Des Cas9-CKO Strategy

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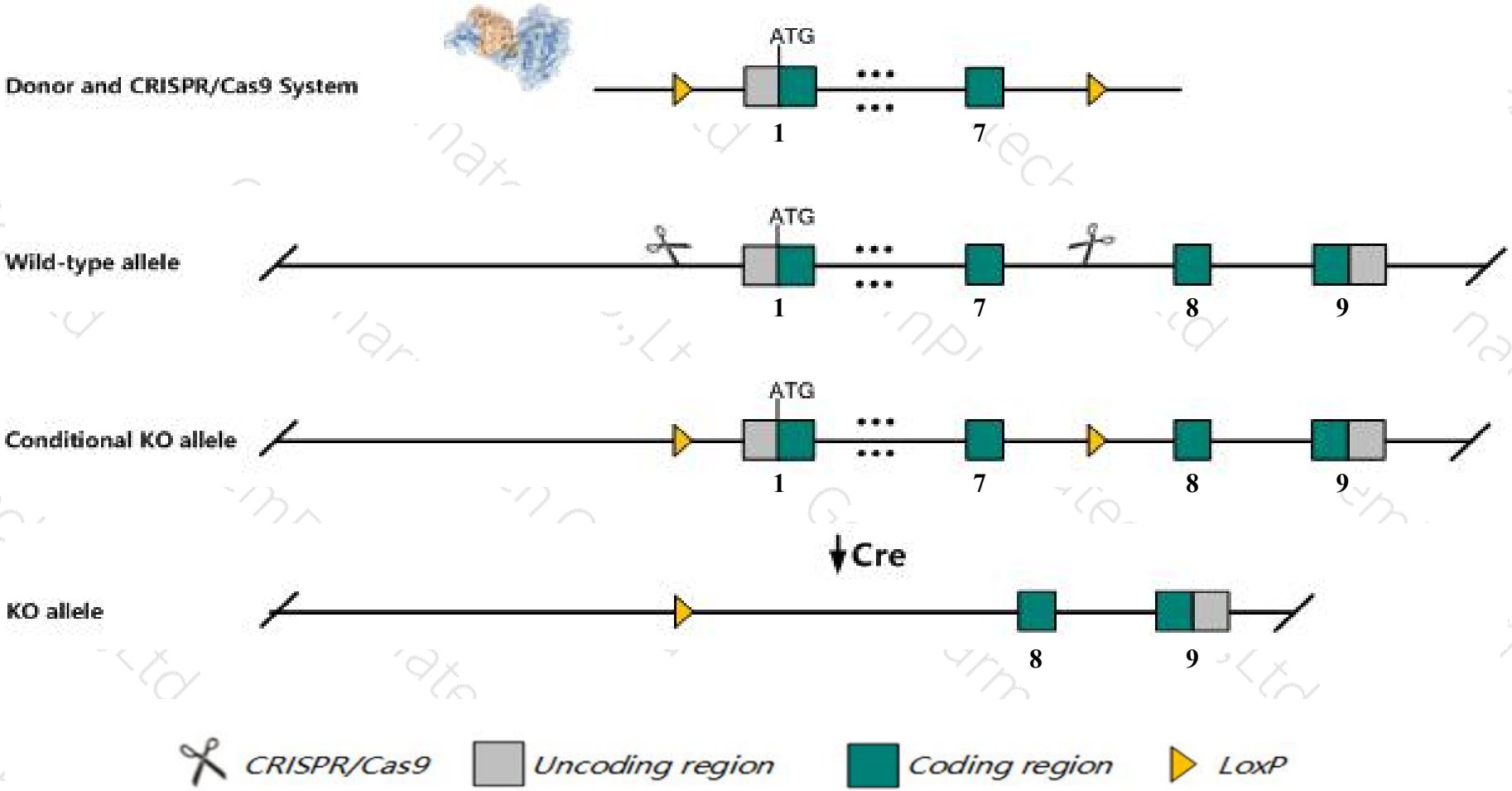
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Des* gene has 3 transcripts. According to the structure of *Des* gene, exon1-exon7 of *Des-201* (ENSMUST00000027409.9) transcript is recommended as the knockout region.The region contains start codon ATG. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Des* 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 histologically detectable defects of cardiac, skeletal, and smooth muscle. Defects in the heart are most severe, and lead to calcification, progressive degeneration, and necrosis of the myocardium.

