

# Tspoap1 Cas9-CKO Strategy

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# Overview

## Target Gene Name

- Tspoap1

## Project Type

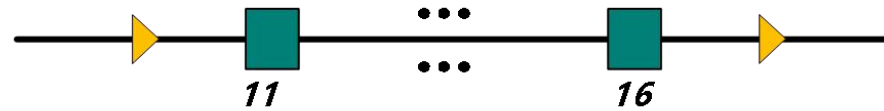
- Cas9-CKO

## Genetic Background

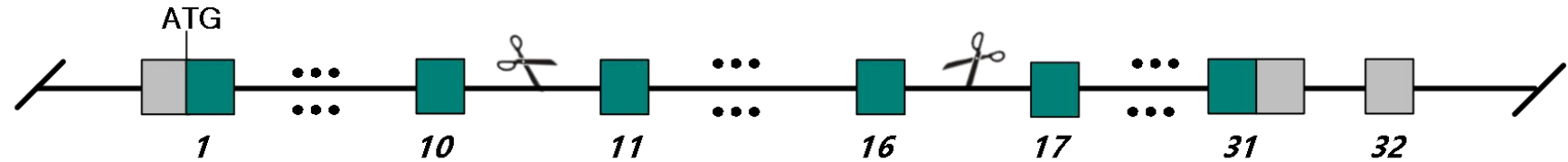
- C57BL/6JGpt

# Strain Strategy

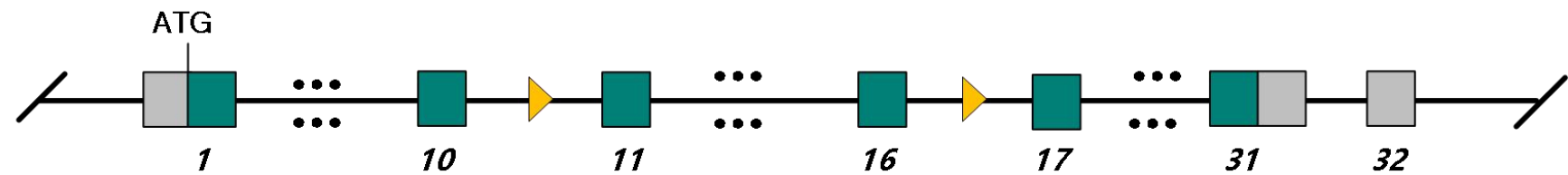
Donor and CRISPR/Cas9 System



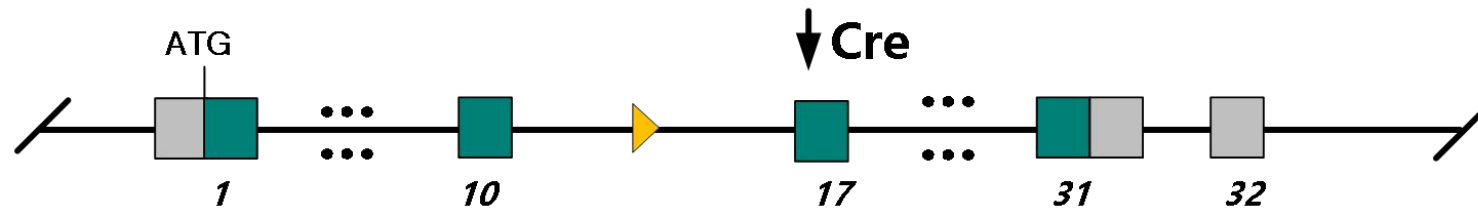
Wild-type allele



Conditional KO allele



KO allele



Schematic representation of CRISPR-Cas9 engineering used to edit the *Tspoap1* gene.

# Technical Information

- The *Tspoap1* gene has 10 transcripts. According to the structure of *Tspoap1* gene, exon11-16 of *Tspoap1*-201 (ENSMUST00000039627.12) transcript is recommended as the knockout region. The region contains 676bp of coding sequences. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Tspoap1* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.

