

# *Efs* Cas9-CKO Strategy

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**Reviewer:**

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**Design Date:**

**2020-2-18**



# Project Overview

**Project Name**

*Efs*

**Project type**

**Cas9-CKO**

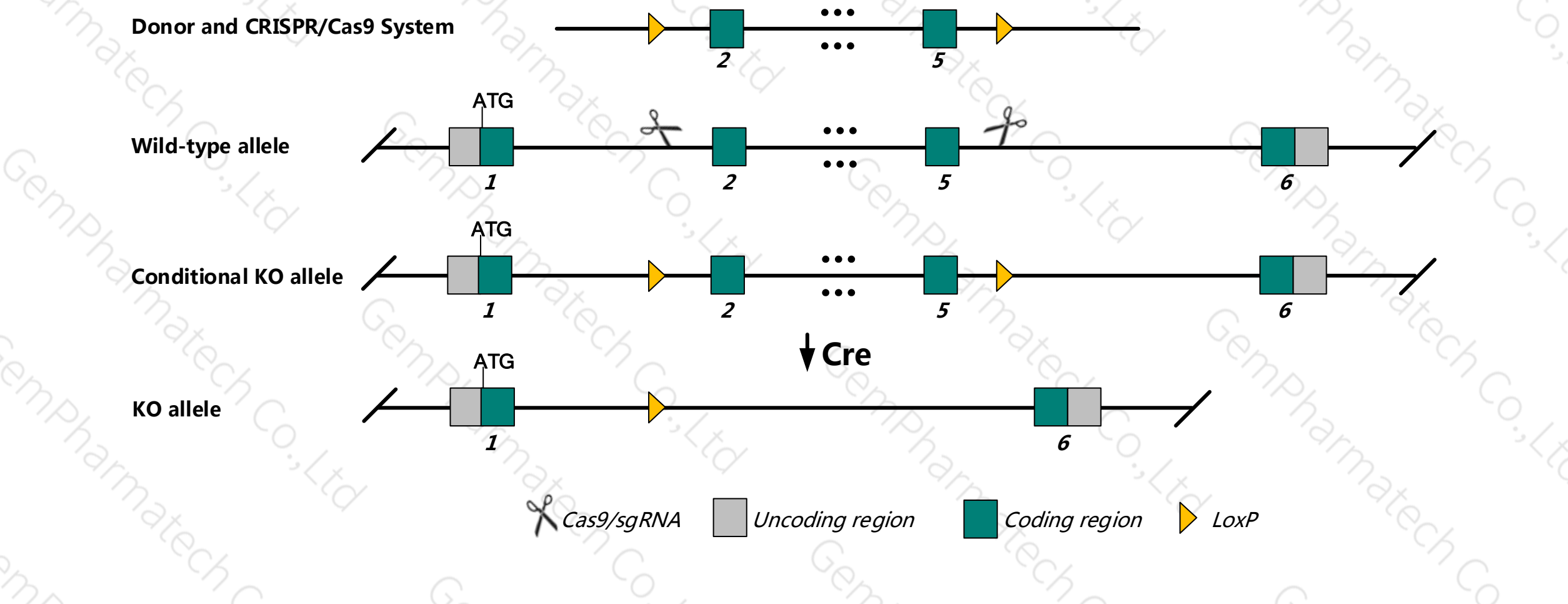
**Strain background**

**C57BL/6JGpt**



# Conditional Knockout strategy

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





- The *Efs* gene has 3 transcripts. According to the structure of *Efs* gene, exon2~exon5 of *Efs*-201 (ENSMUST00000022813.7) transcript is recommended as the knockout region. The region contains most of coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Efs* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.



- According to the existing MGI data , mice homozygous for a disruption in this gene display an increased inflammatory response characterized by excessive T cell responses, enhanced cytokine secretion and antibody production, and intestinal, kidney, liver, and lung inflammation.
- The KO region deletes most of the coding sequence, but does not result in frameshift.
- The *Efs* gene is located on the Chr14. 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.



# Gene information ( NCBI )

## Efs embryonal Fyn-associated substrate [ *Mus musculus* (house mouse) ]

Gene ID: 13644, updated on 12-Aug-2019

### Summary

Official Symbol	Efs provided by <a href="#">MGI</a>
Official Full Name	embryonal Fyn-associated substrate provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:105311</a>
See related	<a href="#">Ensembl:ENSMUSG00000022203</a>
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Broad expression in limb E14.5 (RPKM 32.5), ovary adult (RPKM 23.9) and 18 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

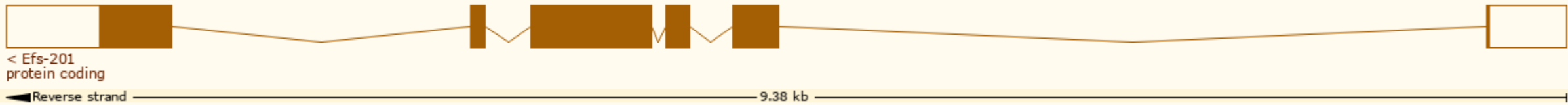


# Transcript information ( Ensembl )

The gene has 3 transcripts, and all transcripts are shown below:

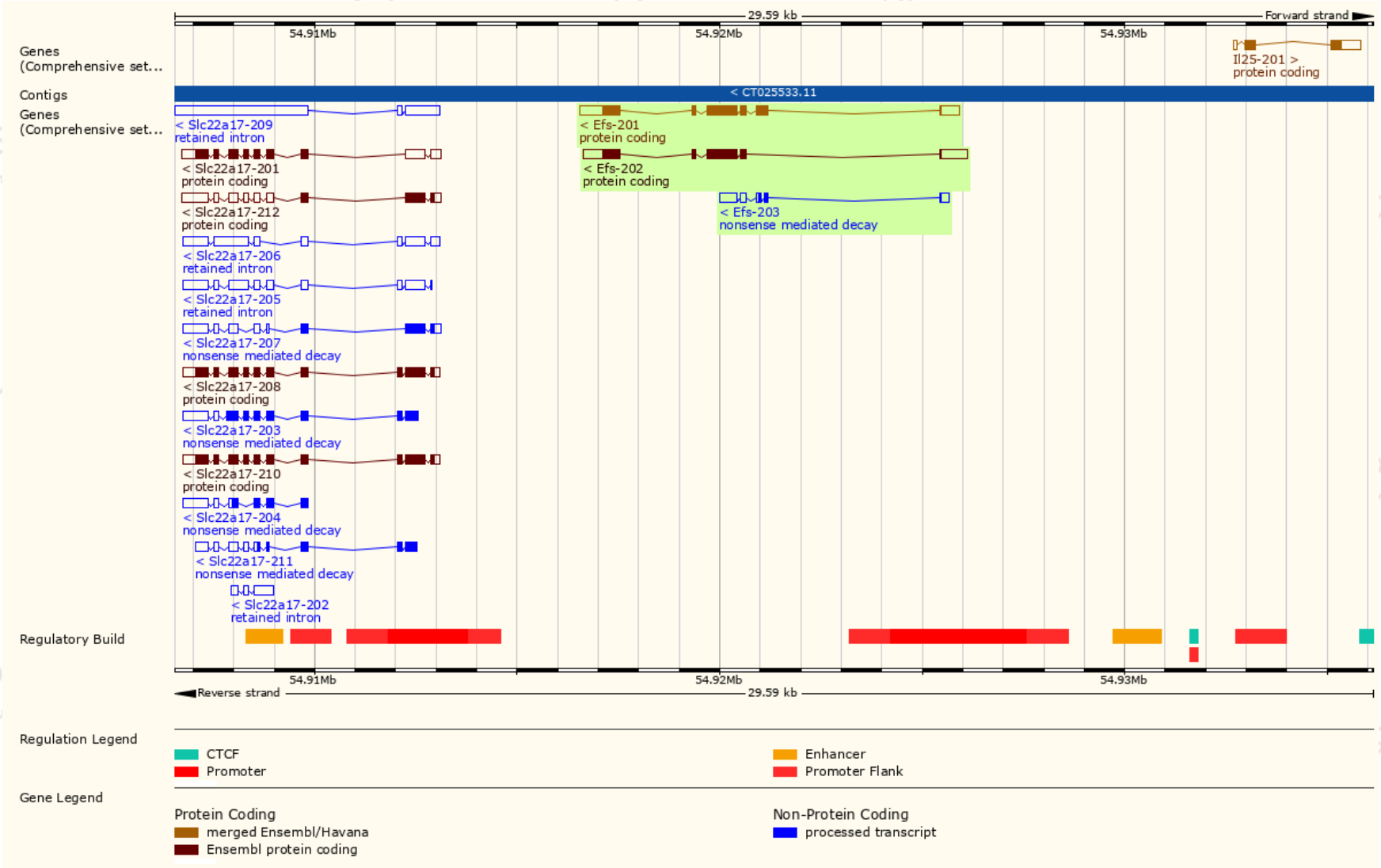
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Efs-201	<a href="#">ENSMUST00000022813.7</a>	2705	<a href="#">560aa</a>	Protein coding	<a href="#">CCDS36925</a>	<a href="#">Q64355</a>	TSL:1 Gencode basic APPRIS P1
Efs-202	<a href="#">ENSMUST000000227037.1</a>	2540	<a href="#">467aa</a>	Protein coding	-	<a href="#">A0A2I3BRP5</a>	Gencode basic
Efs-203	<a href="#">ENSMUST000000227587.1</a>	961	<a href="#">45aa</a>	Nonsense mediated decay	-	<a href="#">A0A2I3BQ52</a>	-

