Dnm2 Cas9-CKO Strategy

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Project Overview

Project Name	***Dnm2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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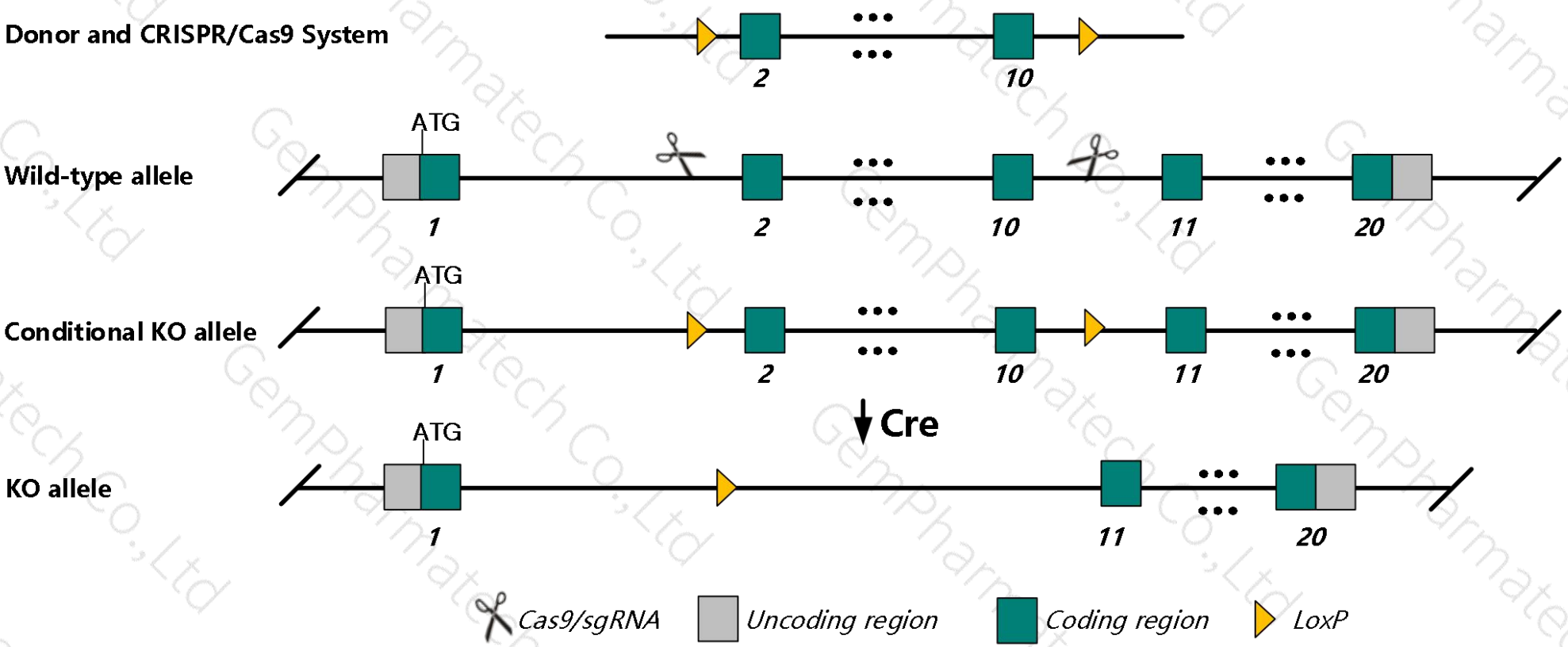
This model will use CRISPR/Cas9 technology to edit the *Dnm2* gene. The schematic diagram is as follows:



Technical routes

- The *Dnm2* gene has 14 transcripts. According to the structure of *Dnm2* gene, exon2-exon10 of *Dnm2-202* (ENSMUST00000091087.12) transcript is recommended as the knockout region.The region contains 1174bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Dnm2* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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,Mice homozygous for a targeted allele die prior to E8-E12. Mice heterozygous for a knock-out allele exhibit muscle atrophy and weakness, intermyofibrillar disorganization, and centrally localized mitochondria and sarcoplasmic reticulum.

