

***Rgs6* Cas9-CKO Strategy**

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Reviewer:

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Project Overview

Project Name

Rgs6

Project type

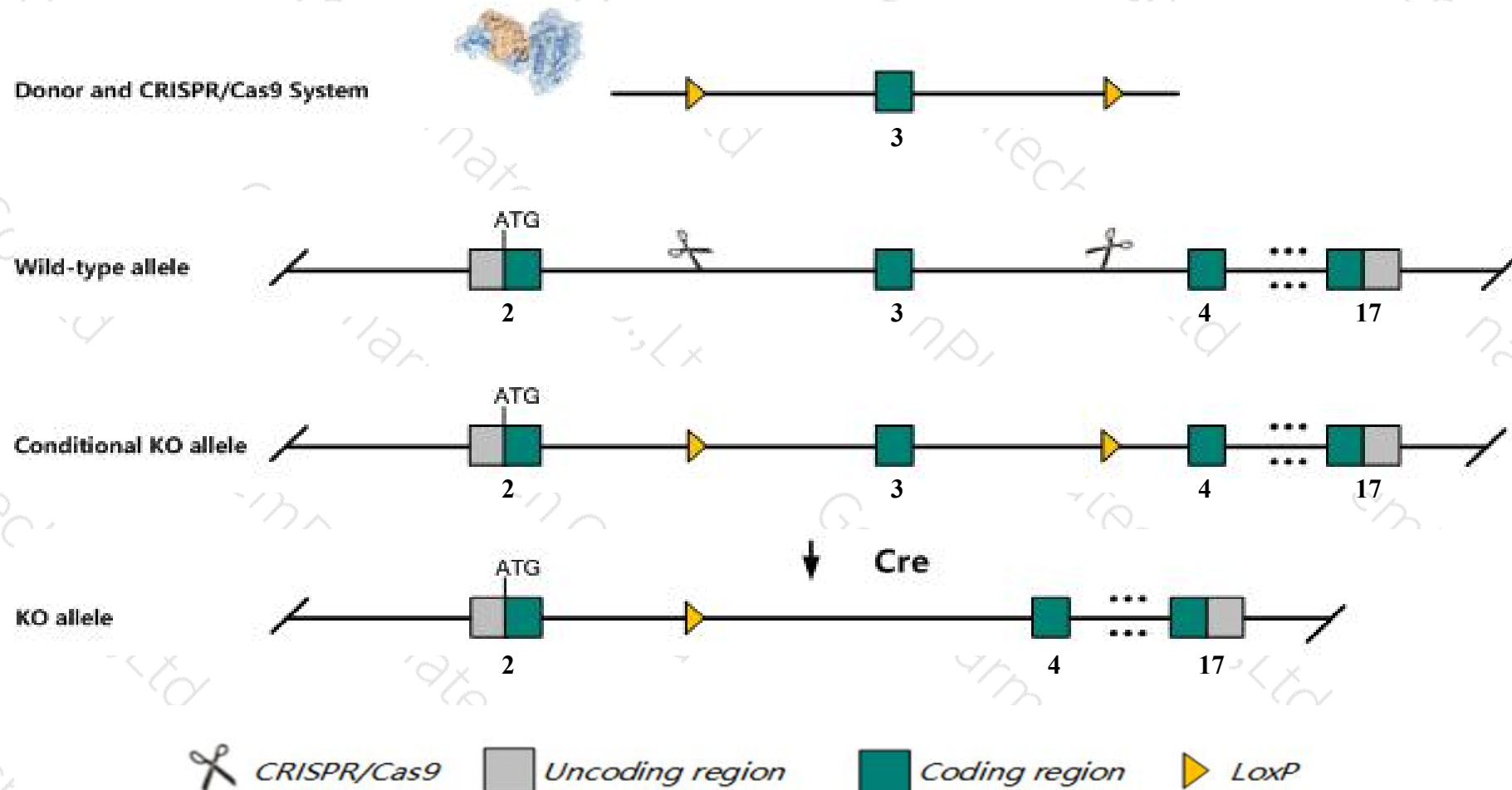
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rgs6* gene. The schematic diagram is as follows:



- The *Rgs6* gene has 22 transcripts. According to the structure of *Rgs6* gene, exon3 of *Rgs6-208* (ENSMUST00000186458.6) transcript is recommended as the knockout region. The region contains 100bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rgs6* 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, Mice homozygous for a knock-out allele exhibit decreased heart rate and abnormal impulse conducting system conduction.
- The *Rgs6* gene is located on the Chr12. 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)

Rgs6 regulator of G-protein signaling 6 [Mus musculus (house mouse)]

