

# ***Eno2* Cas9-CKO Strategy**

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**Reviewer: Daohua Xu**

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# Project Overview

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**Project Name**

***Eno2***

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**Project type**

**Cas9-CKO**

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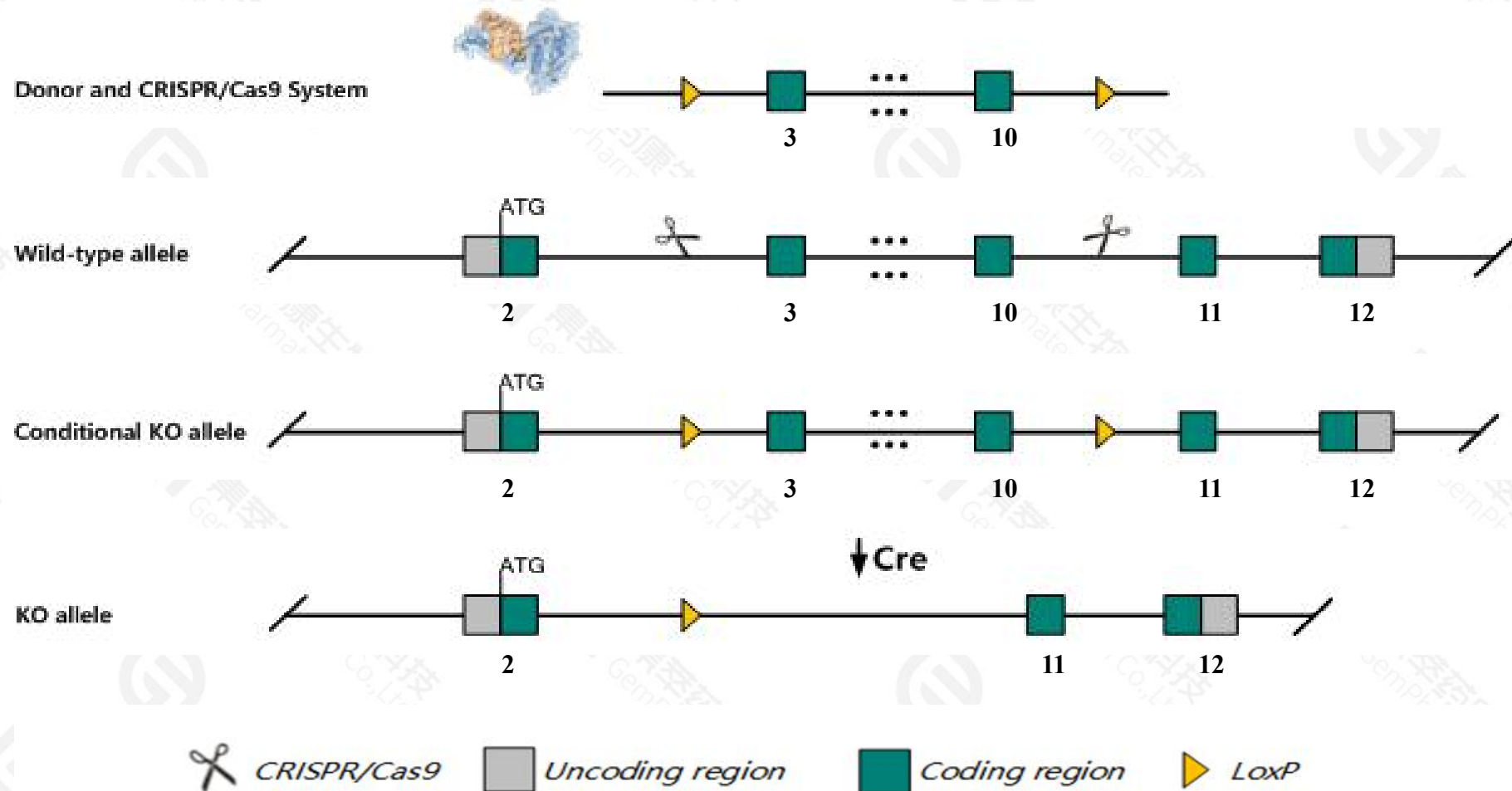
**Strain background**