➢ The *Dnm2* gene is located on the Chr9. 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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Dnm2 dynamin 2 [*Mus musculus* (house mouse)]

Gene ID: 13430, updated on 14-Aug-2019

Summary

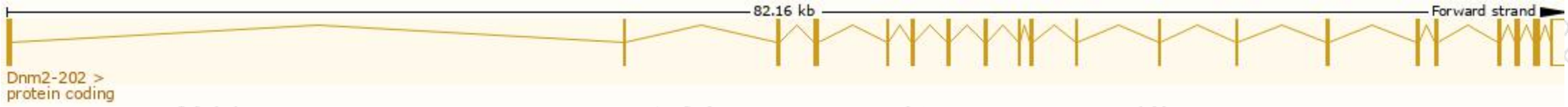
Official Symbol	Dnm2 provided by MGI
Official Full Name	dynamin 2 provided by MGI
Primary source	MGI:MGI:109547
See related	Ensembl:ENSMUSG00000033335
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Dyn2; Udnm; b2b2159Clo
Expression	Ubiquitous expression in duodenum adult (RPKM 68.5), colon adult (RPKM 65.8) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

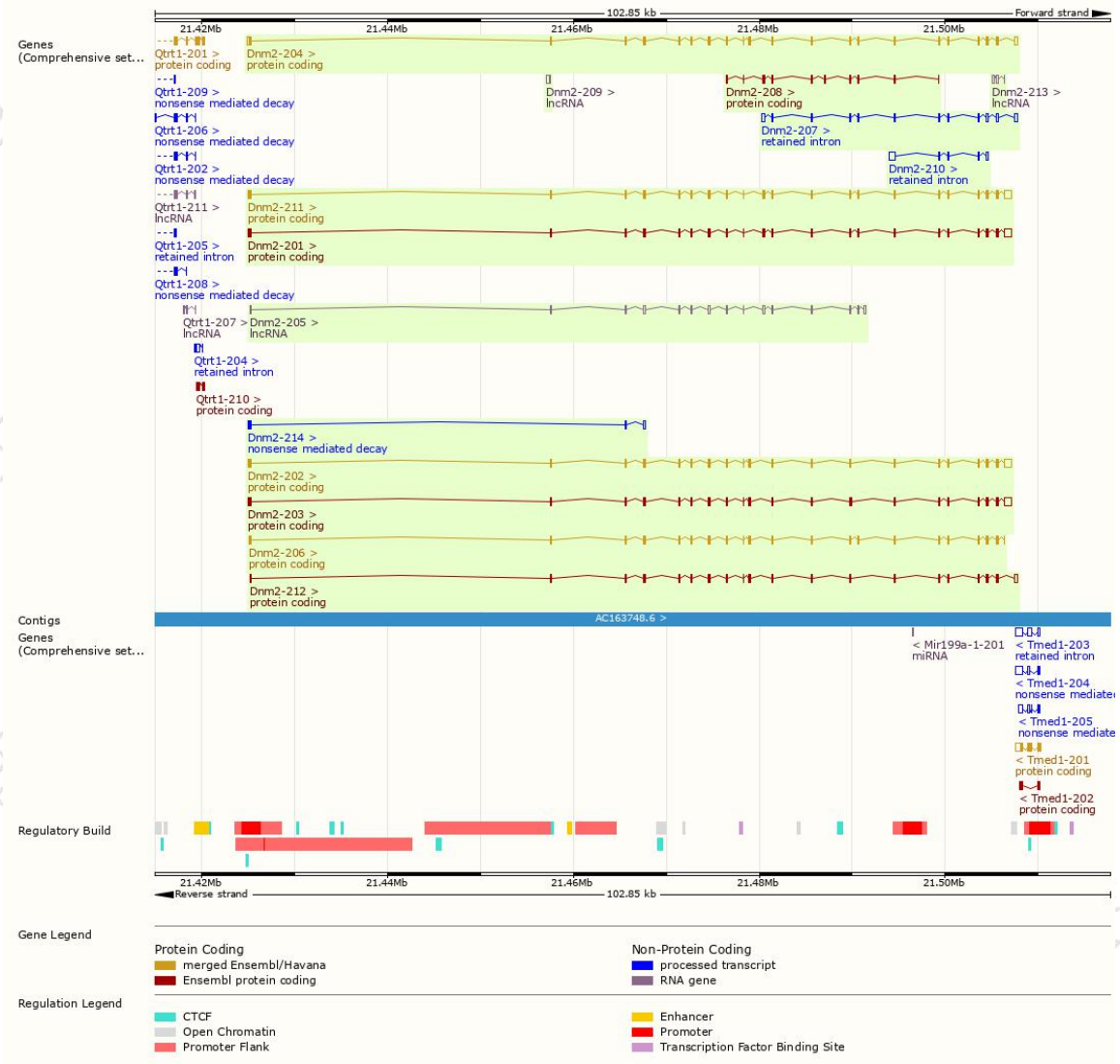
The gene has 14 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dnm2-211	ENSMUST00000173397.7	3510	869aa	Protein coding	CCDS57660	Q3TCR7	TSL:1 GENCODE basic APPRIS ALT1
Dnm2-202	ENSMUST00000091087.12	3450	866aa	Protein coding	CCDS57659	P39054	TSL:1 GENCODE basic APPRIS ALT1
Dnm2-204	ENSMUST00000165766.8	3063	860aa	Protein coding	CCDS57658	Q3T9X3	TSL:1 GENCODE basic
Dnm2-206	ENSMUST00000172482.7	2613	870aa	Protein coding	CCDS57657	P39054	TSL:5 GENCODE basic APPRIS P4
Dnm2-201	ENSMUST00000072362.13	3502	870aa	Protein coding	-	G3X9G4	TSL:1 GENCODE basic APPRIS ALT1
Dnm2-203	ENSMUST00000115404.10	3455	870aa	Protein coding	-	F8WIV5	TSL:5 GENCODE basic APPRIS ALT1
Dnm2-212	ENSMUST00000174050.7	2715	807aa	Protein coding	-	G3UZZ3	CDS 5' incomplete TSL:5
Dnm2-208	ENSMUST00000172833.1	744	248aa	Protein coding	-	G3UXX2	CDS 5' and 3' incomplete TSL:3
Dnm2-214	ENSMUST00000174828.1	608	69aa	Nonsense mediated decay	-	G3UY64	TSL:5
Dnm2-207	ENSMUST00000172763.7	1772	No protein	Retained intron	-	-	TSL:1
Dnm2-210	ENSMUST00000173299.1	1255	No protein	Retained intron	-	-	TSL:5
Dnm2-205	ENSMUST00000169194.8	1591	No protein	lncRNA	-	-	TSL:1
Dnm2-213	ENSMUST00000174243.1	454	No protein	lncRNA	-	-	TSL:3
Dnm2-209	ENSMUST00000172873.1	373	No protein	lncRNA	-	-	TSL:5

