Nr3c2 Cas9-CKO Strategy

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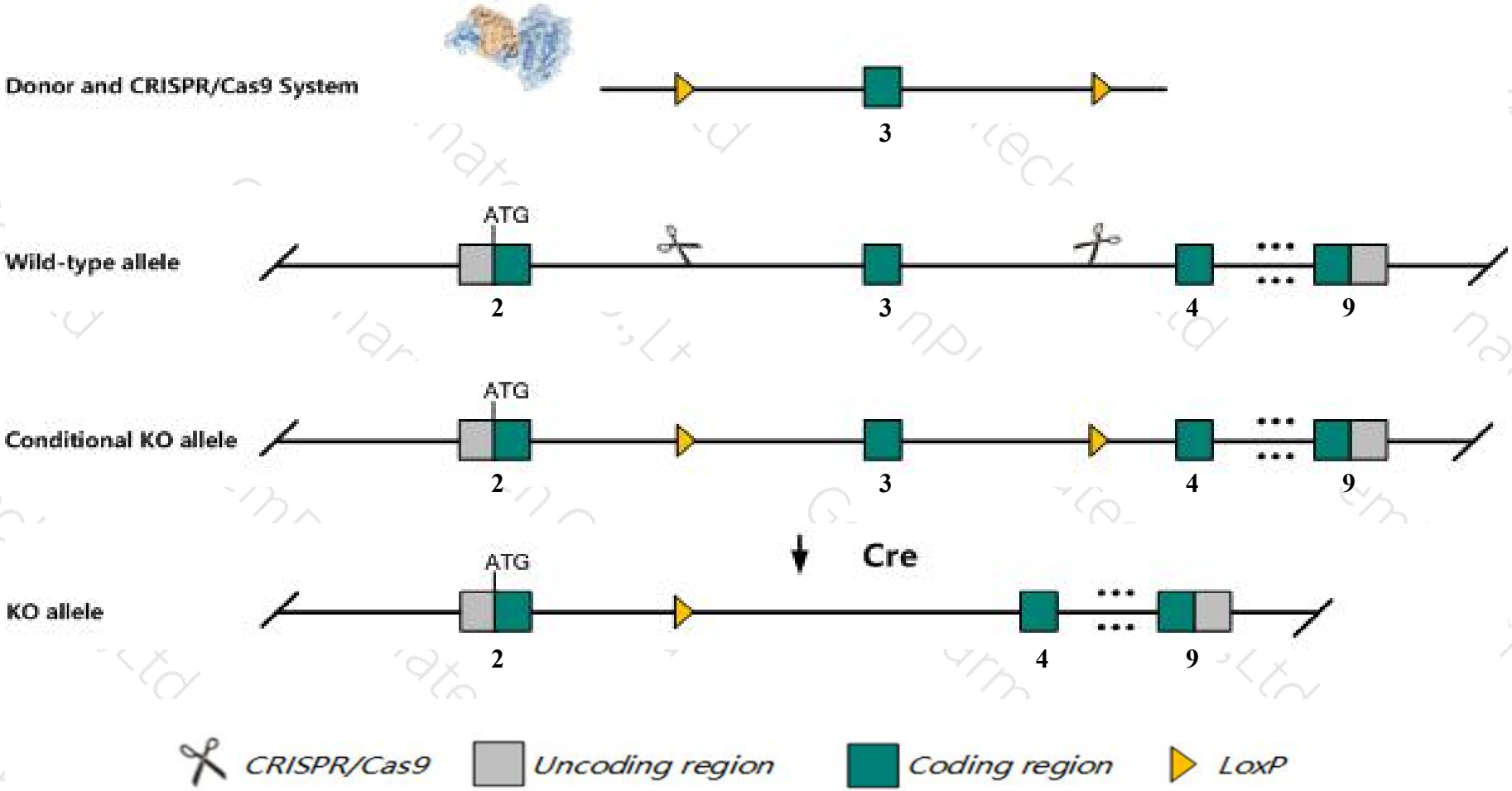
Project Overview

Project Name	***Nr3c2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Nr3c2* gene. The schematic diagram is as follows:



Technical routes

- The *Nr3c2* gene has 8 transcripts. According to the structure of *Nr3c2* gene, exon3 of *Nr3c2-204* (ENSMUST00000109913.8) transcript is recommended as the knockout region.The region contains 140bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Nr3c2* 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,Mice homozygous for a targeted null mutation exhibit weight loss and symptoms of pseudohypoaldosteronism, and eventually die at around day 10 after birth from renal salt wasting and dehydration.

