

# *GnaI* Cas9-CKO Strategy

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# Project Overview

**Project Name**

*Gnal*

**Project type**

**Cas9-CKO**

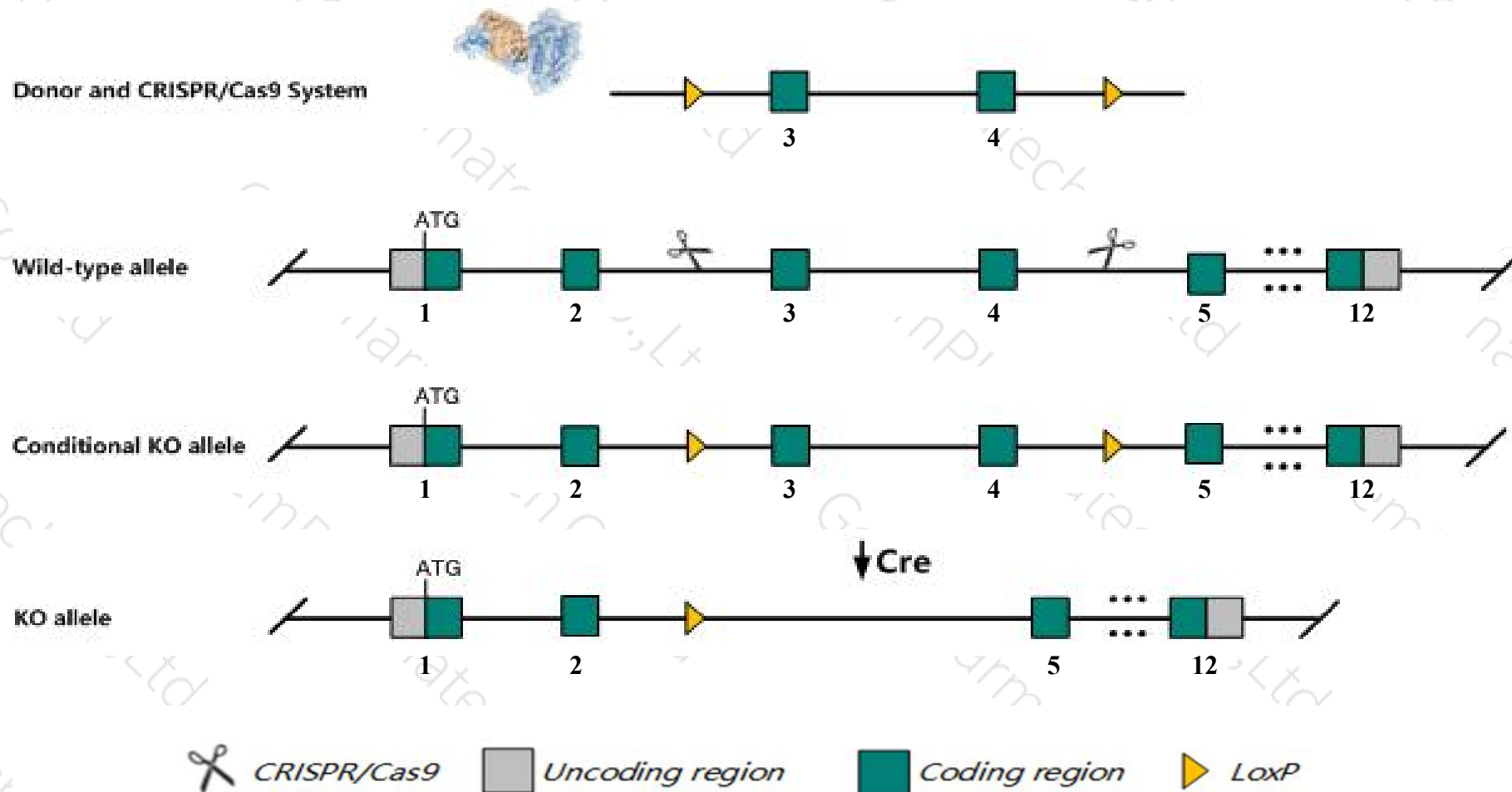
**Strain background**

**C57BL/6JGpt**



# Conditional Knockout strategy

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





# Technical routes

- The *Gnal* gene has 8 transcripts. According to the structure of *Gnal* gene, exon3-exon4 of *Gnal-201* (ENSMUST00000025402.14) transcript is recommended as the knockout region. The region contains 175bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gnal* 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 for a targeted mutation fail to feed, and ~75% die within 2 days after birth. Rare survivors reach sexual maturity and mate but are hyperactive and anosmic, exhibiting severely reduced odor-evoked electrophysical responses and significantly perturbed maternal behaviors.
- Transcript *Gnal*-207 may not be affected.
- The effect on transcript *Gnal*-206 is unknown.
- The *Gnal* gene is located on the Chr18. 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)

