

# ***Pum2 Cas9-KO Strategy***

**Designer:**

**Daohua Xu**

**Reviewer :**

**Huimin Su**

**Design Date:**

**2019-9-28**

# Project Overview

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**Project Name**

*Pum2*

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**Project type**

Cas9-KO

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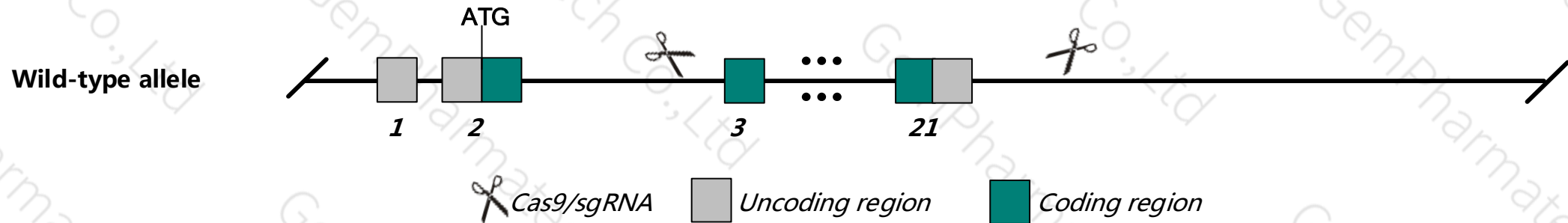
**Animal background**

C57BL/6JGpt

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# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pum2* gene. The schematic diagram is as follows:



# Technical routes

- The *Pum2* gene has 16 transcripts, According to the structure of *Pum2* gene, exon3-exon21 of *Pum2*-204 transcript is recommended as the knockout region. The region contains the most of coding sequence. Knock out the region, result in destruction of protein.
- This project uses CRISPR/Cas9 technology to modify *Pum2* gene. The brief process is as follows: sgRNA was transcribed in vitro, Cas9, sgRNA were microinjected into fertilized eggs of C57BL/6JGpt mice and homologous recombination was carried out to obtain F0 mice. A stable and hereditary F1 generation mouse model was obtained by mating F0 generation mice with C57BL/6JGpt mice which were confirmed positive by PCR-sequencing.

- According to the existing MGI data , Mice homozygous for a gene trapped allele exhibit significantly smaller testes and seminiferous tubule degeneration but are otherwise viable and fertile.
- The *Pum2* gene is located in the Chr12. If the knockout mice are mixed with other mice, two target genes are avoided on the same chromosome as possible, otherwise the offspring of mice with double gene positive and homozygous gene knockout can not be obtained.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of gene transcription and translation processes, all risks cannot be predicted under existing information.

# Gene information ( NCBI )



## Pum2 pumilio RNA-binding family member 2 [ *Mus musculus* (house mouse) ]

Gene ID: 80913, updated on 15-Jan-2019


Summary

Official Symbol	Pum2 provided by MGI
Official Full Name	pumilio RNA-binding family member 2 provided by MGI
Primary source	MGI:MGI:1931751
See related	Ensembl:ENSMUSG00000020594
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
Also known as	Pumm2; 5730503J23Rik
Expression	Ubiquitous expression in thymus adult (RPKM 23.4), whole brain E14.5 (RPKM 23.1) and 28 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