➢ The *Nr3c2* gene is located on the Chr8. 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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Nr3c2 nuclear receptor subfamily 3, group C, member 2 [Mus musculus (house mouse)]

Gene ID: 110784, updated on 19-Mar-2019

Summary

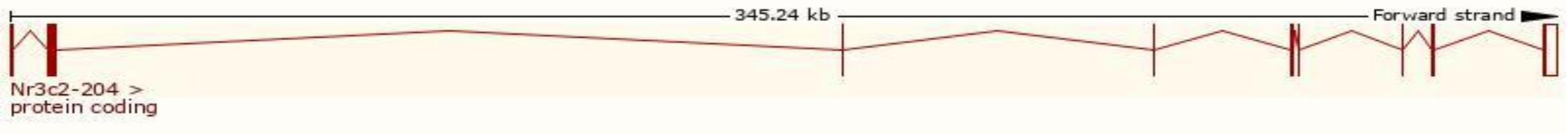
Official Symbol	Nr3c2 provided by MGI
Official Full Name	nuclear receptor subfamily 3, group C, member 2 provided by MGI
Primary source	MGI:MGI:99459
See related	Ensembl:ENSMUSG00000031618
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	MR, Mlr
Expression	Broad expression in colon adult (RPKM 12.0), frontal lobe adult (RPKM 3.4) and 20 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

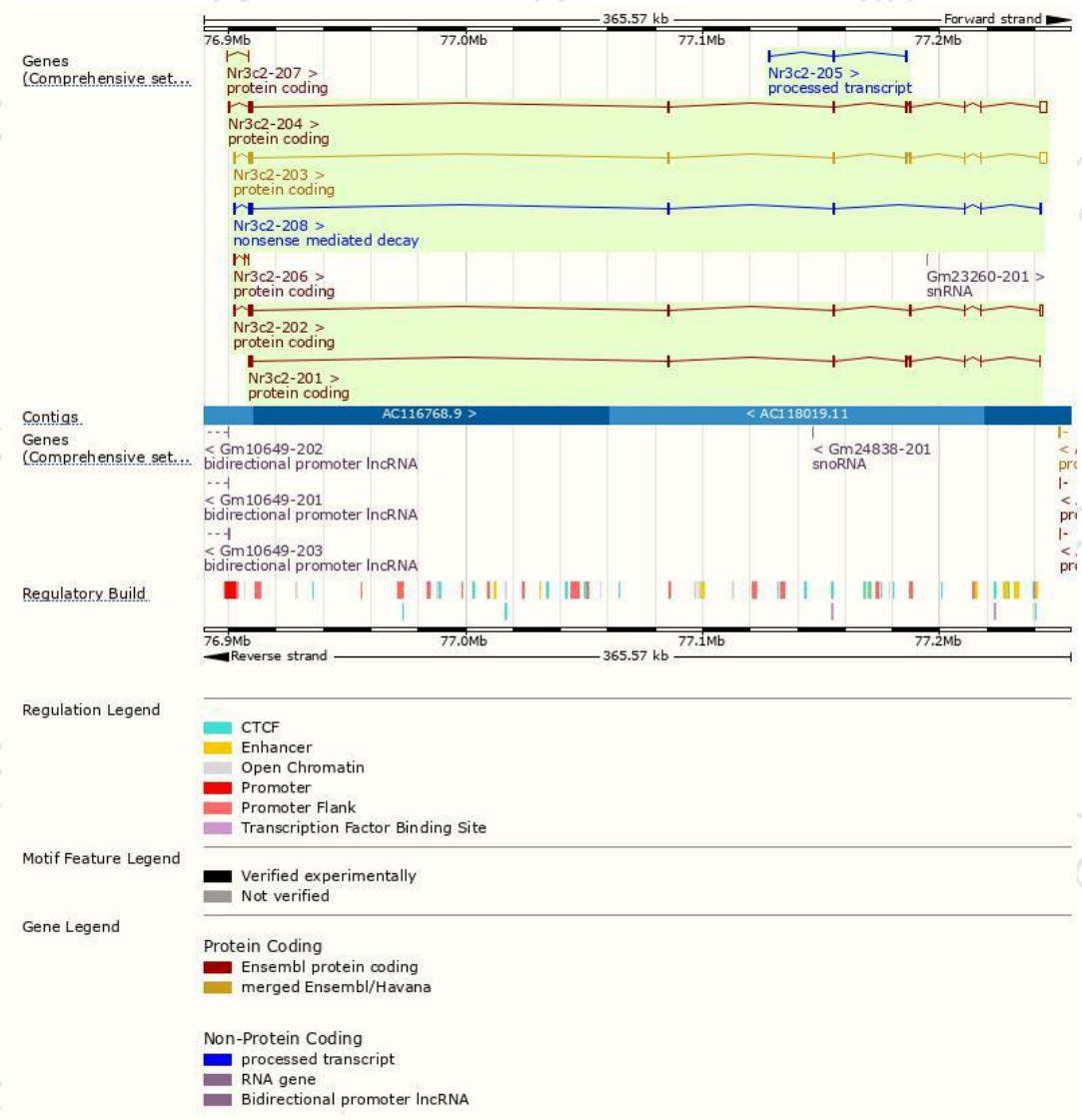
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nr3c2-204	ENSMUST00000109913.8	5916	980aa	Protein coding	CCDS40393	A3KN90	TSL:5 GENCODE basic APPRIS P2
Nr3c2-203	ENSMUST00000109912.7	5675	980aa	Protein coding	CCDS40393	A3KN90	TSL:1 GENCODE basic APPRIS P2
Nr3c2-202	ENSMUST00000109911.7	3804	867aa	Protein coding	-	D3Z473	TSL:5 GENCODE basic
Nr3c2-201	ENSMUST00000034031.5	2955	984aa	Protein coding	-	E9Q8M8	TSL:5 GENCODE basic APPRIS ALT2
Nr3c2-206	ENSMUST00000128862.1	612	128aa	Protein coding	-	D3Z7F2	CDS 3' incomplete TSL:2
Nr3c2-207	ENSMUST00000143284.1	270	47aa	Protein coding	-	D3Z7J4	CDS 3' incomplete TSL:5
Nr3c2-208	ENSMUST00000148106.7	3180	698aa	Nonsense mediated decay	-	D6RIL1	TSL:5
Nr3c2-205	ENSMUST00000126697.1	350	No protein	Processed transcript	-	-	TSL:2