**Gnal** guanine nucleotide binding protein, alpha stimulating, olfactory type [ *Mus musculus* (house mouse) ]

Gene ID: 14680, updated on 10-Oct-2019

## Summary

**Official Symbol** Gnal provided by [MGI](#)  
**Official Full Name** guanine nucleotide binding protein, alpha stimulating, olfactory type provided by [MGI](#)  
**Primary source** [MGI:MG1:95774](#)  
**See related** [Ensembl:ENSMUSG000000024524](#)  
**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** Golf; Gna10; Galphaolf; 2610011C15Rik; 9630020G10Rik  
**Expression** Broad expression in CNS E18 (RPKM 24.2), cortex adult (RPKM 22.2) and 15 other tissues [See more](#)  
**Orthologs** [human](#) [all](#)

## Genomic context

**Location:** 18 E1; 18 39.85 cM

[See Gnal in Genome Data Viewer](#)

**Exon count:** 14

Annotation release	Status	Assembly	Chr	Location
<a href="#">108</a>	current	GRCm38.p6 ( <a href="#">GCF_000001635.26</a> )	18	NC_000084.6 (67088298..67226792)
Build 37.2	previous assembly	MGSCv37 ( <a href="#">GCF_000001635.18</a> )	18	NC_000084.5 (67247990..67386446)



# Transcript information (Ensembl)

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

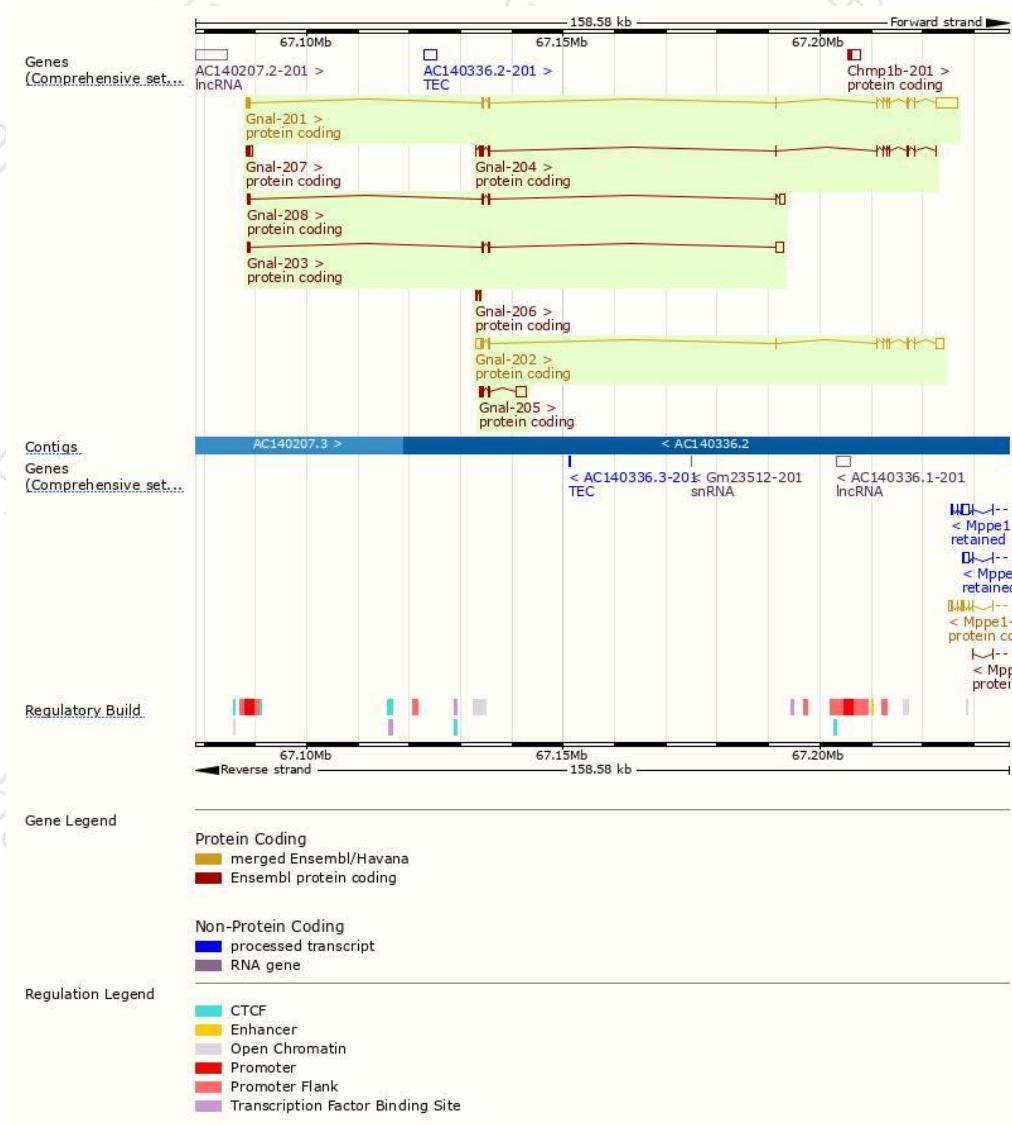
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gnal-201	<a href="#">ENSMUST00000025402.14</a>	5736	<a href="#">448aa</a>	Protein coding	<a href="#">CCDS29317</a>	<a href="#">Q66L47</a>	TSL:1 GENCODE basic
Gnal-202	<a href="#">ENSMUST00000076605.8</a>	3470	<a href="#">381aa</a>	Protein coding	<a href="#">CCDS29318</a>	<a href="#">Q8CGK7</a>	TSL:1 GENCODE basic APPRIS P1
Gnal-204	<a href="#">ENSMUST00000236771.1</a>	1284	<a href="#">381aa</a>	Protein coding	<a href="#">CCDS29318</a>	-	GENCODE basic APPRIS P1
Gnal-205	<a href="#">ENSMUST00000236918.1</a>	2555	<a href="#">134aa</a>	Protein coding	-	<a href="#">Q8C040</a>	GENCODE basic
Gnal-203	<a href="#">ENSMUST00000236551.1</a>	2264	<a href="#">230aa</a>	Protein coding	-	<a href="#">Q8BHK8</a>	GENCODE basic
Gnal-208	<a href="#">ENSMUST00000237985.1</a>	2022	<a href="#">230aa</a>	Protein coding	-	<a href="#">Q8BHK8</a>	GENCODE basic
Gnal-207	<a href="#">ENSMUST00000237445.1</a>	1191	<a href="#">143aa</a>	Protein coding	-	-	GENCODE basic
Gnal-206	<a href="#">ENSMUST00000237304.1</a>	315	<a href="#">13aa</a>	Protein coding	-	-	CDS 3' incomplete

