Grb14 Cas9-CKO Strategy

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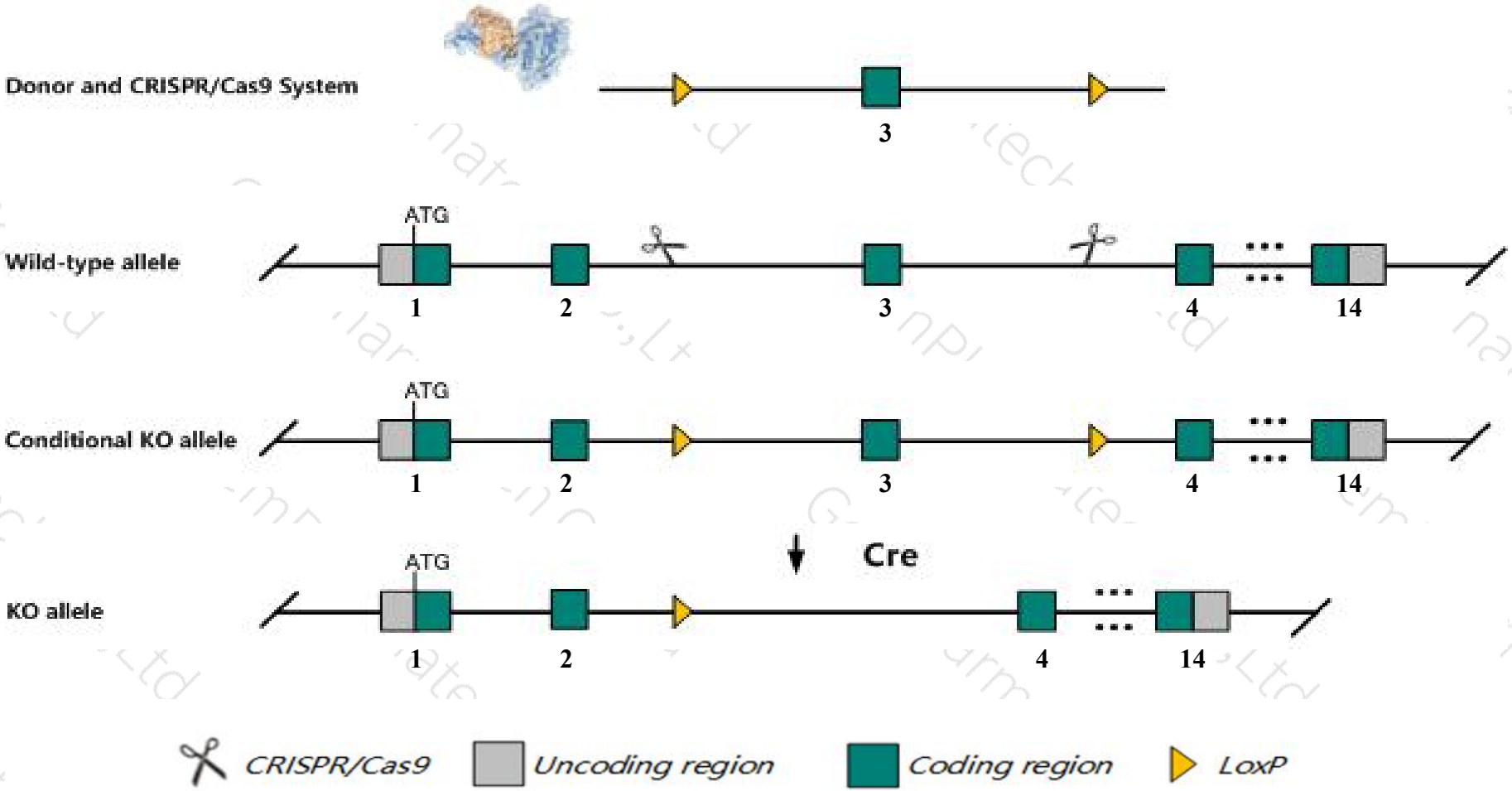
Project Overview

Project Name	***Grb14***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Grb14* gene. The schematic diagram is as follows:



Technical routes

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- The *Grb14* gene has 8 transcripts. According to the structure of *Grb14* gene, exon3 of *Grb14-201* (ENSMUST00000028252.13) transcript is recommended as the knockout region.The region contains 157bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Grb14* 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 mutation of this gene results in improved glucose tolerance, lower circulating insulin levels and increased incorporation of glucose into glycogen in the liver and skeletal muscle of males. Both males and females exhibit a decrease in body size.