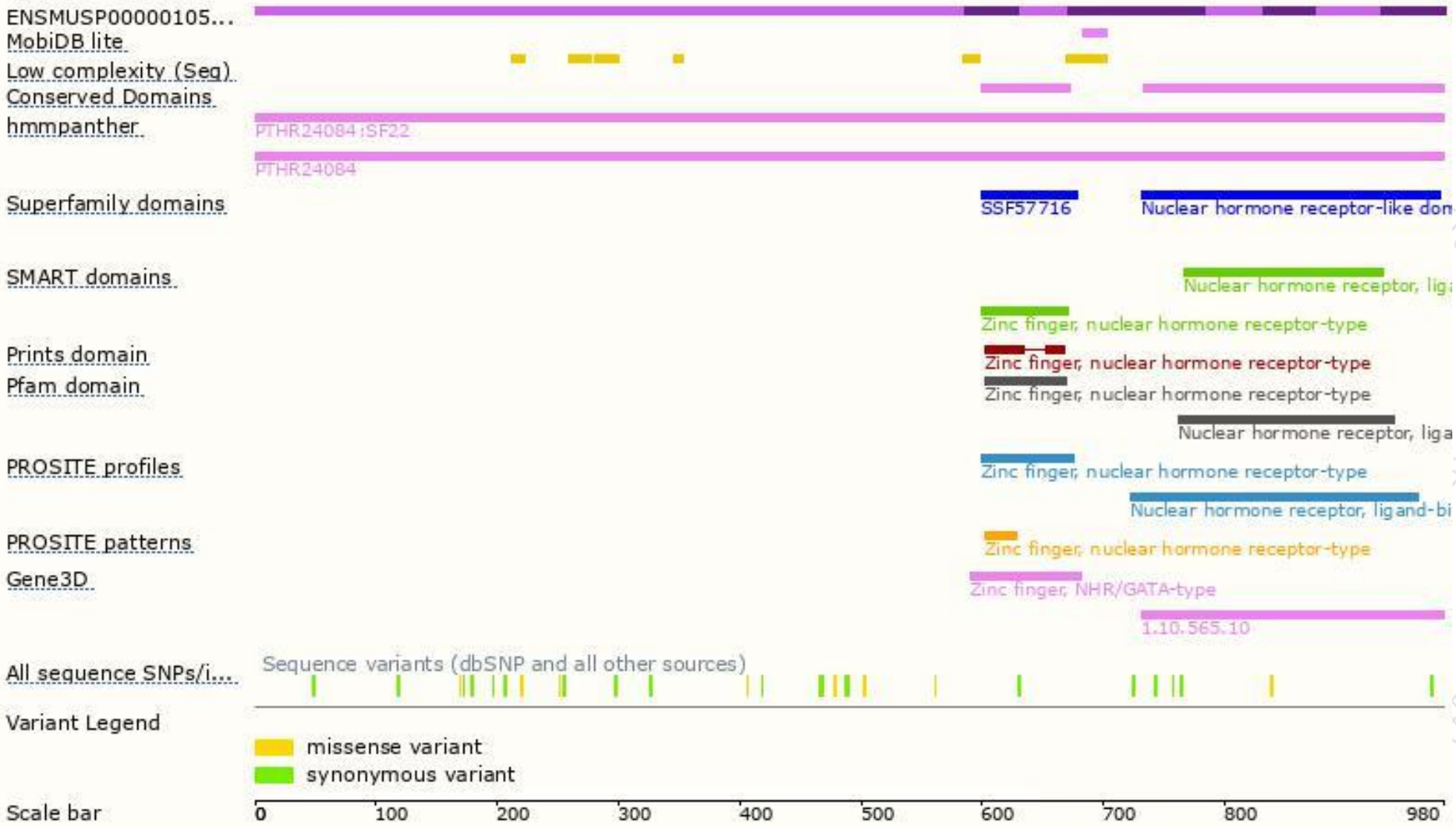
The strategy is based on the design of *Nr3c2-204* transcript,The transcription is shown below



