

# ***Brd2* Cas9-CKO Strategy**

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# Project Overview

**Project Name**

*Brd2*

**Project type**

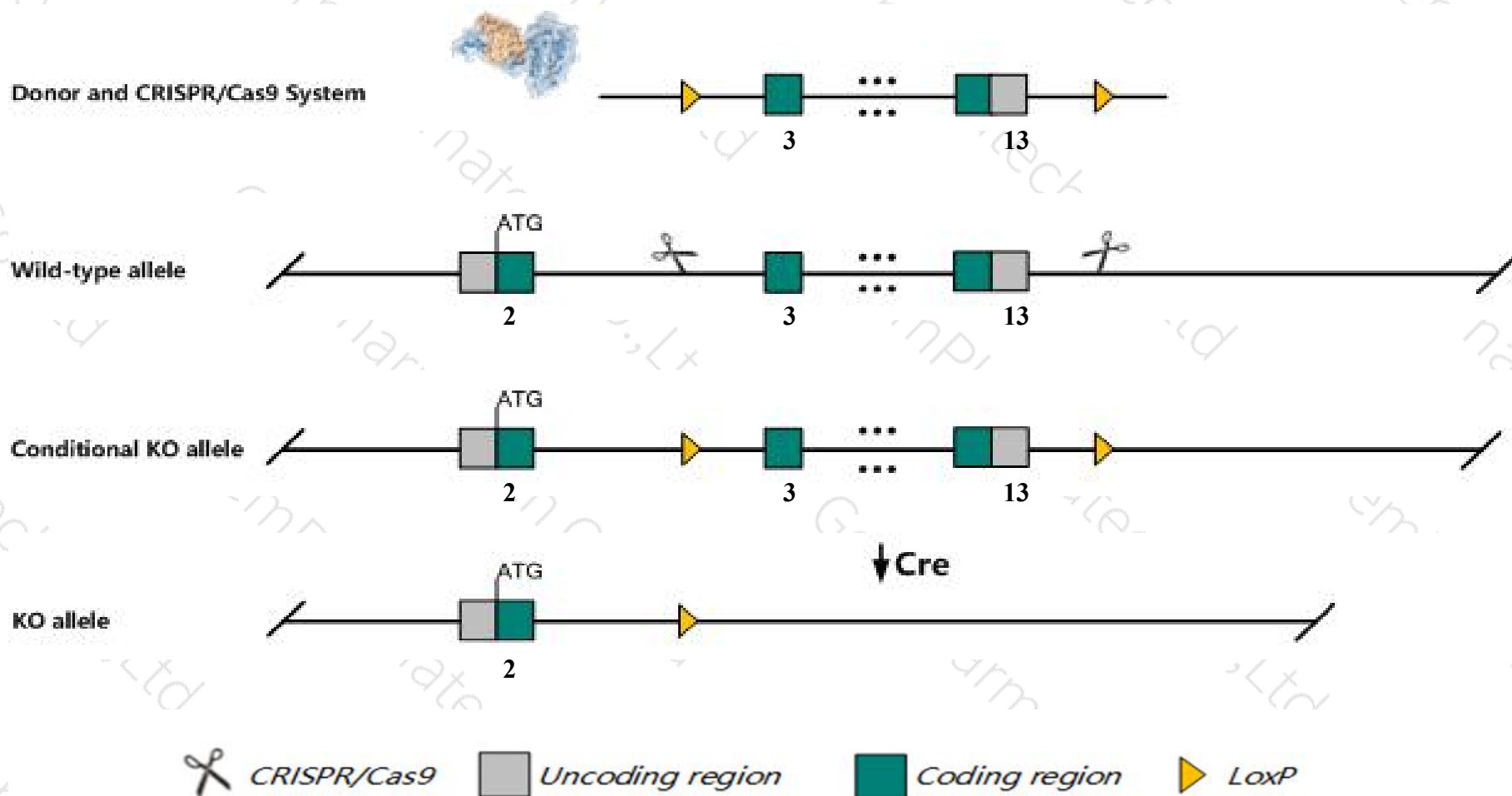
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Brd2* gene. The schematic diagram is as follows:



# Technical routes

- The *Brd2* gene has 19 transcripts. According to the structure of *Brd2* gene, exon3-exon13 of *Brd2-201* (ENSMUST00000025193.13) transcript is recommended as the knockout region. The region contains 2371bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Brd2* 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 null mutation display embryonic lethality during organogenesis with decreased embryo size, decreased cell proliferation, a delay in the cell cycle, and increased cell death. Heterozygous mice also display decreased cell proliferation.
- The *Brd2* gene is located on the Chr17. 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)

