

A4galt Cas9-CKO Strategy

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Project Overview

Project Name

A4galt

Project type

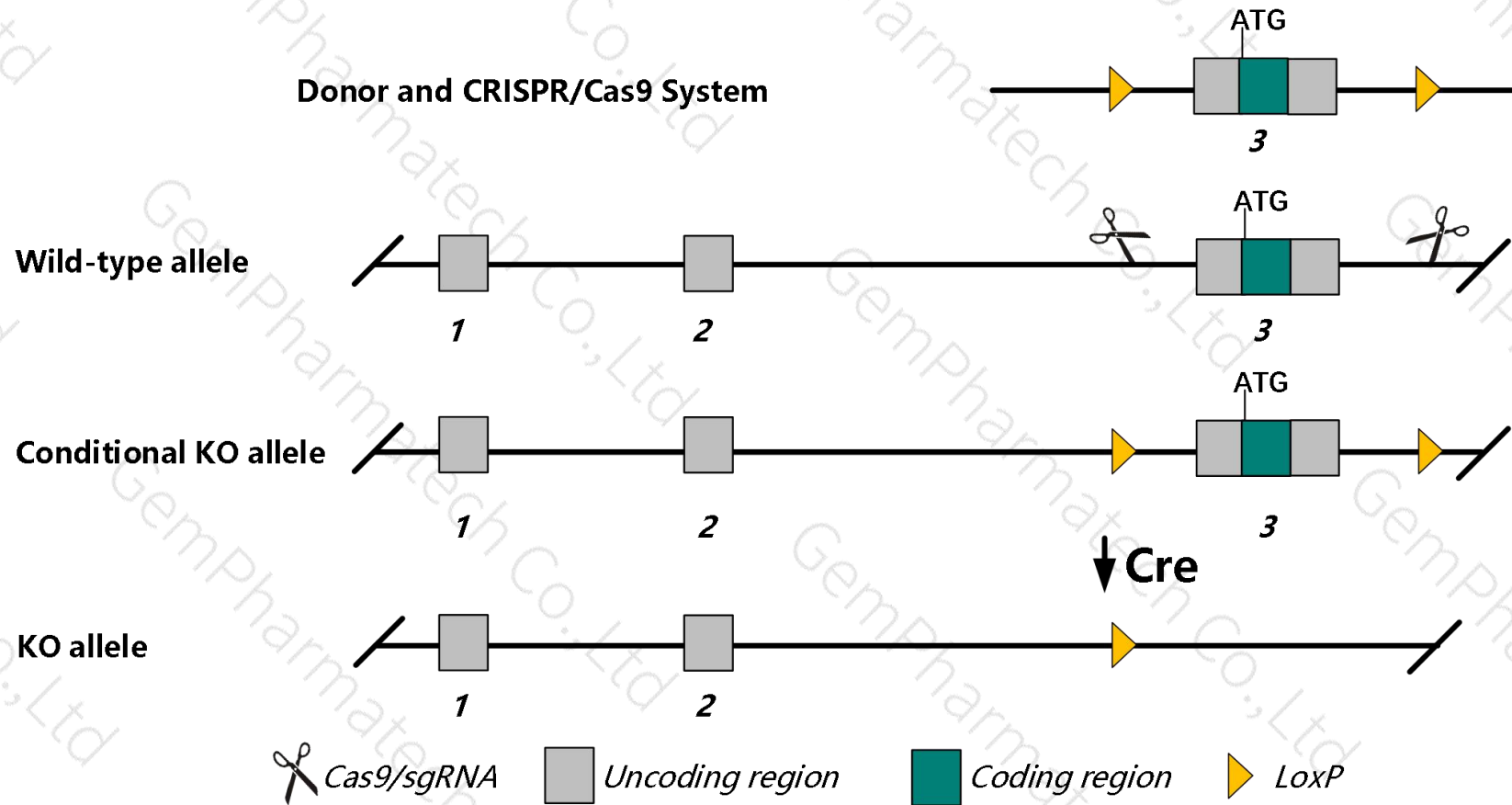
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *A4galt* gene. The schematic diagram is as follows:



- The *A4galt* gene has 4 transcripts. According to the structure of *A4galt* gene, exon3 of *A4galt-201* (ENSMUST00000049530.13) transcript is recommended as the knockout region. The region contains all 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 *A4galt* 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, Homozygous null mice display reduced sensitivity to verotoxin but do not display any gross morphological abnormalities.
- Transcript *A4galt*-203 may not be affected.
- The *A4galt* gene is located on the Chr15. 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)

A4galt alpha 1,4-galactosyltransferase [*Mus musculus* (house mouse)]

Gene ID: 239559, updated on 9-Feb-2020

Summary

- Official Symbol** A4galt provided by [MGI](#)
- Official Full Name** alpha 1,4-galactosyltransferase provided by [MGI](#)
- Primary source** [MGI:MGI:3512453](#)
- See related** [Ensembl:ENSMUSG000000047878](#)
- 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
- Summary** The protein encoded by this gene catalyzes the transfer of galactose to lactosylceramide to form globotriaosylceramide, which has been identified as the P(k) antigen of the P blood group system. The encoded protein, which is a type II membrane protein found in the Golgi, is also required for the synthesis of the bacterial verotoxins receptor. Alternative splicing results in multiple transcript variants.[provided by RefSeq, Jan 2010]
- Expression** Biased expression in ovary adult (RPKM 35.6), bladder adult (RPKM 35.2) and 14 other tissues [See more](#)
- Orthologs** [human](#) [all](#)

Genomic context

Location: 15; 15 E1 [See A4galt in Genome Data Viewer](#)

Exon count: 4

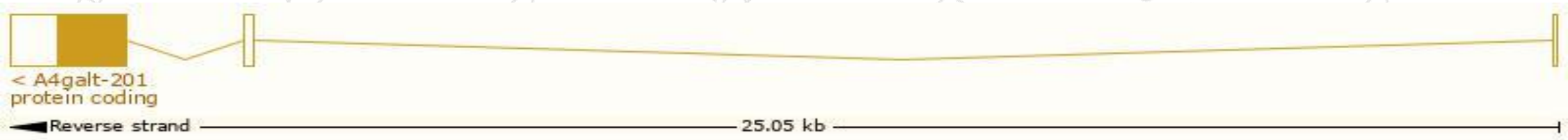
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	15	NC_000081.6 (83226722..83251729, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	15	NC_000081.5 (83057152..83082204, complement)

Transcript information (Ensembl)

