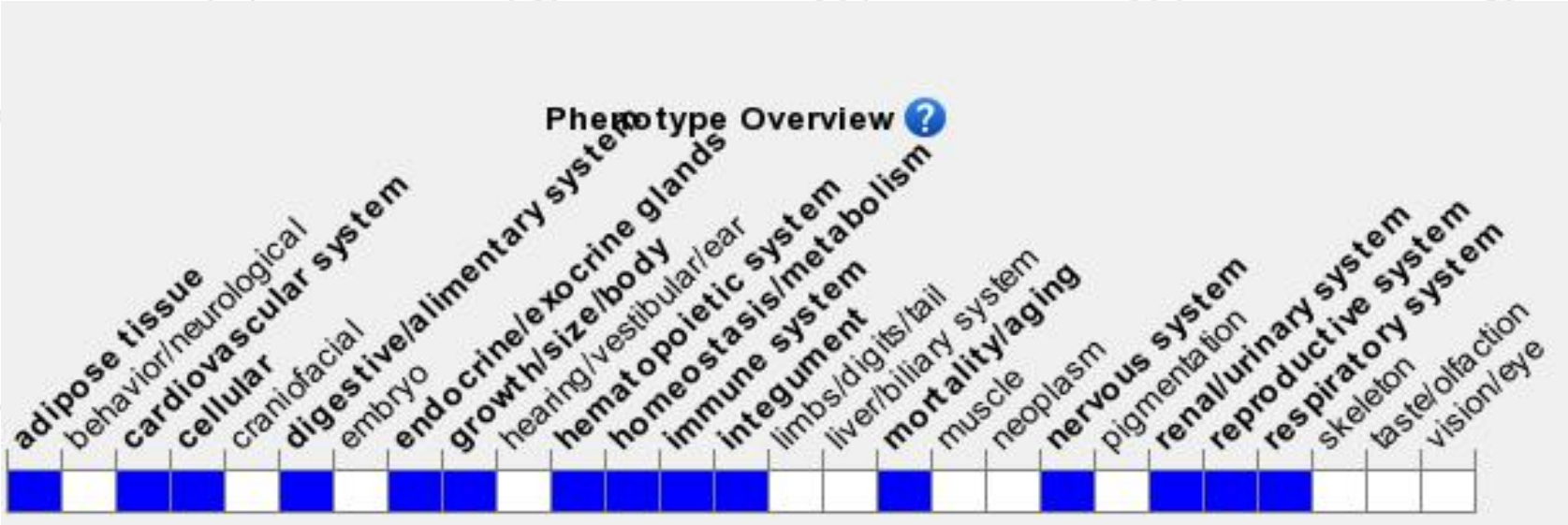
Genomic location distribution



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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