Gene ID: 50779, updated on 31-Jan-2019

Summary



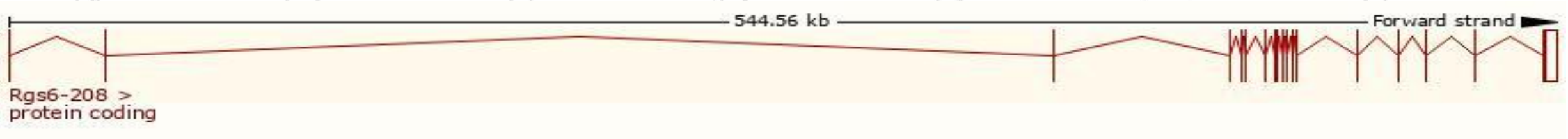
Official Symbol	Rgs6 provided by MGI
Official Full Name	regulator of G-protein signaling 6 provided by MGI
Primary source	MGI:MGI:1354730
See related	Ensembl:ENSMUSG000000021219
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Summary	This gene encodes a member of the RGS (regulator of G protein signaling) family of proteins, which are defined by the presence of a RGS domain that confers the GTPase-activating activity of these proteins toward certain G alpha subunits. This protein also belongs to a subfamily of RGS proteins characterized by the presence of DEP (Dishevelled, Egl-10, and Pleckstrin) and GGL (G-protein gamma like) domains, the latter a G beta 5-interacting domain. The RGS proteins negatively regulate G protein signaling, and may modulate neuronal, cardiovascular, lymphocytic activities, and cancer risk. Mice lacking this gene exhibit decreased heart rate. Alternative splicing results in multiple transcript variants, however, the full-length nature of some of these variants is not known. [provided by RefSeq, Sep 2015]
Expression	Biased expression in cortex adult (RPKM 4.8), frontal lobe adult (RPKM 3.7) and 10 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