**C57BL/6JGpt**

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# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Eno2* gene. The schematic diagram is as follows:



- The *Eno2* gene has 16 transcripts. According to the structure of *Eno2* gene, exon3-exon10 of *Eno2*-201(ENSMUST00000004378.15) transcript is recommended as the knockout region. The region contains 1091bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Eno2* 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.



- The Intron2 and Intron10 are only 441bp and 946bp, loxp insertion may affect mRNA splicing.
- The *Eno2* 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)

## Eno2 enolase 2, gamma neuronal [Mus musculus (house mouse)]

Gene ID: 13807, updated on 13-Mar-2020

### Summary

**Official Symbol** Eno2 provided by [MGI](#)

**Official Full Name** enolase 2, gamma neuronal provided by [MGI](#)

**Primary source** [MGI:MGI:95394](#)

**See related** [Ensembl:ENSMUSG000000004267](#)

**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** AI837106, D6Ert375e, Eno-2, NSE

**Expression** Biased expression in cerebellum adult (RPKM 147.0), frontal lobe adult (RPKM 120.3) and 4 other tissues [See more](#)

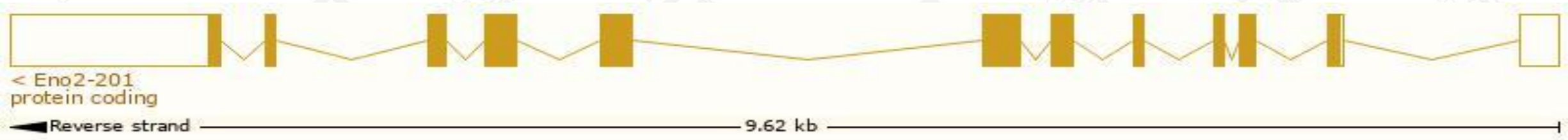
**Orthologs** [human](#) [all](#)

# Transcript information (Ensembl)

The gene has 16 transcripts,all transcripts are shown below:

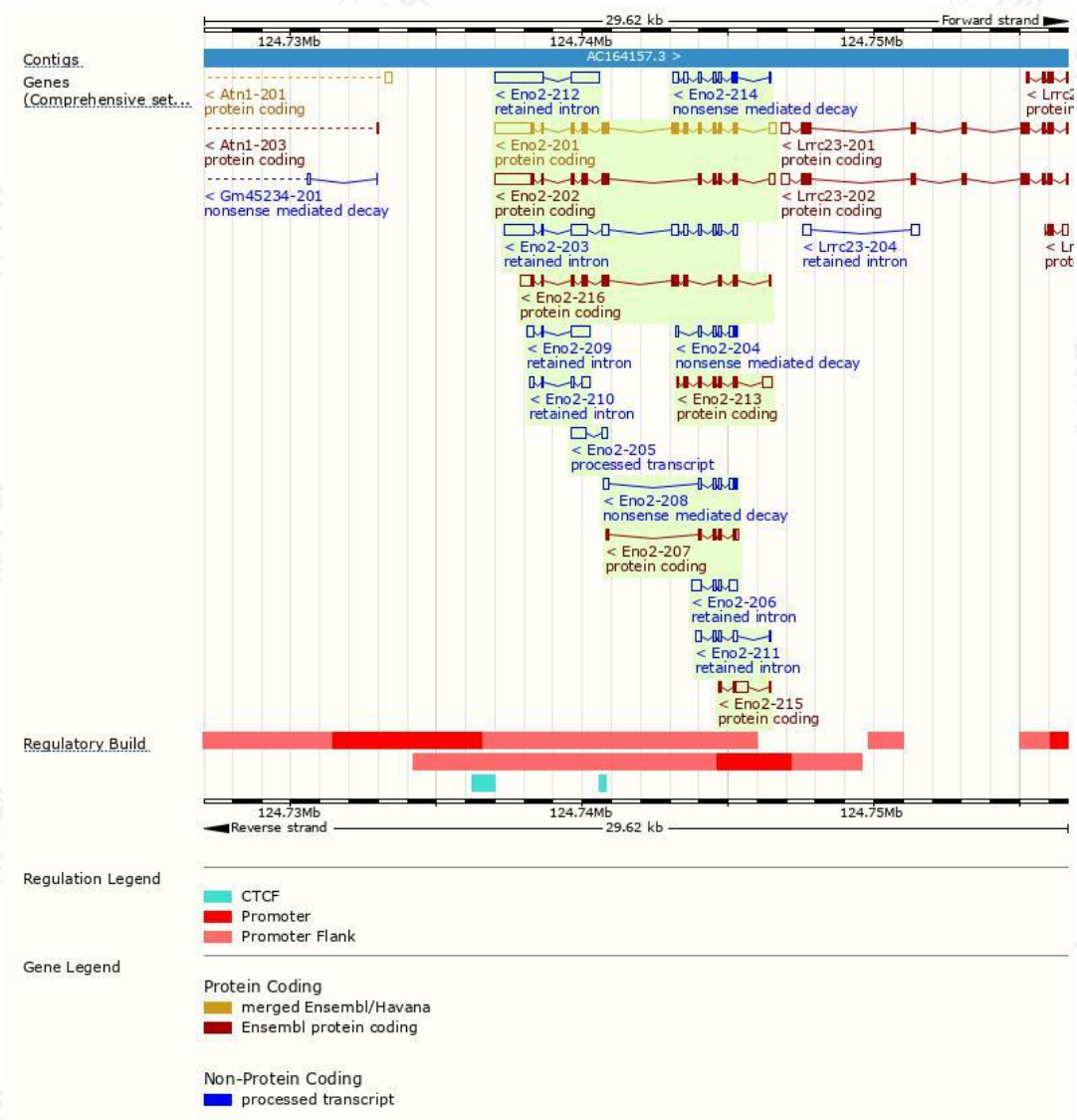
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Eno2-201	<a href="#">ENSMUST00000004378.14</a>	2795	<a href="#">434aa</a>	Protein coding	<a href="#">CCDS20527</a>	<a href="#">P17183 Q545V3</a>	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Eno2-202	<a href="#">ENSMUST00000112476.7</a>	2386	<a href="#">315aa</a>	Protein coding	<a href="#">CCDS80609</a>	<a href="#">D3Z6E4</a>	TSL:1 GENCODE basic
Eno2-216	<a href="#">ENSMUST00000204896.2</a>	1640	<a href="#">391aa</a>	Protein coding	-	<a href="#">A0A0N4SUX5</a>	TSL:5 GENCODE basic
Eno2-213	<a href="#">ENSMUST00000149652.1</a>	804	<a href="#">162aa</a>	Protein coding	-	<a href="#">D3Z2S4</a>	CDS 3' incomplete TSL:2
Eno2-215	<a href="#">ENSMUST00000156033.1</a>	622	<a href="#">58aa</a>	Protein coding	-	<a href="#">A0A0N4SUI6</a>	CDS 3' incomplete TSL:5
Eno2-207	<a href="#">ENSMUST00000135626.7</a>	419	<a href="#">121aa</a>	Protein coding	-	<a href="#">D3YVD3</a>	CDS 3' incomplete TSL:3