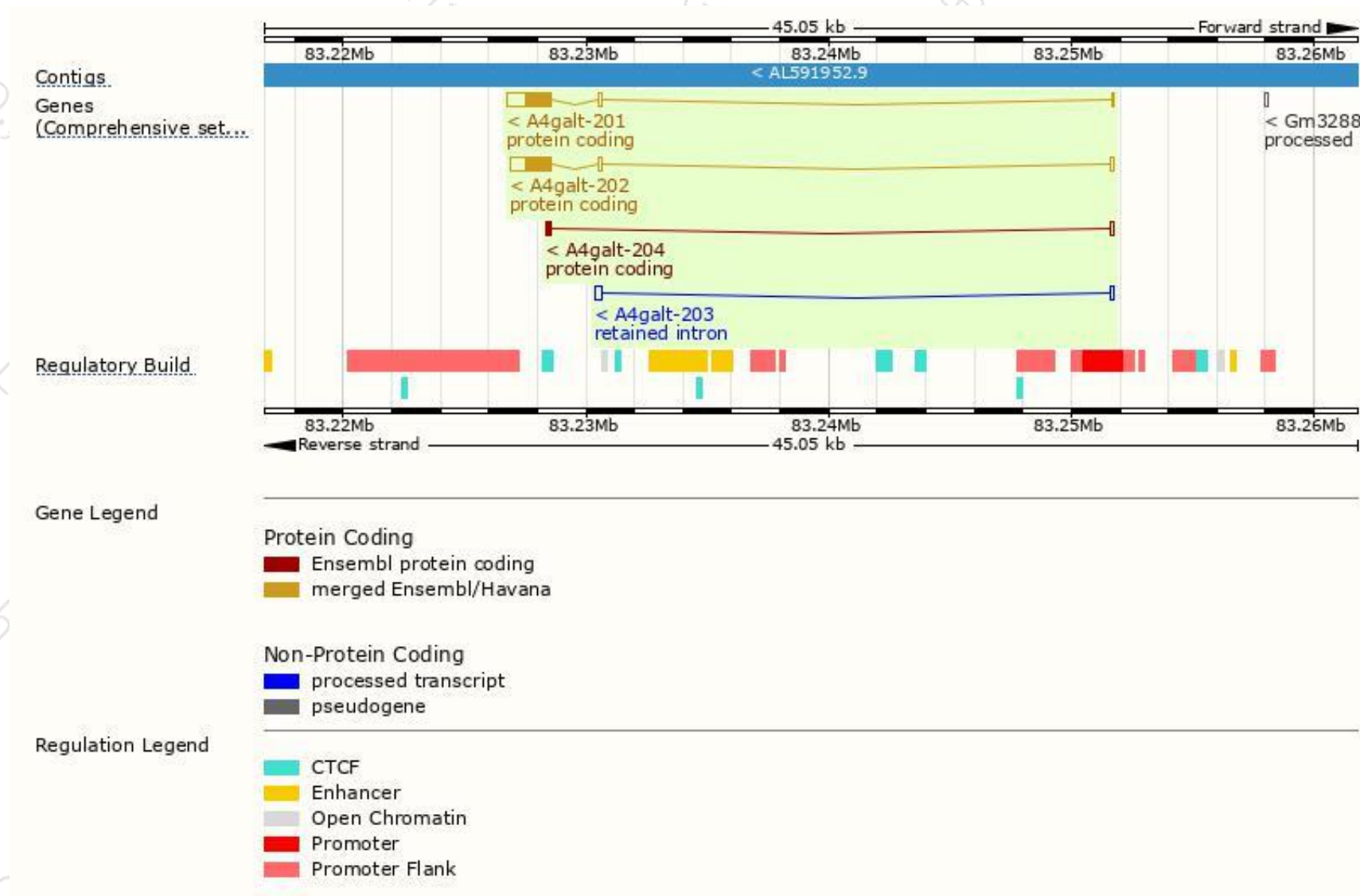
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
A4galt-201	ENSMUST00000049530.13	2092	359aa	Protein coding	CCDS27699	Q3UF00 Q67BJ4	TSL:1 GENCODE basic APPRIS P1
A4galt-202	ENSMUST00000164614.2	1974	359aa	Protein coding	CCDS27699	Q3UF00 Q67BJ4	TSL:1 GENCODE basic APPRIS P1
A4galt-204	ENSMUST00000229687.1	375	82aa	Protein coding	-	A0A2R8VI36	CDS 3' incomplete
A4galt-203	ENSMUST00000229454.1	360	No protein	Retained intron	-	-	

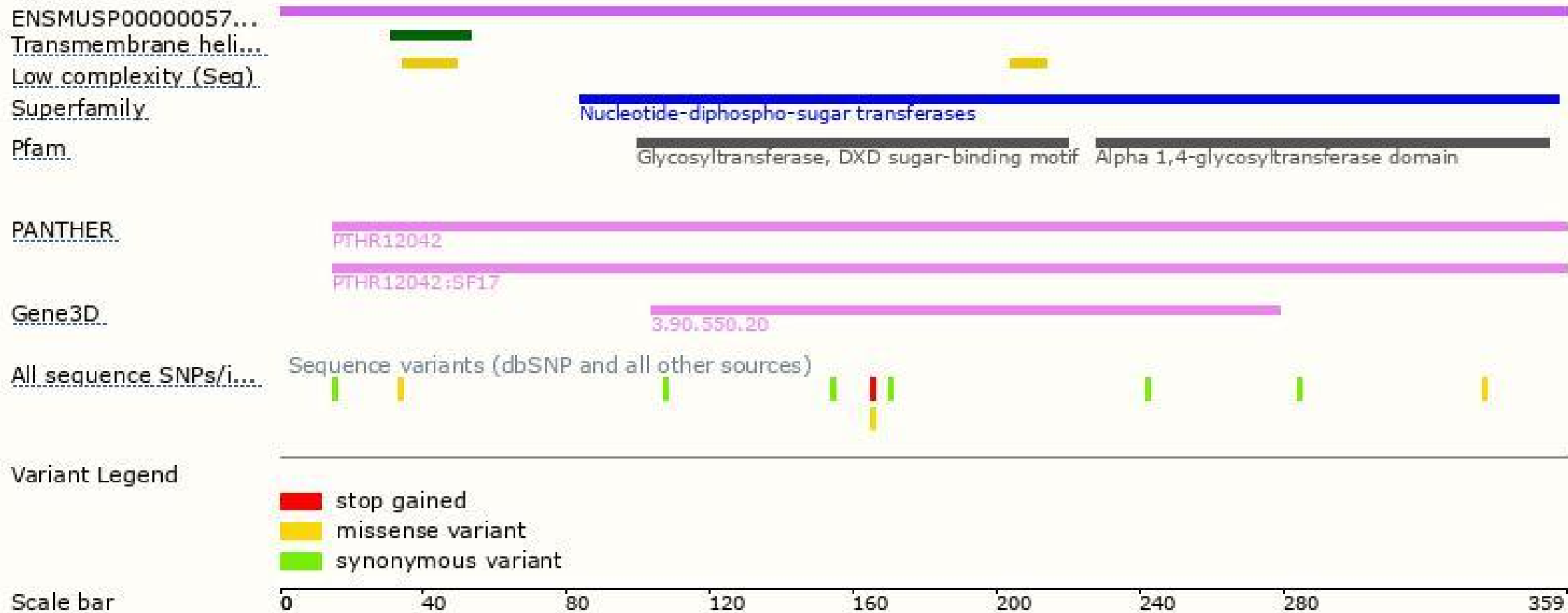
The strategy is based on the design of *A4galt-201* transcript,The transcription is shown below



