

***Wnk1* Cas9-CKO Strategy**

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Design Date: 2019-8-22

Project Overview

Project Name

Wnk1

Project type

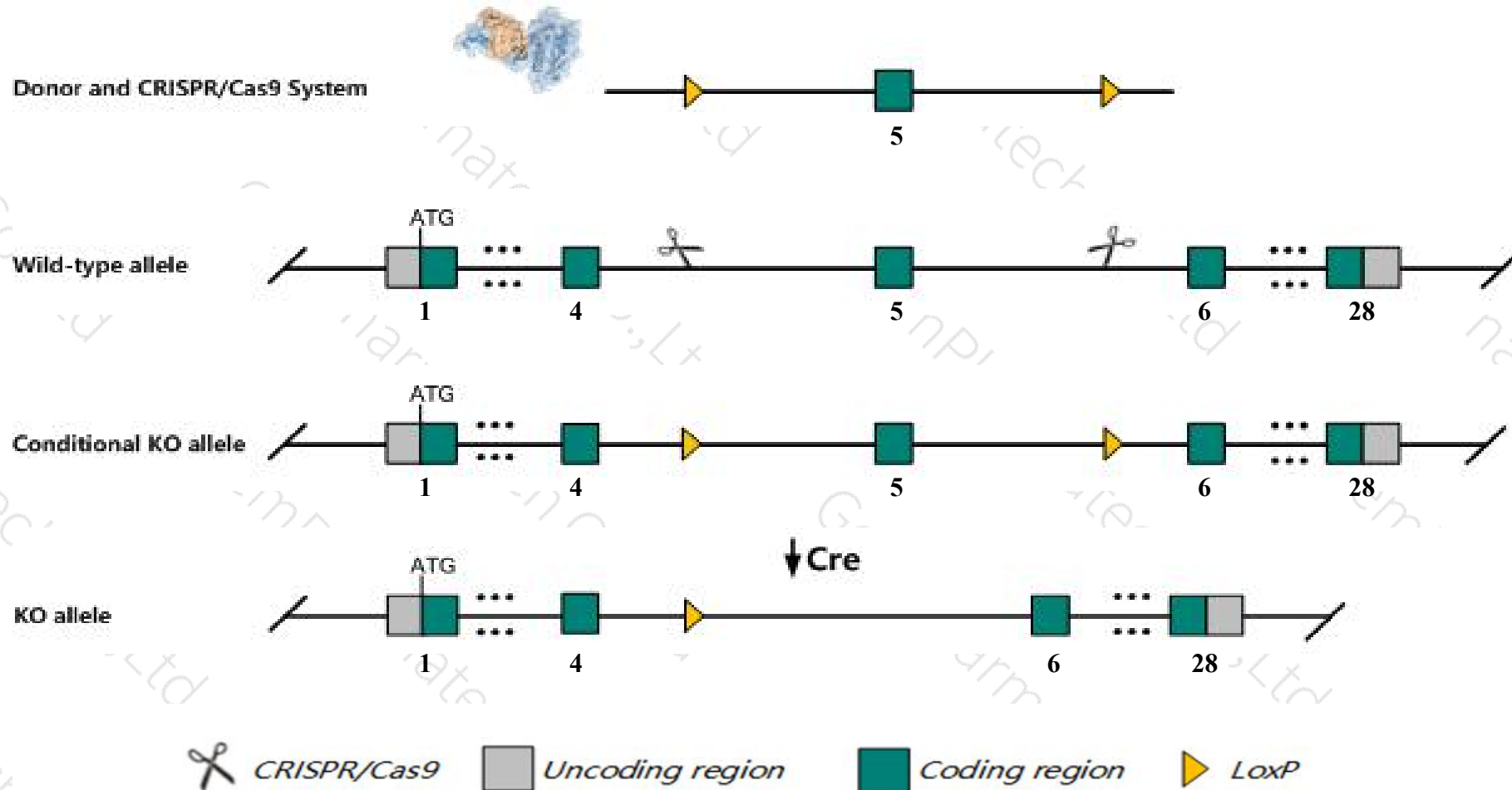
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Wnk1* gene. The schematic diagram is as follows:



- The *Wnk1* gene has 26 transcripts. According to the structure of *Wnk1* gene, exon5 of *Wnk1-203* (ENSMUST00000088644.12) transcript is recommended as the knockout region. The region contains 89bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Wnk1* 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, Homozygous mutant mice die before birth, whereas heterozygotes survive and exhibit decreased blood pressure. Mice homozygous for an allele that does not produce the kidney isoform exhibit a slight increase in systemic arterial diastolic blood pressure and reduced sensitivity to amiloride.
- The *Wnk1* gene is located on the Chr6. 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)