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



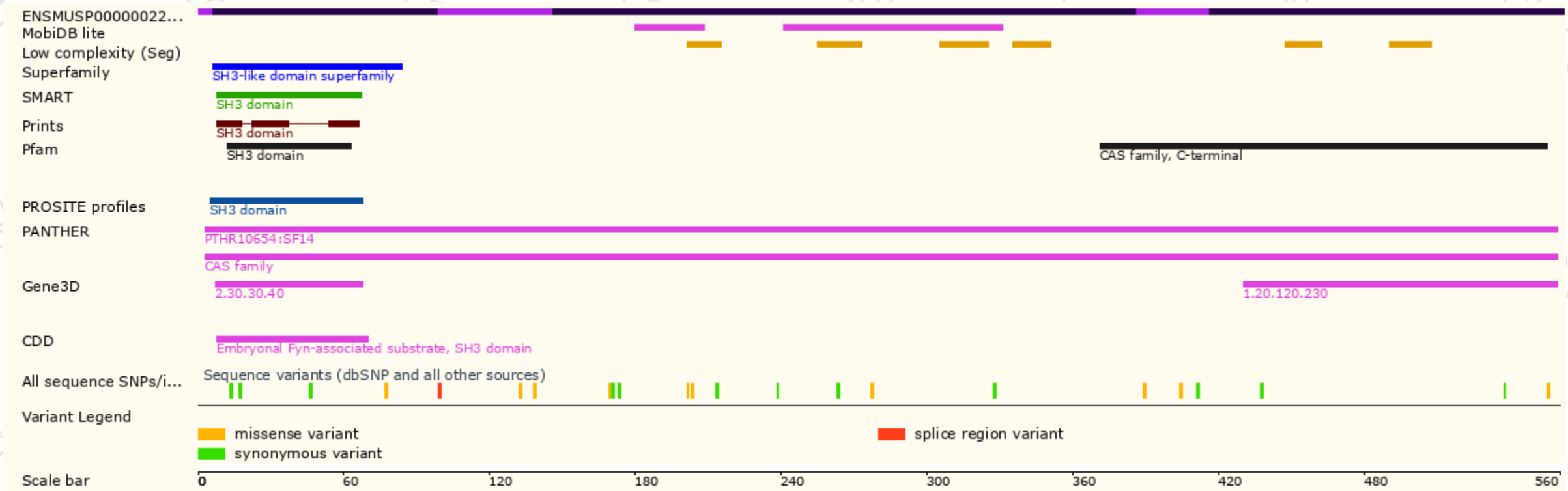


# Genomic location distribution



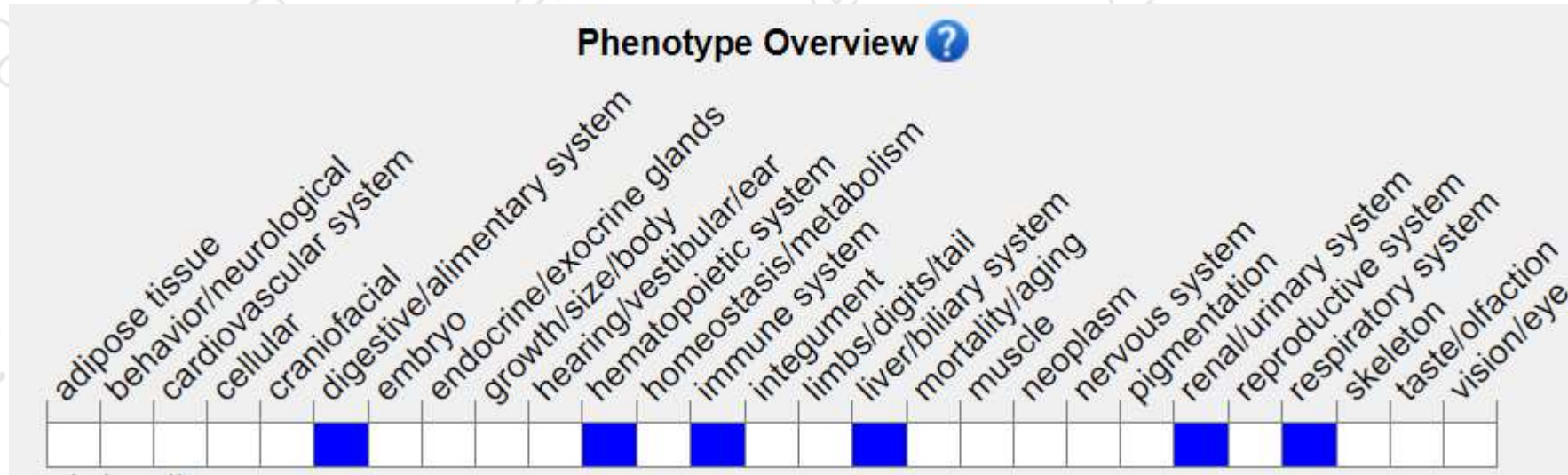


# Protein domain





# 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 disruption in this gene display an increased inflammatory response characterized by excessive T cell responses, enhanced cytokine secretion and antibody production, and intestinal, kidney, liver, and lung inflammation.



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