➢ Transcript 206,207 CDS 5' incomplete the influences is unknown.

➢ The *Grb14* gene is located on the Chr2. 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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Grb14 growth factor receptor bound protein 14 [Mus musculus (house mouse)]

Gene ID: 50915, updated on 31-Jan-2019

Summary

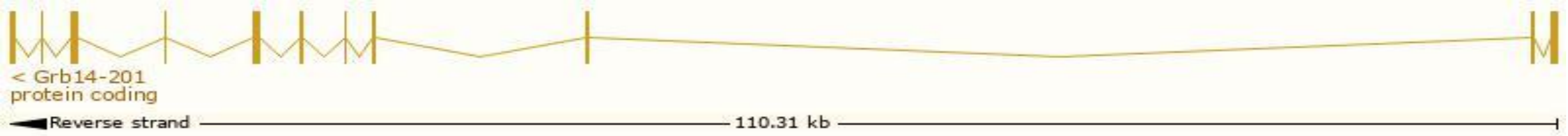
Official Symbol	Grb14 provided by MGI
Official Full Name	growth factor receptor bound protein 14 provided by MGI
Primary source	MGI:MGI:1355324
See related	Ensembl:ENSMUSG00000026888
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI505286
Expression	Broad expression in heart adult (RPKM 19.5), ovary adult (RPKM 12.9) and 18 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

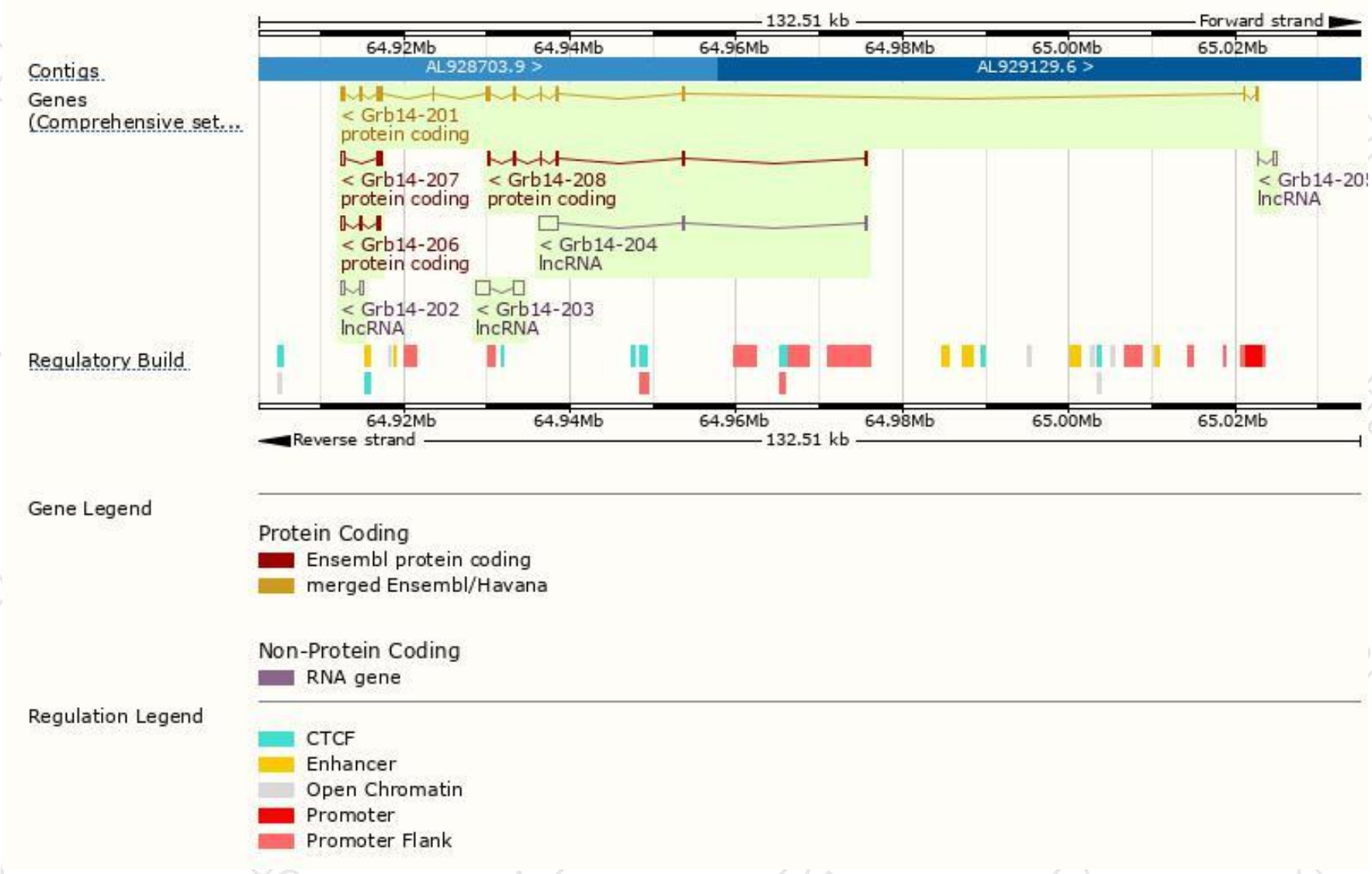
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Grb14-201	ENSMUST00000028252.13	1986	538aa	Protein coding	CCDS16071	A2ASX2 Q9JLM9	TSL:1 GENCODE basic APPRIS P1
Grb14-208	ENSMUST00000156765.1	813	221aa	Protein coding	-	A2ASX1	CDS 3' incomplete TSL:5
Grb14-206	ENSMUST00000149193.7	697	109aa	Protein coding	-	A2ASX4	CDS 5' incomplete TSL:5
Grb14-207	ENSMUST00000150643.7	656	93aa	Protein coding	-	A2ASX3	CDS 5' incomplete TSL:3
Grb14-203	ENSMUST00000137085.1	2797	No protein	lncRNA	-	-	TSL:1
Grb14-204	ENSMUST00000145603.1	2547	No protein	lncRNA	-	-	TSL:1
Grb14-202	ENSMUST00000134020.1	682	No protein	lncRNA	-	-	TSL:2
Grb14-205	ENSMUST00000146807.1	380	No protein	lncRNA	-	-	TSL:5