- The *Des* gene is located on the Chr1. 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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Des desmin [*Mus musculus* (house mouse)]

Gene ID: 13346, updated on 1-Oct-2019

Summary

Official Symbol	Des provided by MGI
Official Full Name	desmin provided by MGI
Primary source	MGI:MGI:94885
See related	Ensembl:ENSMUSG00000026208
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 muscle-specific class III intermediate filament. Homopolymers of this protein form a stable intracytoplasmic filamentous network connecting myofibrils to each other and to the plasma membrane and are essential for maintaining the strength and integrity of skeletal, cardiac and smooth muscle fibers. Mutations in this gene affect assembly of intermediate filaments. Mice lacking this gene are able to develop and reproduce but exhibit abnormal muscle fibers. Mutations in the human gene are associated with myofibrillar myopathy, dilated cardiomyopathy, neurogenic scapuloperoneal syndrome and autosomal recessive limb-girdle muscular dystrophy, type 2R. [provided by RefSeq, Jan 2014]
Expression	Biased expression in bladder adult (RPKM 808.0), stomach adult (RPKM 385.6) and 10 other tissues See more
Orthologs	human all

Genomic context

Location: 1 C4; 1 38.85 cM

See Des in Genome Data Viewer

Exon count: 9

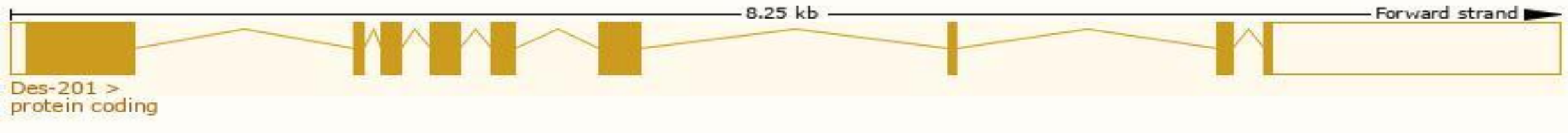
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	1	NC_000067.6 (75360292..75368579)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	1	NC_000067.5 (75356919..75364291)