## Brd2 bromodomain containing 2 [Mus musculus (house mouse)]

Gene ID: 14312, updated on 31-Jan-2019

### Summary



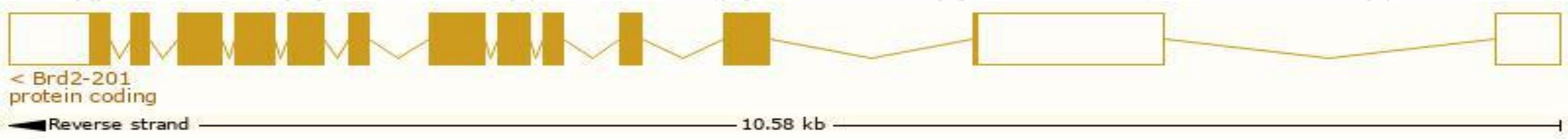
<b>Official Symbol</b>	Brd2 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	bromodomain containing 2 provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:99495</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000024335</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	VALIDATED
<b>Organism</b>	<a href="#">Mus musculus</a>
<b>Lineage</b>	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
<b>Also known as</b>	AW228947, D17H6S113E, Frg-1, Fsrg-1, Fsrg1, Nat, Ring3, Rnf3, mKIAA4005
<b>Expression</b>	Ubiquitous expression in thymus adult (RPKM 36.3), adrenal adult (RPKM 32.4) and 28 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

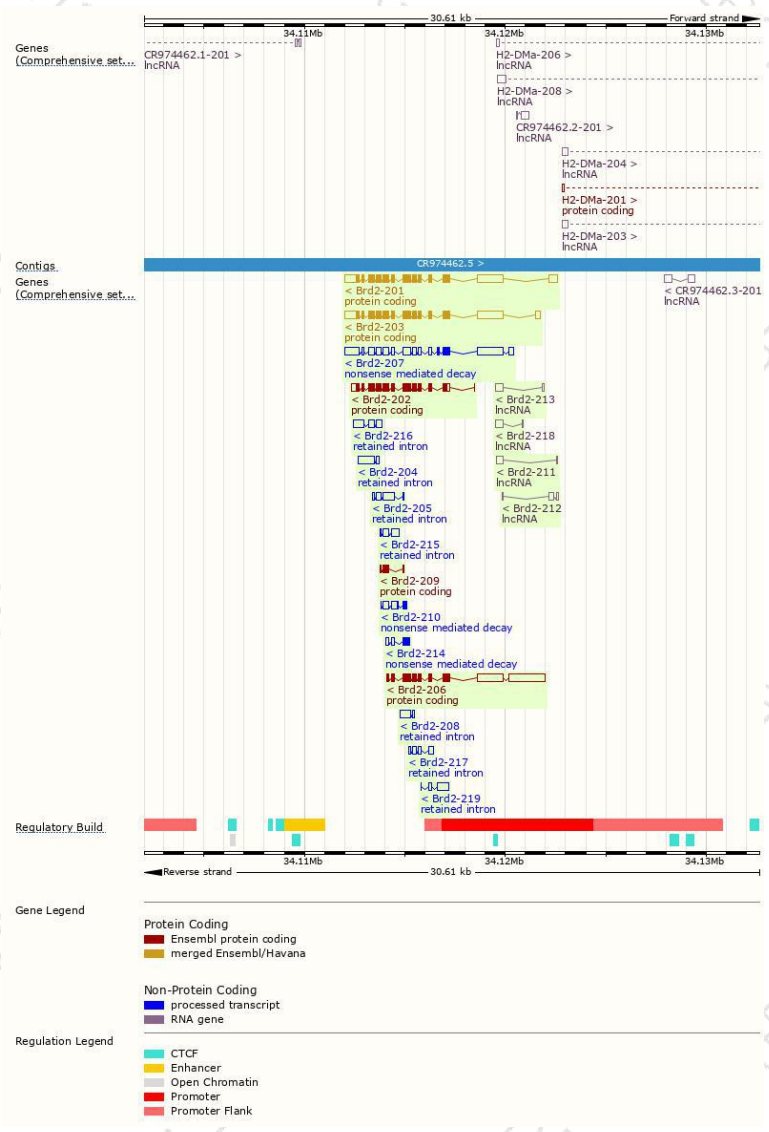
The gene has 19 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Brd2-201	<a href="#">ENSMUST00000025193.13</a>	4657	<a href="#">798aa</a>	Protein coding	<a href="#">CCDS28641</a>	<a href="#">B2RS09_Q7JJ13</a>	TSL:1 GENCODE basic APPRIS P2
Brd2-203	<a href="#">ENSMUST00000114242.8</a>	4483	<a href="#">798aa</a>	Protein coding	<a href="#">CCDS28641</a>	<a href="#">B2RS09_Q7JJ13</a>	TSL:1 GENCODE basic APPRIS P2
Brd2-206	<a href="#">ENSMUST00000151986.1</a>	4482	<a href="#">473aa</a>	Protein coding	-	<a href="#">I7HPW1</a>	CDS 3' incomplete TSL:1
Brd2-202	<a href="#">ENSMUST00000095347.12</a>	2617	<a href="#">752aa</a>	Protein coding	-	<a href="#">U3KL T0</a>	TSL:5 GENCODE basic APPRIS ALT2