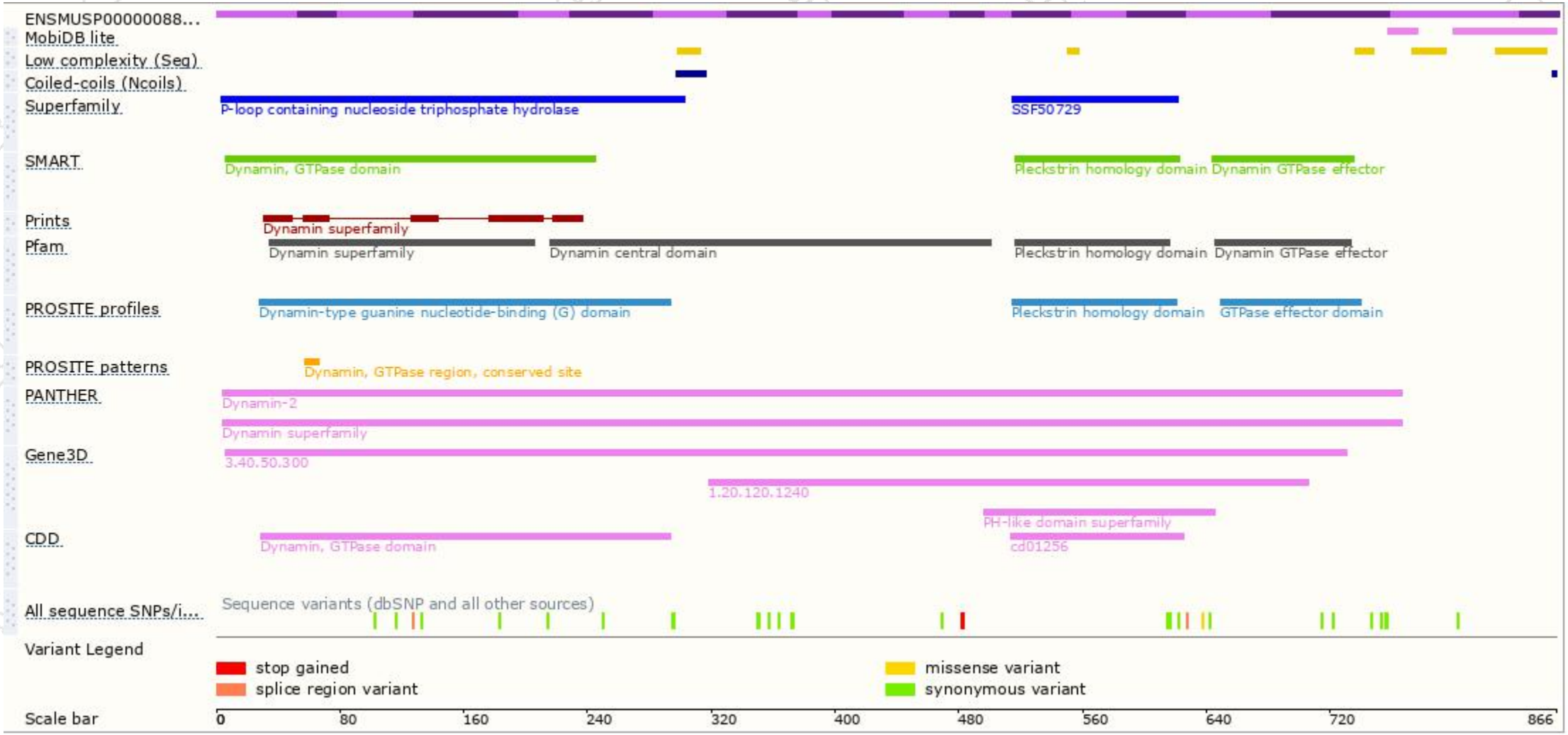
The strategy is based on the design of *Dnm2-202* transcript,The transcription is shown below