Wnk1 WNK lysine deficient protein kinase 1 [Mus musculus (house mouse)]

Gene ID: 232341, updated on 31-Jan-2019

Summary



Official Symbol	Wnk1 provided by MGI
Official Full Name	WNK lysine deficient protein kinase 1 provided by MGI
Primary source	MGI:MGI:2442092
See related	Ensembl:ENSMUSG00000045962
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	6430573H23Rik, EG406236, Hsn2, Prkwk1, mKIAA0344
Expression	Ubiquitous expression in heart adult (RPKM 40.9), thymus adult (RPKM 32.8) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

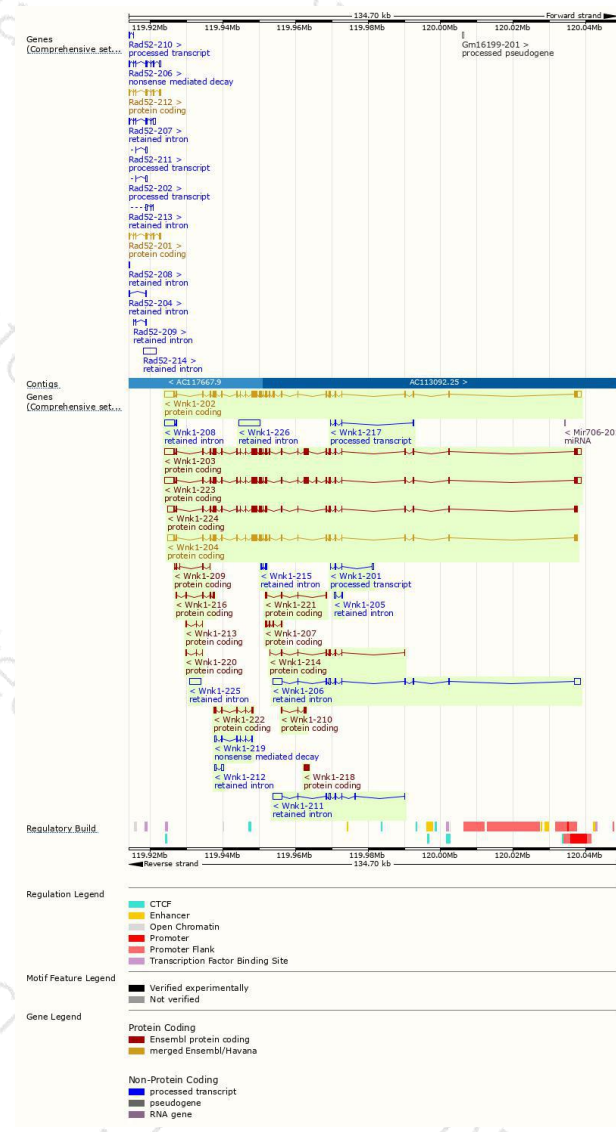
The gene has 26 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Wnk1-203	ENSMUST0000088644.12	11330	2635aa	Protein coding	CCDS57444	P83741	TSL5 GENCODE basic APPRIS ALT2
Wnk1-223	ENSMUST00000177761.7	11303	2626aa	Protein coding	CCDS57445	P83741	TSL5 GENCODE basic APPRIS ALT2
Wnk1-202	ENSMUST0000060043.12	10575	2377aa	Protein coding	CCDS20478	P83741	TSL1 GENCODE basic APPRIS P3
Wnk1-204	ENSMUST0000088646.11	8364	2195aa	Protein coding	CCDS51888	P83741	TSL1 GENCODE basic APPRIS ALT2
Wnk1-224	ENSMUST00000203030.2	8163	2128aa	Protein coding	CCDS85147	P83741	TSL1 GENCODE basic APPRIS ALT2
Wnk1-218	ENSMUST00000161512.2	1426	433aa	Protein coding	-	A0A0R4J274	TSLNA GENCODE basic
Wnk1-222	ENSMUST00000162541.7	1221	407aa	Protein coding	-	F7CZC4	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL3
Wnk1-214	ENSMUST00000161048.7	1215	405aa	Protein coding	-	F7CC62	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL1
Wnk1-216	ENSMUST00000161243.1	843	281aa	Protein coding	-	F6U2S8	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL5
Wnk1-207	ENSMUST00000159785.1	755	252aa	Protein coding	-	F6TW99	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL3
Wnk1-210	ENSMUST00000160886.1	716	238aa	Protein coding	-	J3QPKQ	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL3
Wnk1-221	ENSMUST00000162368.7	640	213aa	Protein coding	-	F7CBK9	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL3
Wnk1-209	ENSMUST00000160493.7	600	200aa	Protein coding	-	F6TAR7	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL2
Wnk1-220	ENSMUST00000162190.7	201	67aa	Protein coding	-	F7B663	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL5
Wnk1-213	ENSMUST00000160872.1	181	61aa	Protein coding	-	F6V9V5	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL5
Wnk1-219	ENSMUST00000161802.1	845	85aa	Nonsense mediated decay	-	F6S7V4	CDS 5' incomplete TSL3
Wnk1-201	ENSMUST0000035513.6	646	No protein	Processed transcript	-	-	TSL5
Wnk1-217	ENSMUST00000161392.7	516	No protein	Processed transcript	-	-	TSL5
Wnk1-226	ENSMUST00000204207.1	5825	No protein	Retained intron	-	-	TSLNA
Wnk1-206	ENSMUST00000159397.7	5745	No protein	Retained intron	-	-	TSL5
Wnk1-211	ENSMUST00000160769.7	3623	No protein	Retained intron	-	-	TSL1
Wnk1-225	ENSMUST00000203335.1	3065	No protein	Retained intron	-	-	TSLNA
Wnk1-208	ENSMUST00000159903.1	2934	No protein	Retained intron	-	-	TSL1
Wnk1-212	ENSMUST00000160820.1	748	No protein	Retained intron	-	-	TSL2
Wnk1-215	ENSMUST00000161185.1	731	No protein	Retained intron	-	-	TSL3
Wnk1-205	ENSMUST00000159151.1	628	No protein	Retained intron	-	-	TSL2