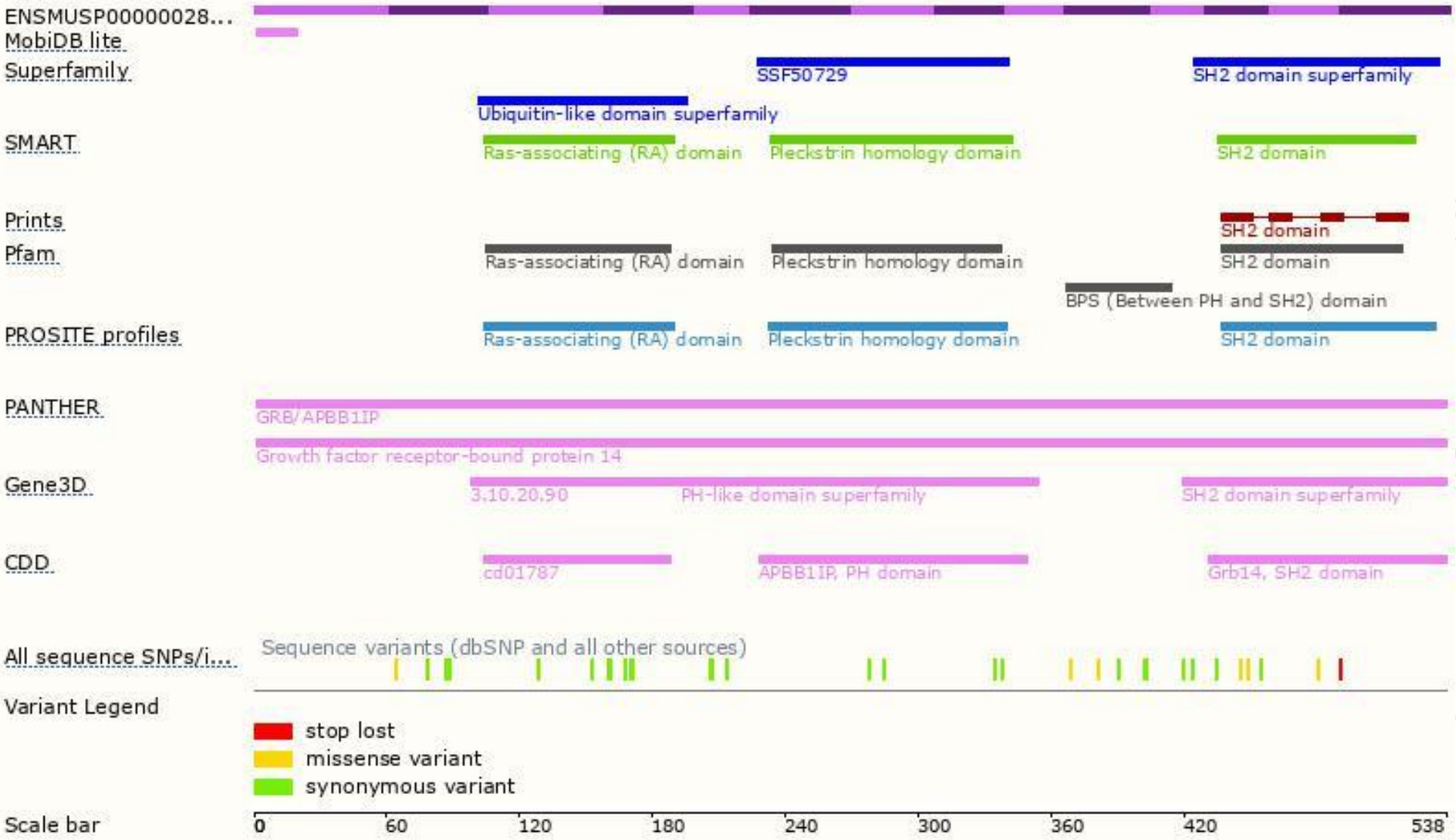
The strategy is based on the design of *Grb14-201* transcript,The transcription is shown below