Transcript information（Ensembl）

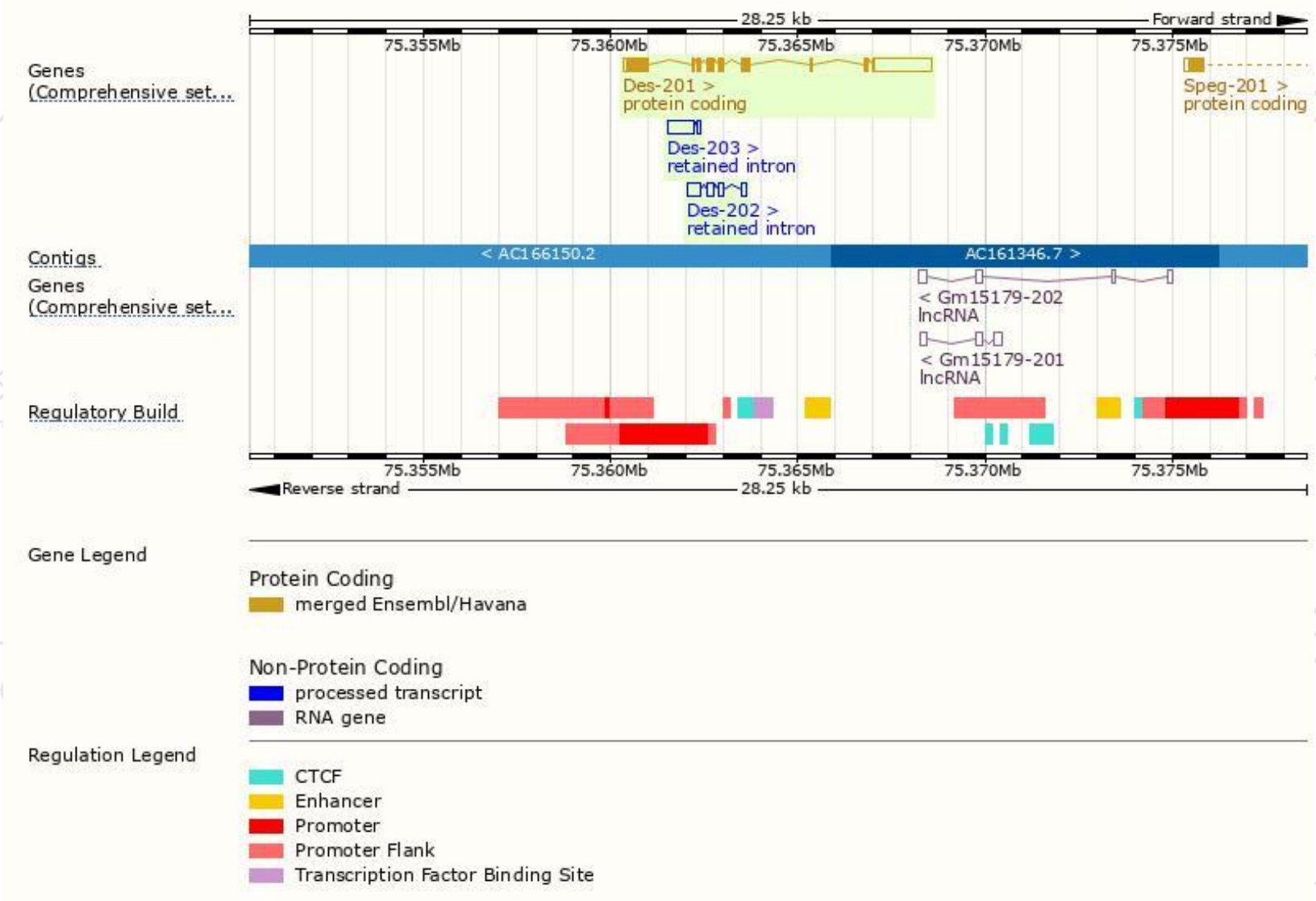
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Des-201	ENSMUST00000027409.9	3028	469aa	Protein coding	CCDS15071	P31001 Q3V1K9	TSL:1 GENCODE basic APPRIS P1
Des-202	ENSMUST00000125948.1	835	No protein	Retained intron	-	-	TSL:2
Des-203	ENSMUST00000144894.1	784	No protein	Retained intron	-	-	TSL:3

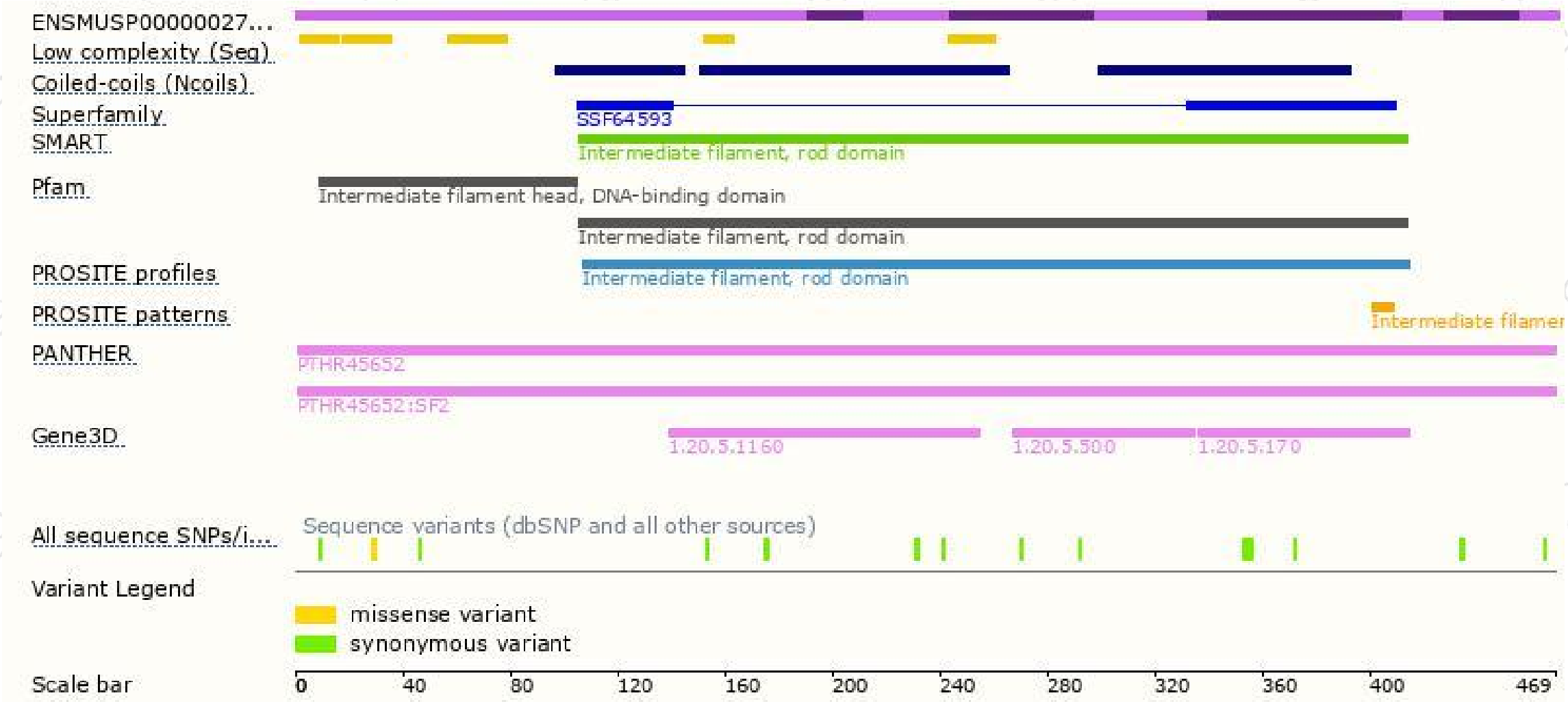
The strategy is based on the design of *Des-201* transcript,The transcription is shown below