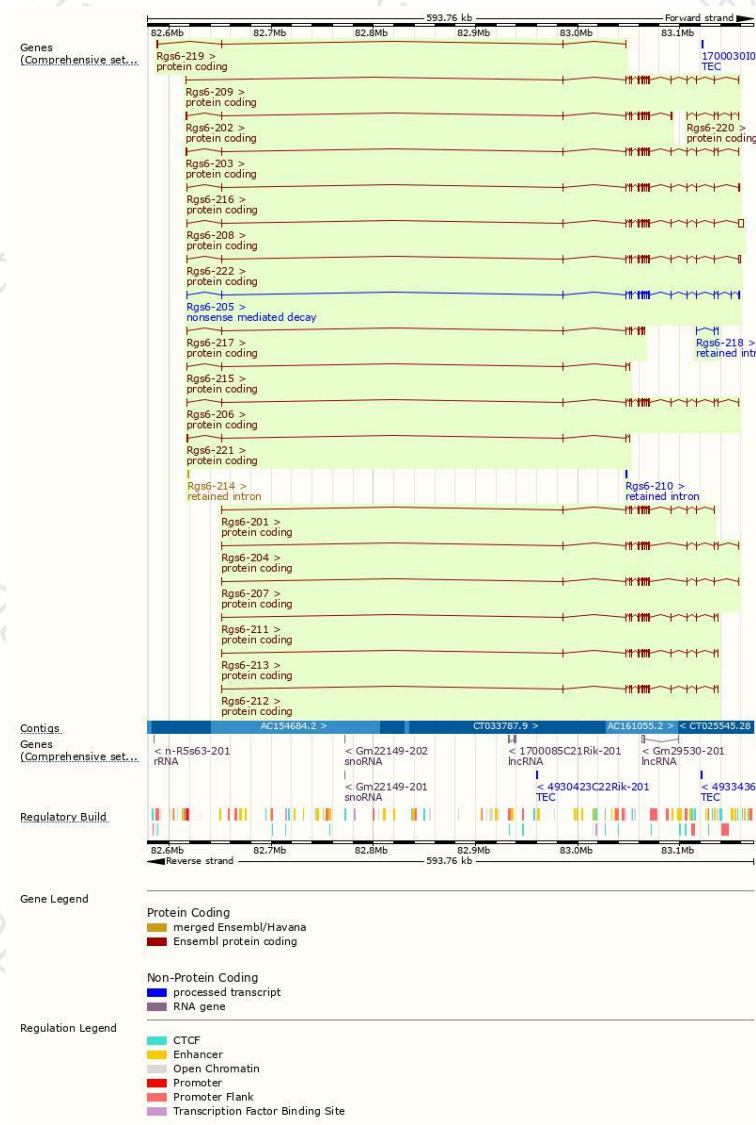
The gene has 22 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rgs6-208	ENSMUST00000186458.6	5588	472aa	Protein coding	CCDS79132	Q9Z2H2	TSL:1 GENCODE basic APPRIS ALT:1
Rgs6-222	ENSMUST00000202210.3	3069	472aa	Protein coding	CCDS79132	Q9Z2H2	TSL:5 GENCODE basic APPRIS ALT:1
Rgs6-216	ENSMUST00000200911.3	2626	472aa	Protein coding	CCDS79132	Q9Z2H2	TSL:1 GENCODE basic APPRIS ALT:1
Rgs6-203	ENSMUST00000185665.6	2201	490aa	Protein coding	CCDS79131	A0A087WNZ9	TSL:5 GENCODE basic APPRIS P:4
Rgs6-206	ENSMUST00000186309.6	1693	490aa	Protein coding	CCDS79131	A0A087WNZ9	TSL:5 GENCODE basic APPRIS P:4
Rgs6-202	ENSMUST00000161801.7	2141	368aa	Protein coding	-	Q3UTM5	TSL:1 GENCODE basic
Rgs6-211	ENSMUST00000191107.6	1621	505aa	Protein coding	-	A0A087WPD1	TSL:5 GENCODE basic
Rgs6-213	ENSMUST00000191352.6	1507	502aa	Protein coding	-	A0A087WPC4	CDS 3' incomplete TSL:5
Rgs6-212	ENSMUST00000191311.6	1486	495aa	Protein coding	-	A0A087WRP8	CDS 3' incomplete TSL:5
Rgs6-209	ENSMUST00000186848.4	1472	437aa	Protein coding	-	A0A087WSH2	TSL:5 GENCODE basic
Rgs6-201	ENSMUST00000101234.8	1368	456aa	Protein coding	-	F6WDS8	TSL:5 GENCODE basic
Rgs6-204	ENSMUST00000185674.6	1362	453aa	Protein coding	-	A0A087WPW1	TSL:5 GENCODE basic
Rgs6-207	ENSMUST00000186323.6	1308	435aa	Protein coding	-	A0A087WSJ9	TSL:5 GENCODE basic
Rgs6-220	ENSMUST00000201767.1	782	221aa	Protein coding	-	A0A0J9YTS6	CDS 5' incomplete TSL:5
Rgs6-217	ENSMUST00000201271.3	663	206aa	Protein coding	-	A0A0J9YUE4	CDS 3' incomplete TSL:5
Rgs6-221	ENSMUST00000201861.3	535	88aa	Protein coding	-	A0A0J9YUY2	CDS 3' incomplete TSL:5
Rgs6-215	ENSMUST00000200861.3	525	109aa	Protein coding	-	A0A0J9YUD1	CDS 3' incomplete TSL:3
Rgs6-219	ENSMUST00000201602.3	426	72aa	Protein coding	-	A0A0J9YU74	CDS 3' incomplete TSL:5
Rgs6-205	ENSMUST00000186081.6	2091	465aa	Nonsense mediated decay	-	A0A087WQG6	TSL:5
Rgs6-210	ENSMUST00000190964.1	1131	No protein	Retained intron	-	-	TSL:NA
Rgs6-214	ENSMUST00000193448.1	968	No protein	Retained intron	-	-	TSL:NA
Rgs6-218	ENSMUST00000201512.1	403	No protein	Retained intron	-	-	TSL:5