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



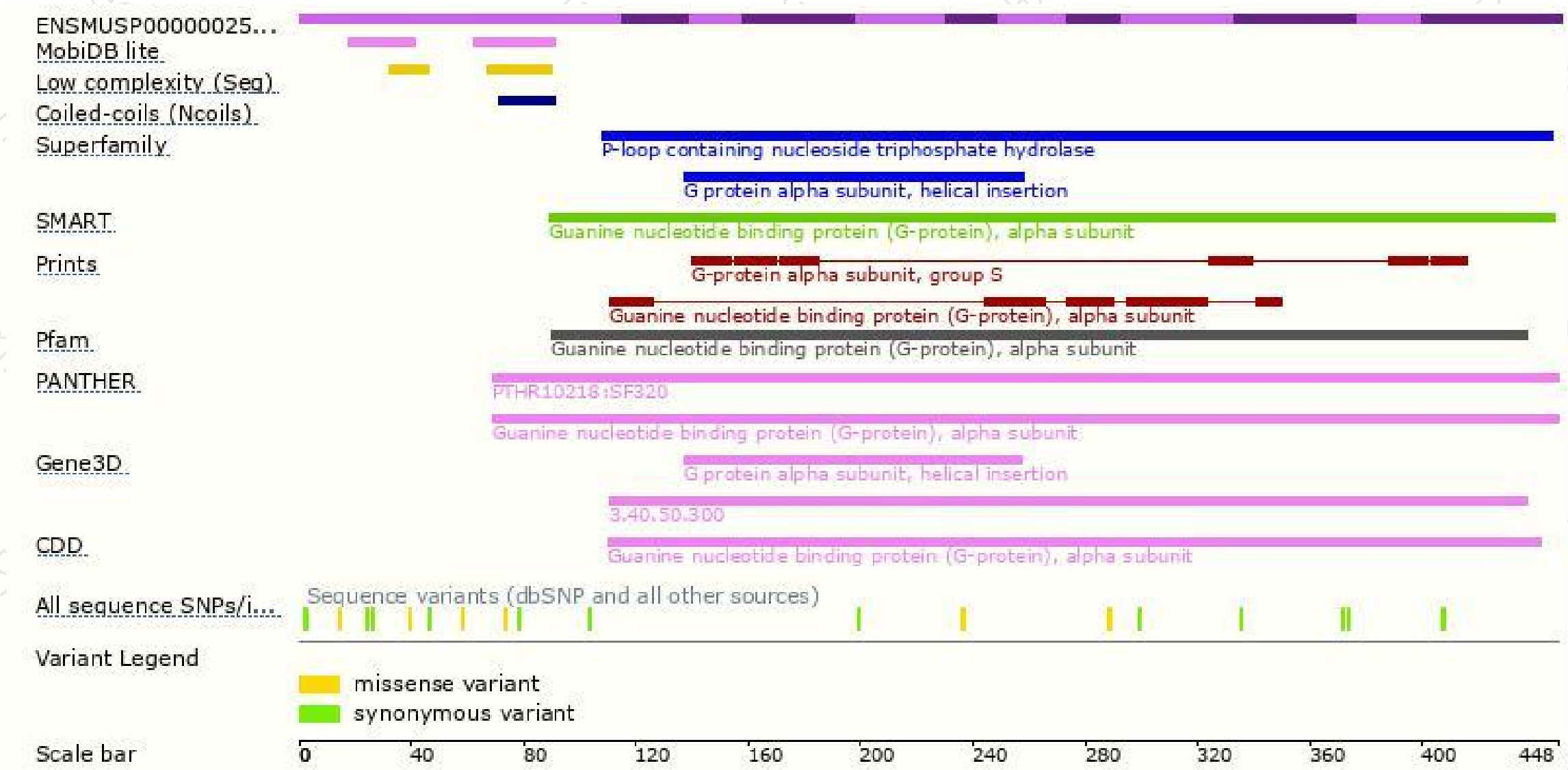


# Genomic location distribution



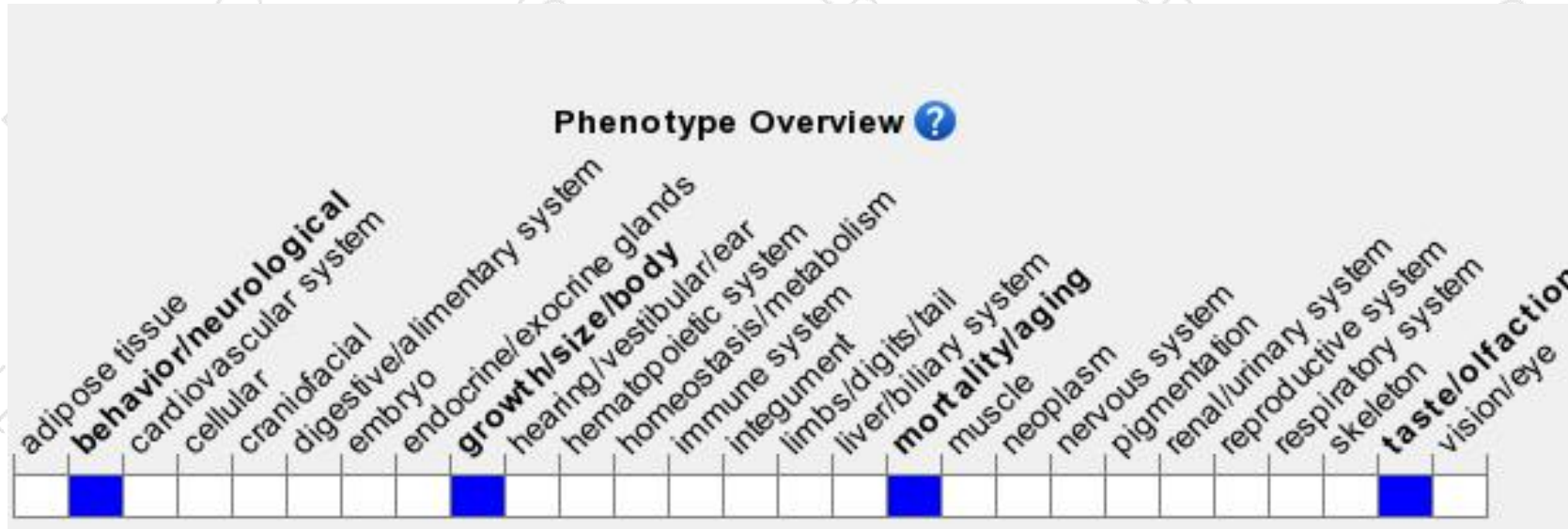


# Protein domain





# 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, Homozygous for a targeted mutation fail to feed, and ~75% die within 2 days after birth. Rare survivors reach sexual maturity and mate but are hyperactive and anosmic, exhibiting severely reduced odor-evoked electrophysical responses and significantly perturbed maternal behaviors.



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

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