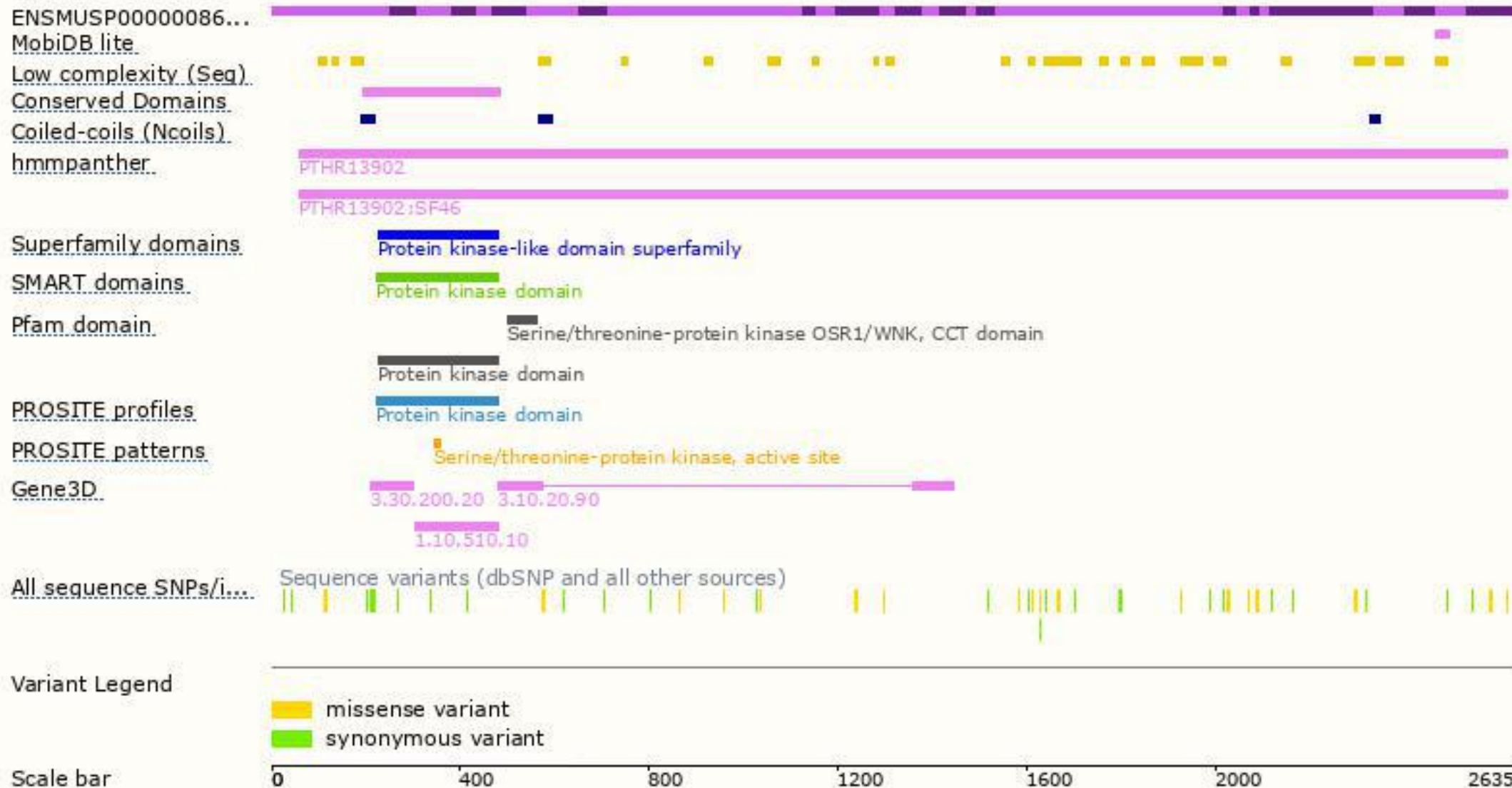
The strategy is based on the design of *Wnk1-203* transcript,The transcription is shown below