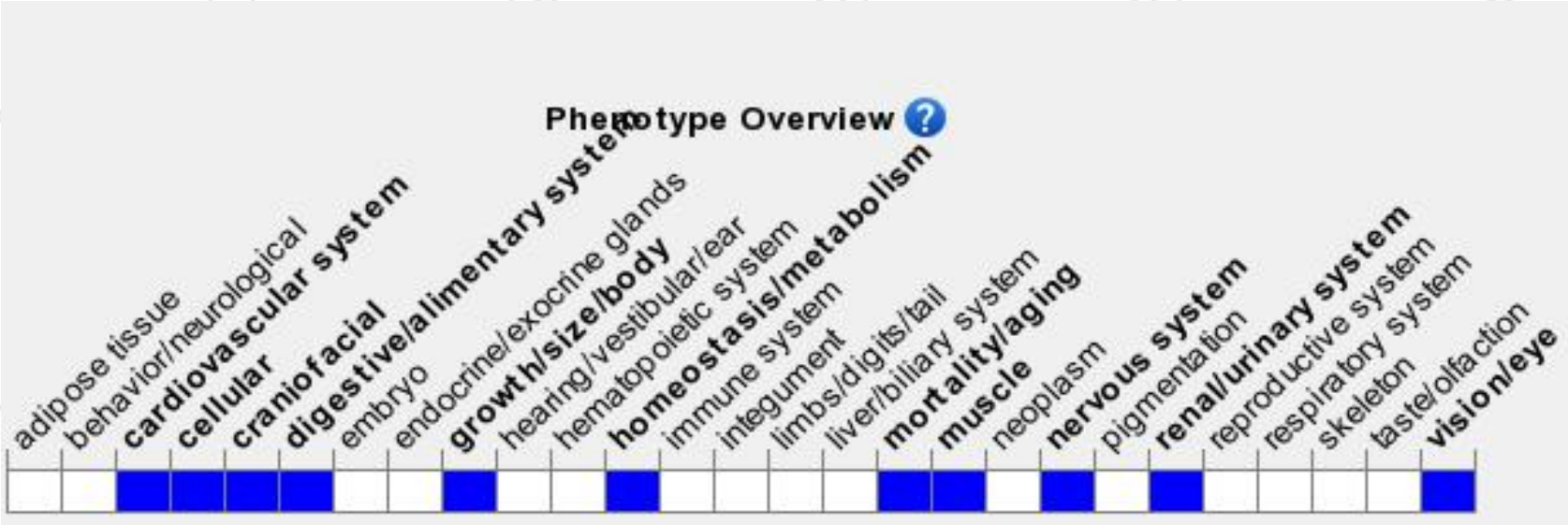
Genomic location distribution



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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