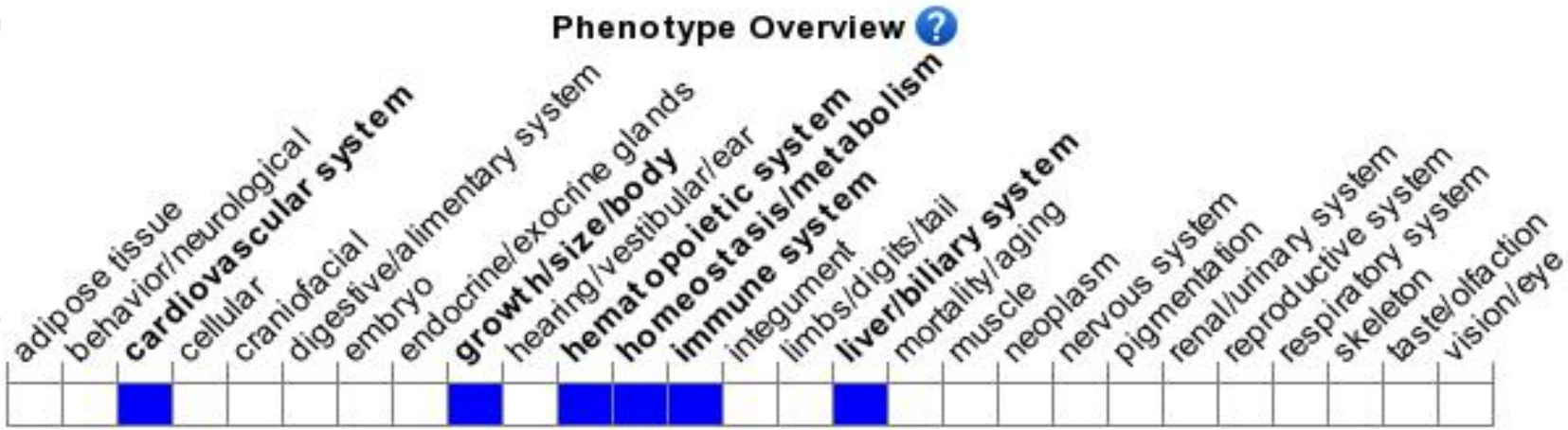
Genomic location distribution



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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