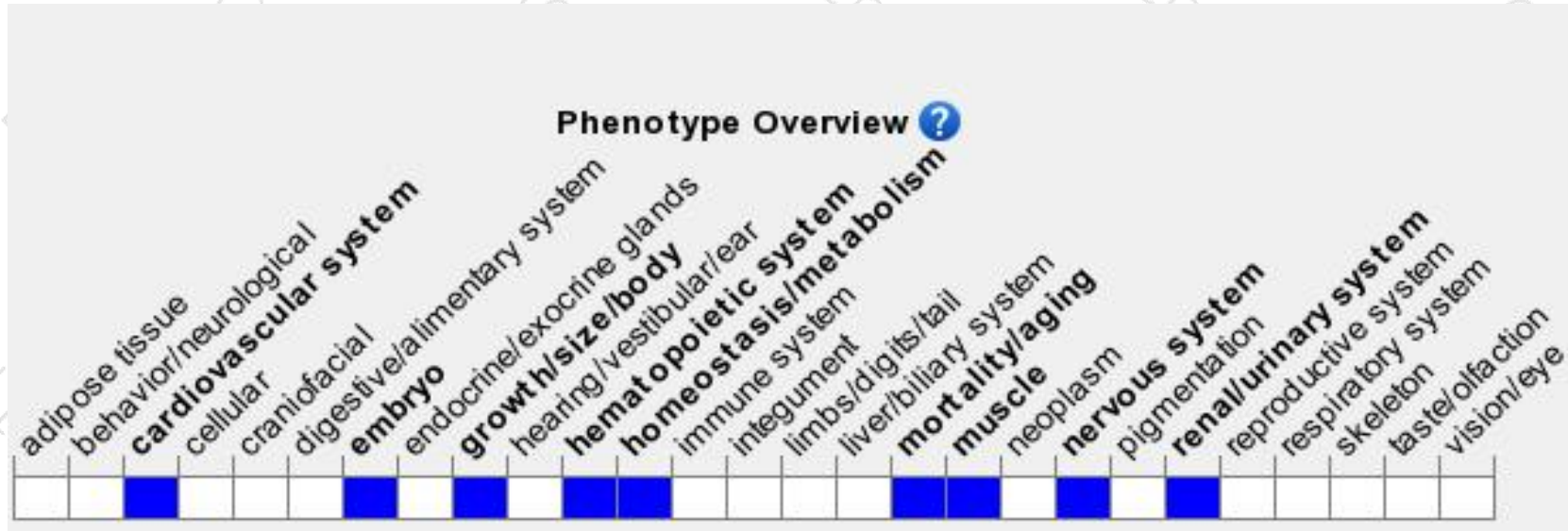
Genomic location distribution



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, Homozygous mutant mice die before birth, whereas heterozygotes survive and exhibit decreased blood pressure. Mice homozygous for an allele that does not produce the kidney isoform exhibit a slight increase in systemic arterial diastolic blood pressure and reduced sensitivity to amiloride.

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

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