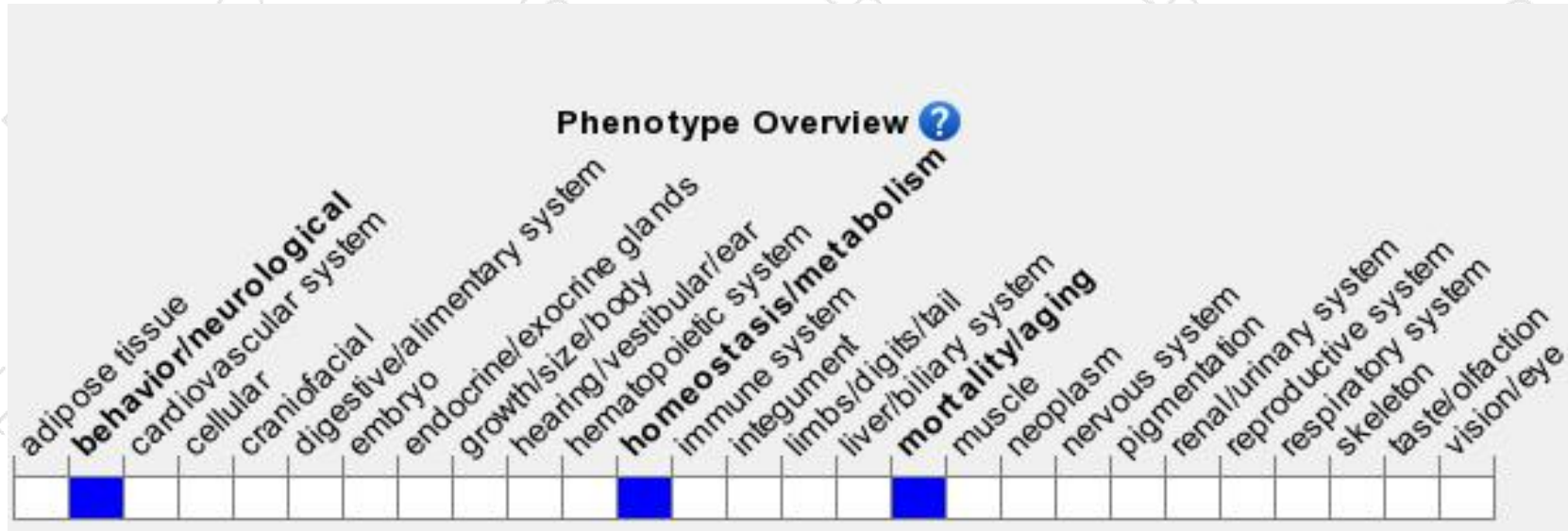
Genomic location distribution



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/>).

According to the existing MGI data, Homozygous null mice display reduced sensitivity to verotoxin but do not display any gross morphological abnormalities.

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

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