Brd2-209	<a href="#">ENSMUST00000173032.1</a>	406	<a href="#">136aa</a>	Protein coding	-	<a href="#">G3UZR9</a>	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Brd2-207	<a href="#">ENSMUST00000154232.8</a>	4520	<a href="#">146aa</a>	Nonsense mediated decay	-	<a href="#">Q3TH63</a>	TSL:5
Brd2-210	<a href="#">ENSMUST00000173204.1</a>	771	<a href="#">64aa</a>	Nonsense mediated decay	-	<a href="#">S4R1Z1</a>	CDS 5' incomplete TSL:3
Brd2-214	<a href="#">ENSMUST00000235313.1</a>	638	<a href="#">132aa</a>	Nonsense mediated decay	-	-	CDS 5' incomplete
Brd2-216	<a href="#">ENSMUST00000236090.1</a>	1109	No protein	Retained intron	-	-	
Brd2-205	<a href="#">ENSMUST00000148143.2</a>	975	No protein	Retained intron	-	-	TSL:5
Brd2-204	<a href="#">ENSMUST00000142570.1</a>	961	No protein	Retained intron	-	-	TSL:1
Brd2-219	<a href="#">ENSMUST00000237872.1</a>	747	No protein	Retained intron	-	-	
Brd2-215	<a href="#">ENSMUST00000235347.1</a>	684	No protein	Retained intron	-	-	
Brd2-217	<a href="#">ENSMUST00000236781.1</a>	654	No protein	Retained intron	-	-	
Brd2-208	<a href="#">ENSMUST00000155286.1</a>	610	No protein	Retained intron	-	-	TSL:2
Brd2-213	<a href="#">ENSMUST00000179722.1</a>	463	No protein	lncRNA	-	-	TSL:2
Brd2-218	<a href="#">ENSMUST00000237345.1</a>	442	No protein	lncRNA	-	-	
Brd2-211	<a href="#">ENSMUST00000177828.1</a>	396	No protein	lncRNA	-	-	TSL:3
Brd2-212	<a href="#">ENSMUST00000179687.1</a>	366	No protein	lncRNA	-	-	TSL:5