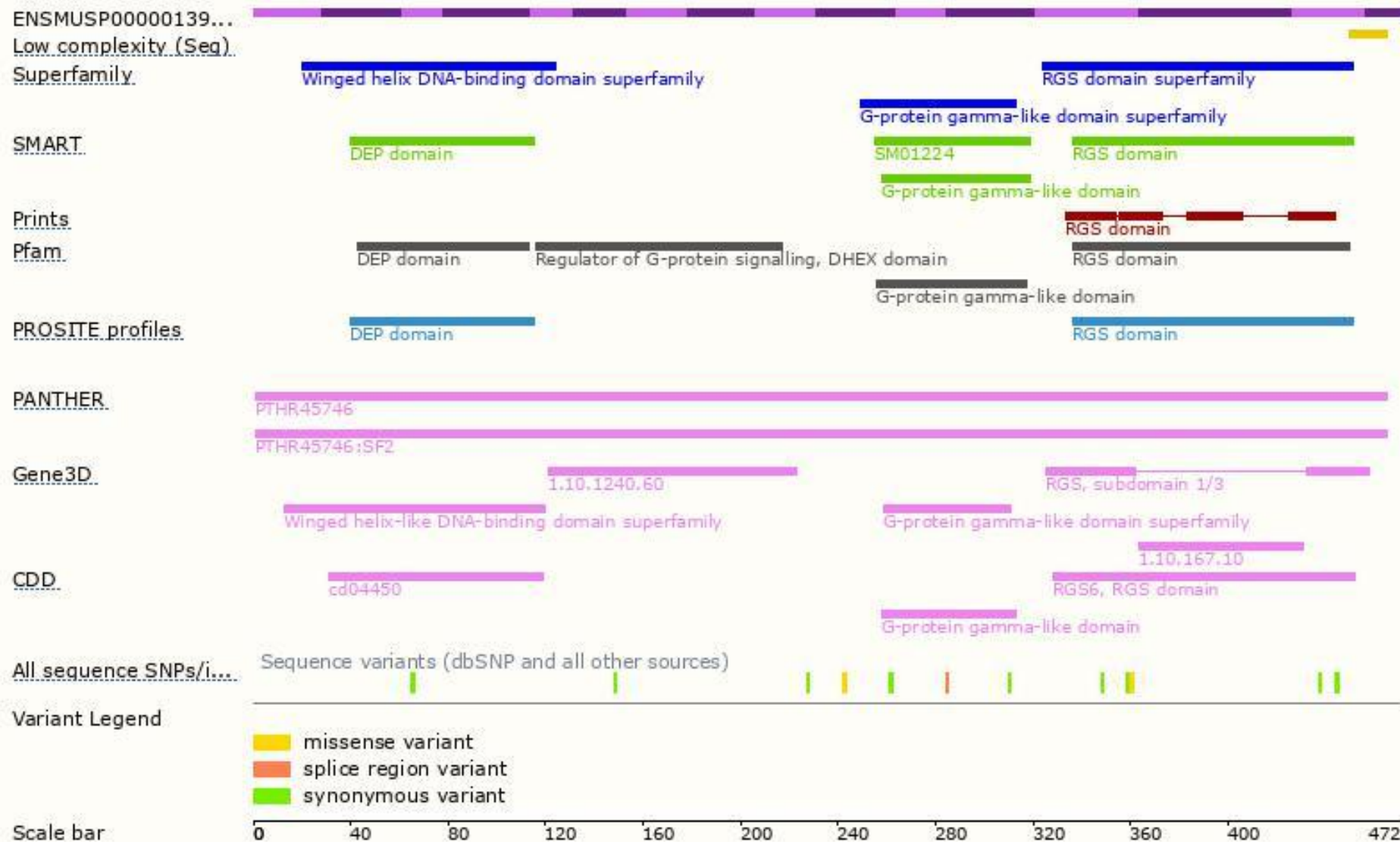
The strategy is based on the design of *Rgs6-208* transcript,The transcription is shown below



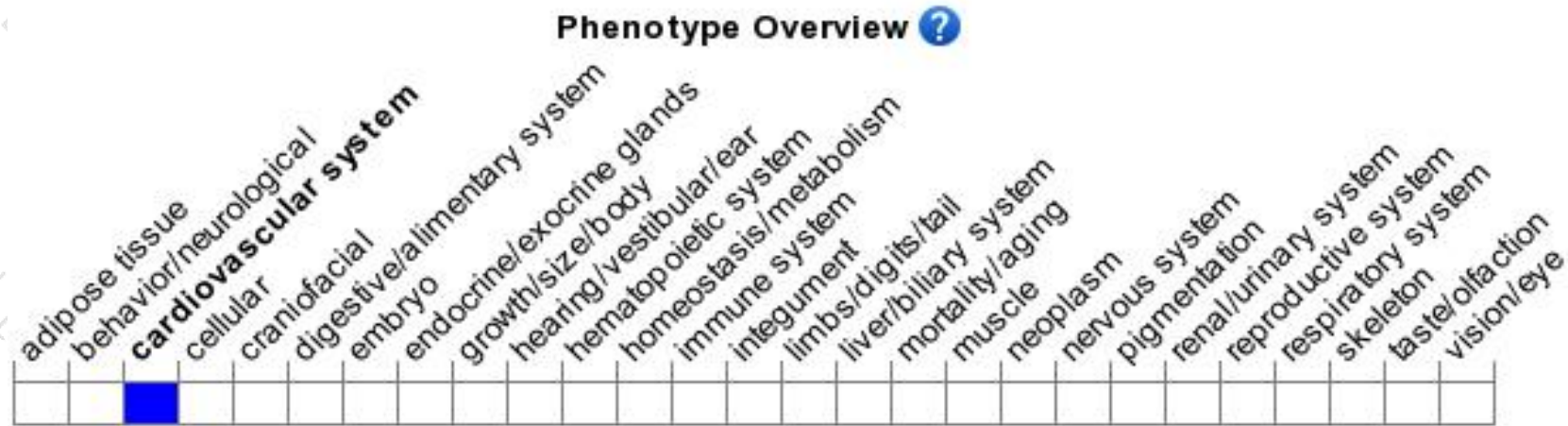
Genomic location distribution



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 knock-out allele exhibit decreased heart rate and abnormal impulse conducting system conduction.

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

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