Eno2-214	<a href="#">ENSMUST00000151214.7</a>	745	<a href="#">37aa</a>	Nonsense mediated decay	-	<a href="#">A0A0N4SUW8</a>	TSL:5
Eno2-208	<a href="#">ENSMUST00000138770.7</a>	616	<a href="#">37aa</a>	Nonsense mediated decay	-	<a href="#">A0A0N4SUW8</a>	TSL:3
Eno2-204	<a href="#">ENSMUST00000127274.7</a>	565	<a href="#">37aa</a>	Nonsense mediated decay	-	<a href="#">A0A0N4SUW8</a>	TSL:3
Eno2-205	<a href="#">ENSMUST00000128598.1</a>	716	No protein	Processed transcript	-	-	TSL:3
Eno2-212	<a href="#">ENSMUST00000147307.1</a>	2614	No protein	Retained intron	-	-	TSL:2
Eno2-203	<a href="#">ENSMUST00000125436.7</a>	2491	No protein	Retained intron	-	-	TSL:1
Eno2-209	<a href="#">ENSMUST00000139763.7</a>	898	No protein	Retained intron	-	-	TSL:3
Eno2-206	<a href="#">ENSMUST00000128701.7</a>	728	No protein	Retained intron	-	-	TSL:3
Eno2-210	<a href="#">ENSMUST00000146712.1</a>	632	No protein	Retained intron	-	-	TSL:2
Eno2-211	<a href="#">ENSMUST00000147004.1</a>	490	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Eno2-201* transcript,the transcription is shown below:



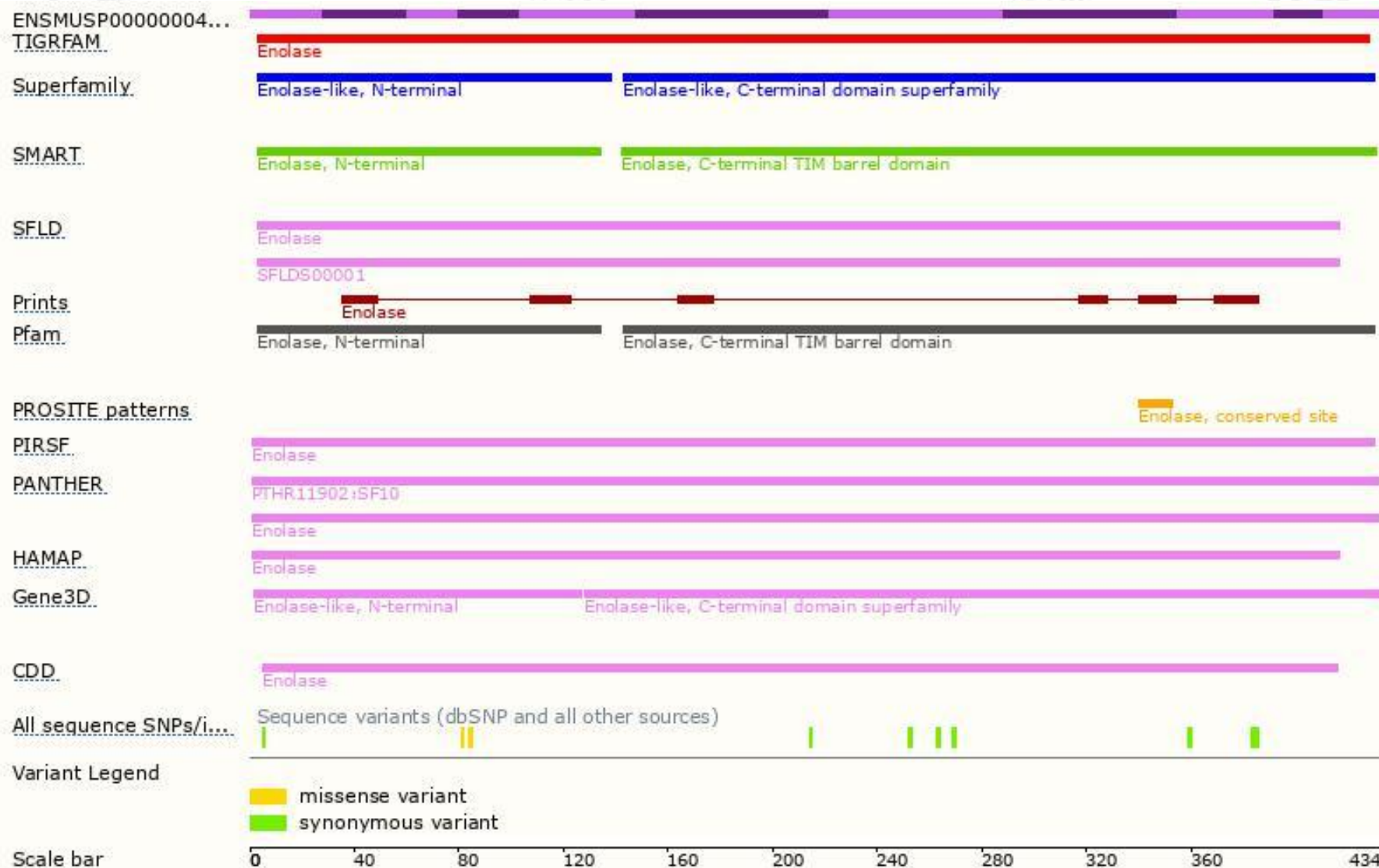


# Genomic location distribution

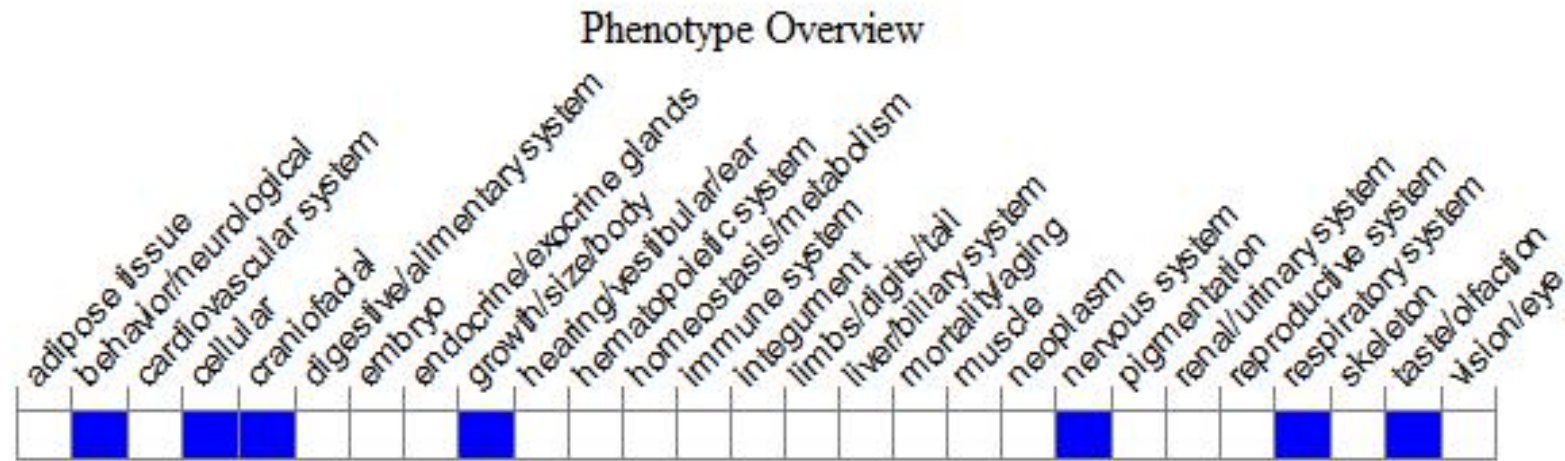




# Protein domain



# Mouse phenotype description(MGI )



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

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