# Gene Information

**Tspoap1** TSPO associated protein 1 [ *Mus musculus* (house mouse) ]

[Download Datasets](#)

Gene ID: 207777, updated on 8-Nov-2022

## Summary

<b>Official Symbol</b>	Tspoap1 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	TSPO associated protein 1 provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:2450877</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000034156</a> <a href="#">AllianceGenome:MGI:2450877</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>	Bzrap1; PRAX-1; mKIAA0612; D230016K05
<b>Summary</b>	Enables voltage-gated calcium channel activity involved in regulation of presynaptic cytosolic calcium levels. Located in calyx of Held. Is active in glutamatergic synapse. Is expressed in lung. Orthologous to human TSPOAP1 (TSPO associated protein 1). [provided by Alliance of Genome Resources, Apr 2022]
<b>Expression</b>	Biased expression in frontal lobe adult (RPKM 29.5), cortex adult (RPKM 26.7) and 7 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>
<b>NEW</b>	Try the new <a href="#">Gene table</a> Try the new <a href="#">Transcript table</a>

Source: <https://www.ncbi.nlm.nih.gov/>

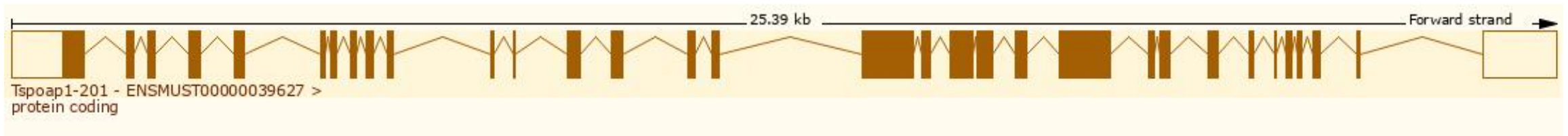


# Transcript Information

The gene has 10 transcripts, all transcripts are shown below:

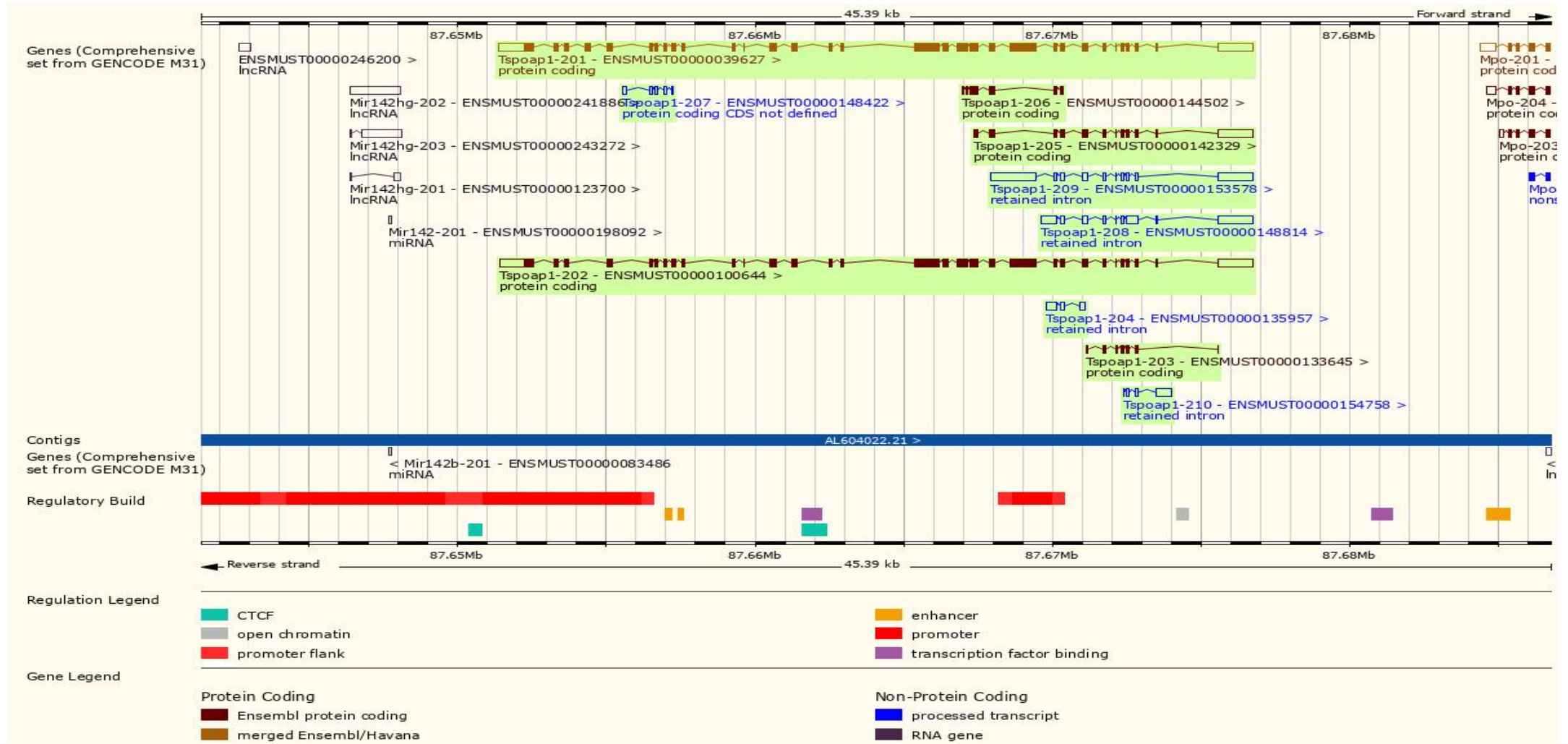
Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
<a href="#">ENSMUST00000039627.12</a>	Tspoap1-201	7616	<a href="#">1846aa</a>	Protein coding	<a href="#">CCDS48879</a>	<a href="#">Q7TNF8-1</a>	Ensembl Canonical GENCODE basic APPRIS ALT2 TSL:5
<a href="#">ENSMUST00000100644.10</a>	Tspoap1-202	7390	<a href="#">1786aa</a>	Protein coding		<a href="#">Q5NCP6</a>	GENCODE basic APPRIS P4 TSL:5
<a href="#">ENSMUST00000142329.8</a>	Tspoap1-205	2367	<a href="#">384aa</a>	Protein coding		<a href="#">F6WV49</a>	TSL:1 CDS 5' incomplete
<a href="#">ENSMUST00000144502.8</a>	Tspoap1-206	752	<a href="#">251aa</a>	Protein coding		<a href="#">F7CD74</a>	TSL:5 CDS 5' and 3' incomplete
<a href="#">ENSMUST00000133645.2</a>	Tspoap1-203	506	<a href="#">169aa</a>	Protein coding		<a href="#">F7AUI0</a>	TSL:5 CDS 5' and 3' incomplete
<a href="#">ENSMUST00000148422.2</a>	Tspoap1-207	386	No protein	Protein coding CDS not defined		-	TSL:2
<a href="#">ENSMUST00000153578.8</a>	Tspoap1-209	3514	No protein	Retained intron		-	TSL:1
<a href="#">ENSMUST00000148814.8</a>	Tspoap1-208	2678	No protein	Retained intron		-	TSL:2
<a href="#">ENSMUST00000154758.2</a>	Tspoap1-210	732	No protein	Retained intron		-	TSL:3
<a href="#">ENSMUST00000135957.2</a>	Tspoap1-204	714	No protein	Retained intron		-	TSL:3