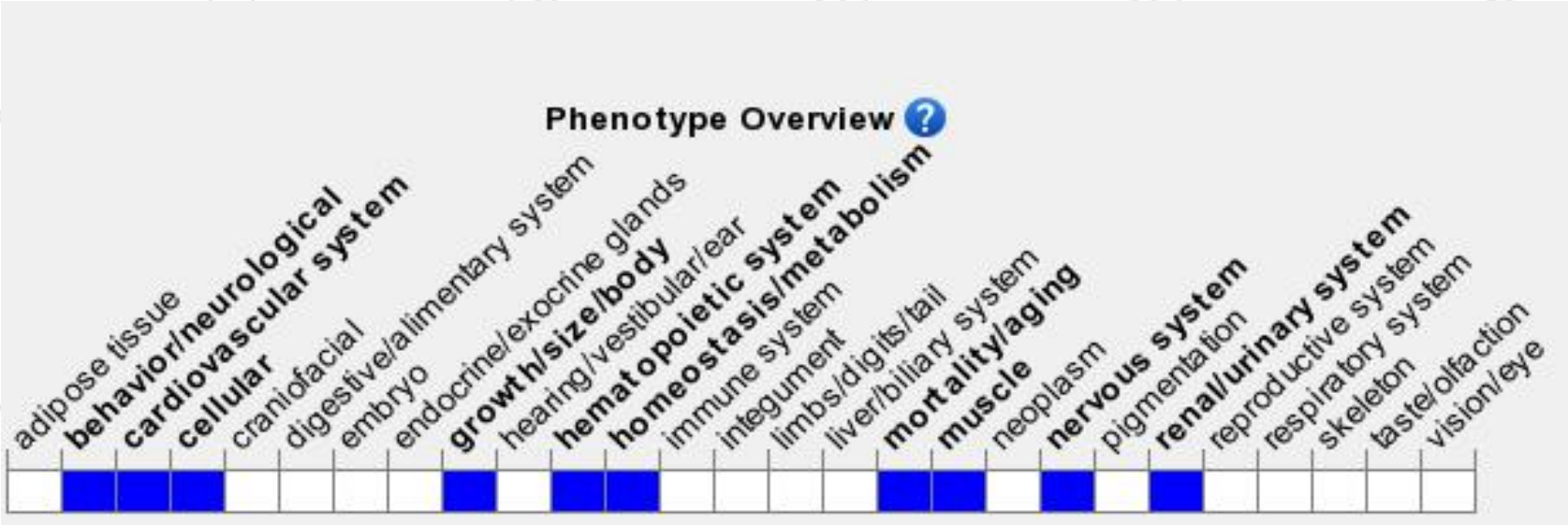
Genomic location distribution



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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