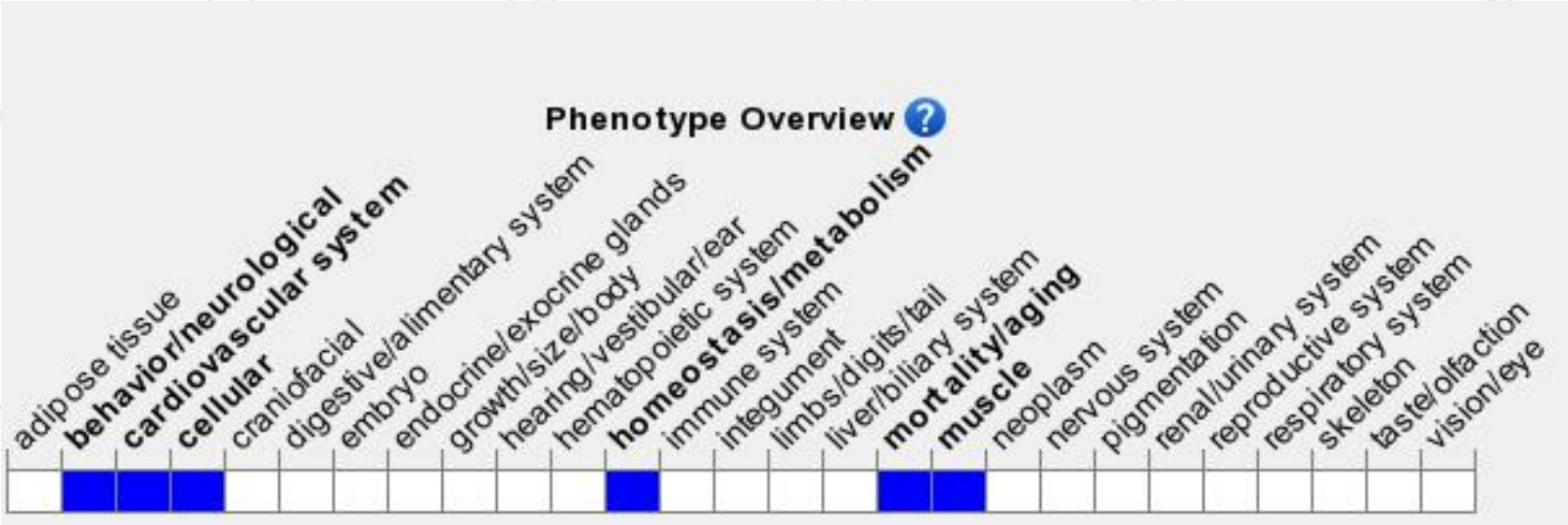
Genomic location distribution



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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