The strategy is based on the design of *Tspoap1*-201 transcript, the transcription is shown below:

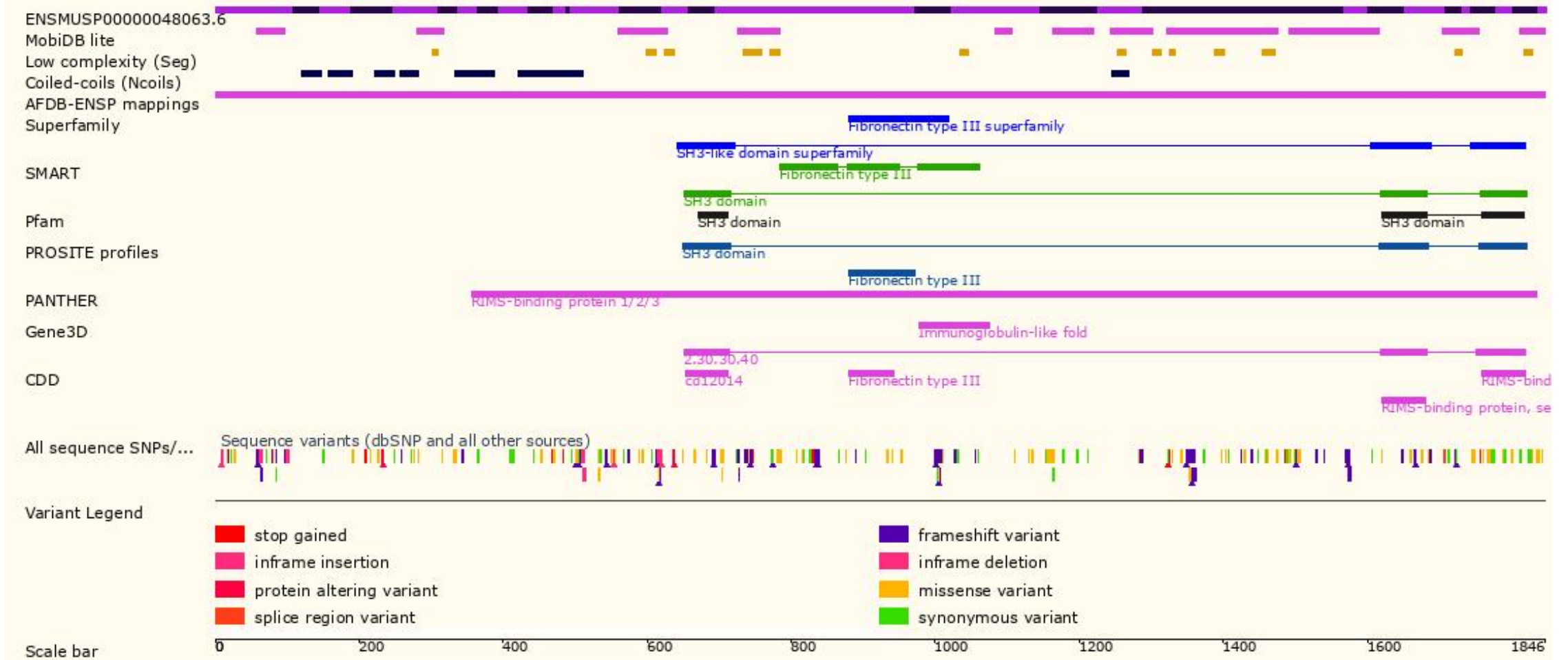


Source: <https://www.ensembl.org>

# Genomic Information

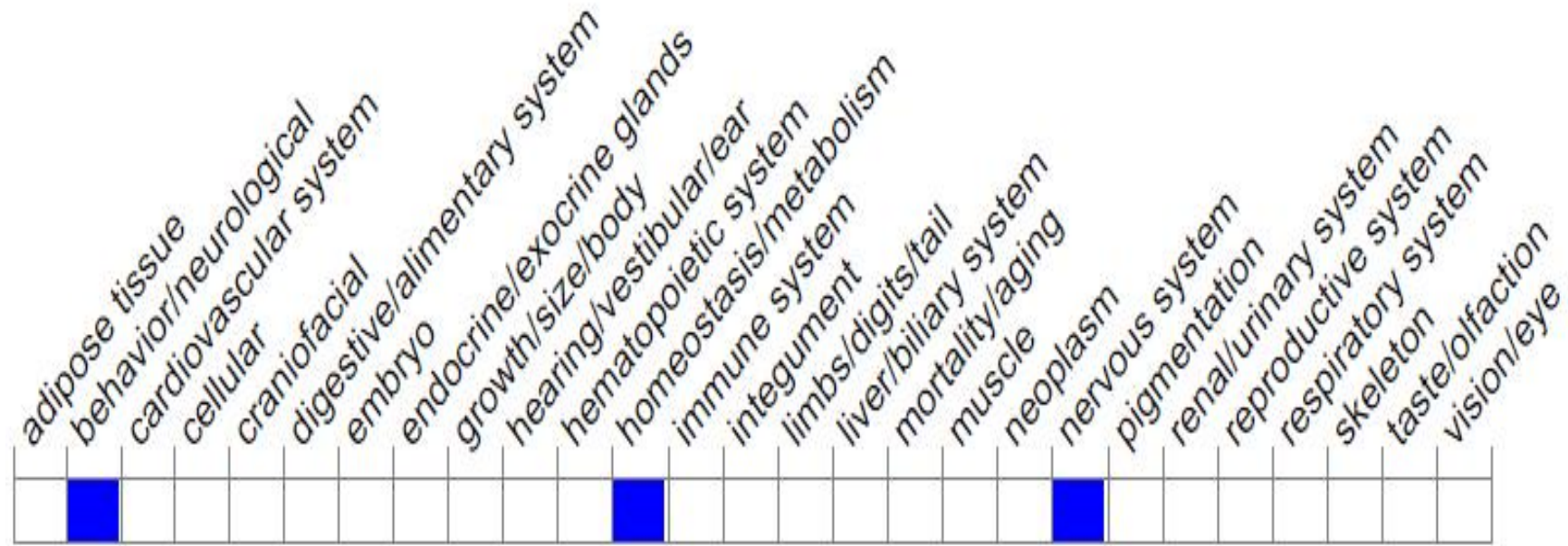


# Protein Information





# Mouse Phenotype Information (MGI)



- Homozygous double-KO with  $Rimbp2^{tm1.2Geno}$  does not exacerbate the phenotype of the latter single KO.

# Important Information

- Transcript *Tspoap1*-205&206&203 may not be affected.
- *Tspoap1* is located on Chr11. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.