The strategy is based on the design of *Brd2-201* transcript,The transcription is shown below

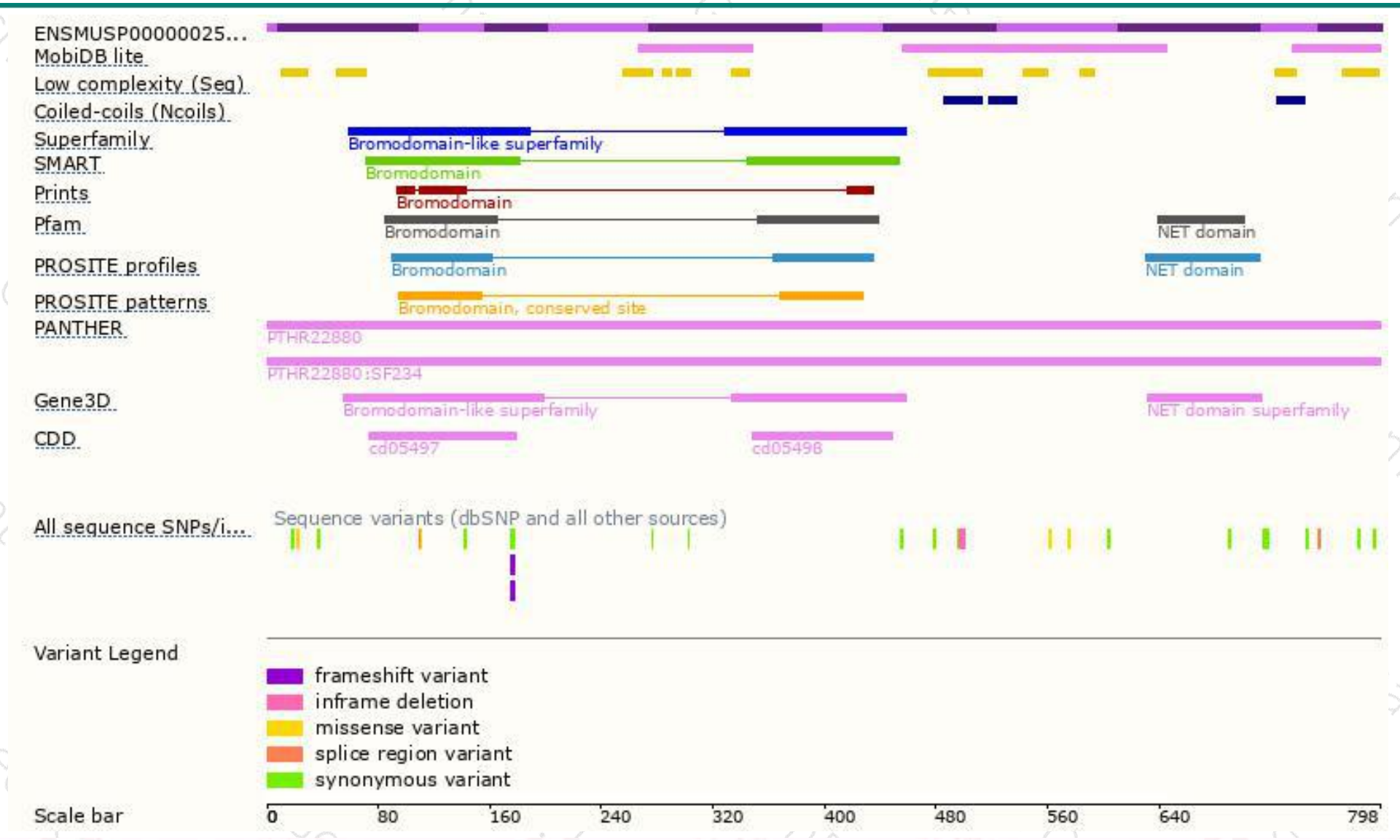


# Genomic location distribution

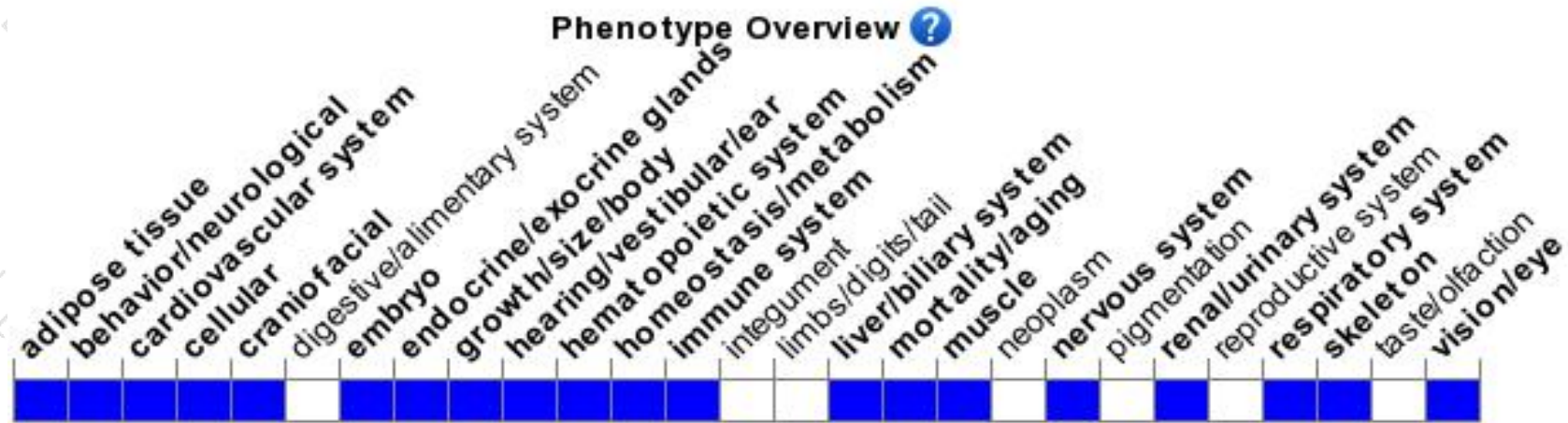




# Protein domain



# Mouse phenotype description(MGI)



*Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).*

According to the existing MGI data, Mice homozygous for a null mutation display embryonic lethality during organogenesis with decreased embryo size, decreased cell proliferation, a delay in the cell cycle, and increased cell death. Heterozygous mice also display decreased cell proliferation.

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

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