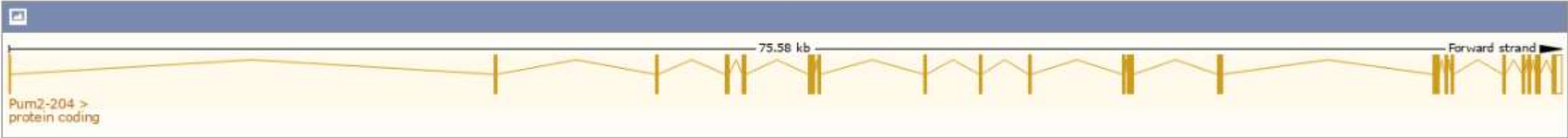


# Transcript information ( Ensembl )

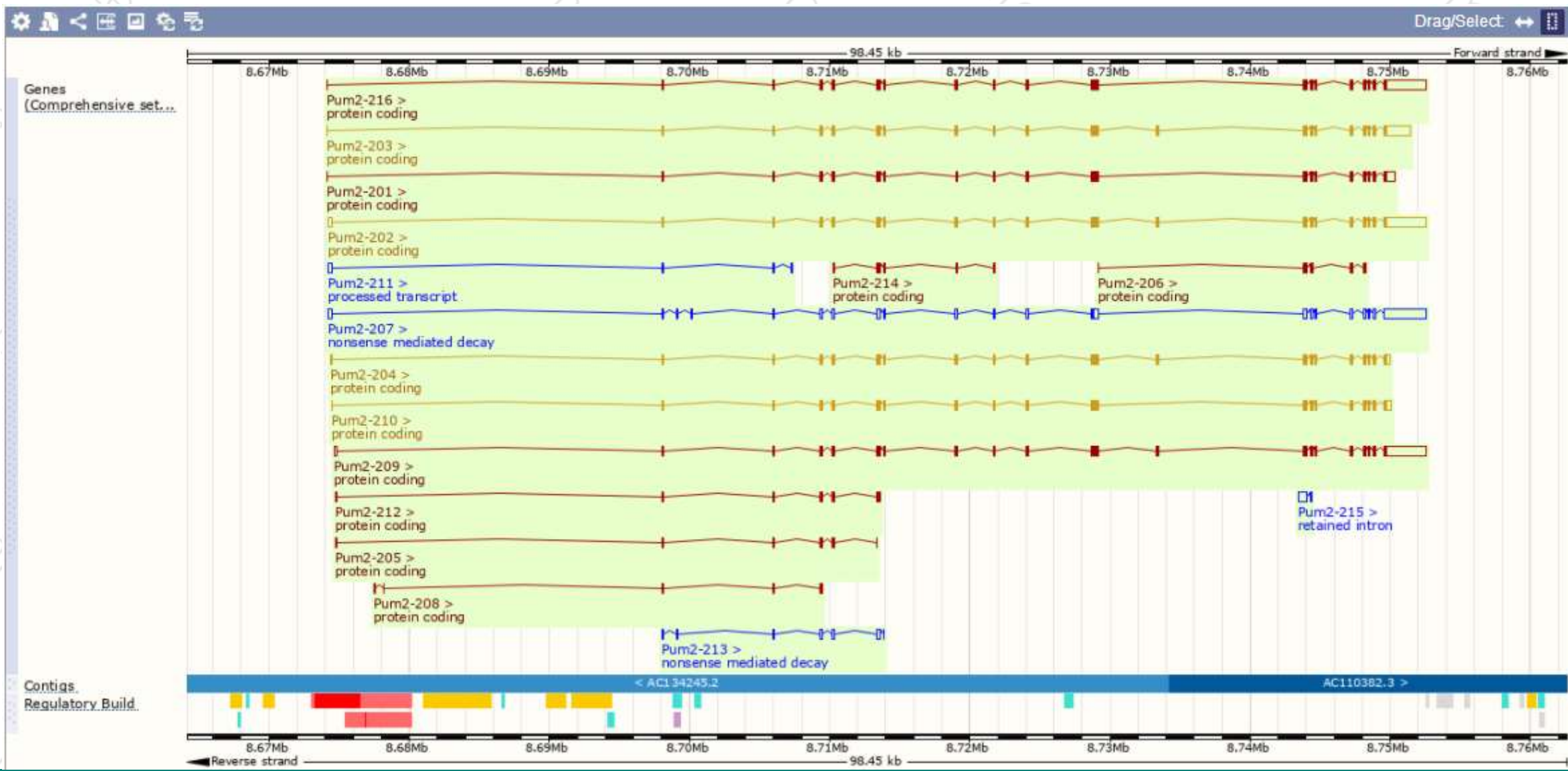
The gene has 16 transcripts, and all transcripts are shown below :

Show/hide columns (1 hidden)										Filter			
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags					
Pum2-202	<a href="#">ENSMUST00000111122.8</a>	6311	<a href="#">1064aa</a>	 Protein coding	<a href="#">CCDS49025</a>	<a href="#">Q3UR91</a>	<a href="#">NM_001160220</a> <a href="#">NP_001153692</a>	TSL:1	GENCODE basic	APPRIS ALT2			
Pum2-209	<a href="#">ENSMUST00000168361.7</a>	6226	<a href="#">1066aa</a>	 Protein coding	<a href="#">CCDS25802</a>	<a href="#">Q80U58</a>	<a href="#">NM_030723</a> <a href="#">NP_109648</a>	TSL:5	GENCODE basic	APPRIS P3			
Pum2-216	<a href="#">ENSMUST00000178015.7</a>	5874	<a href="#">985aa</a>	 Protein coding	<a href="#">CCDS49026</a>	<a href="#">Q80U58</a>	<a href="#">NM_001160222</a> <a href="#">NP_001153694</a>	TSL:5	GENCODE basic	APPRIS ALT2			
Pum2-203	<a href="#">ENSMUST00000111123.8</a>	4984	<a href="#">1064aa</a>	 Protein coding	<a href="#">CCDS49025</a>	<a href="#">Q3UR91</a>	<a href="#">NM_001160221</a> <a href="#">NP_001153693</a>	TSL:1	GENCODE basic	APPRIS ALT2			
Pum2-201	<a href="#">ENSMUST00000020915.9</a>	3587	<a href="#">980aa</a>	 Protein coding	<a href="#">CCDS79105</a>	<a href="#">Q3TQ29</a>	<a href="#">NM_001310519</a> <a href="#">NP_001297448</a>	TSL:1 GENCODE basic					
Pum2-204	<a href="#">ENSMUST00000163569.7</a>	3560	<a href="#">1066aa</a>	 Protein coding	<a href="#">CCDS25802</a>	<a href="#">Q80U58</a>	<a href="#">NM_001160219</a> <a href="#">NP_001153691</a>	TSL:1	GENCODE basic	APPRIS P3			
Pum2-210	<a href="#">ENSMUST00000169089.7</a>	3295	<a href="#">985aa</a>	 Protein coding	<a href="#">CCDS49026</a>	<a href="#">Q80U58</a>	-	TSL:5	GENCODE basic	APPRIS ALT2			
Pum2-212	<a href="#">ENSMUST00000169750.7</a>	902	<a href="#">262aa</a>	 Protein coding	-	<a href="#">E9PW51</a>	-	CDS 3' incomplete TSL:5					
Pum2-214	<a href="#">ENSMUST00000171418.2</a>	706	<a href="#">235aa</a>	 Protein coding	-	<a href="#">F7A034</a>	-	CDS 5' and 3' incomplete TSL:3					
Pum2-206	<a href="#">ENSMUST00000164818.2</a>	665	<a href="#">222aa</a>	 Protein coding	-	<a href="#">F6YD01</a>	-	CDS 5' and 3' incomplete TSL:5					
Pum2-205	<a href="#">ENSMUST00000163730.7</a>	648	<a href="#">185aa</a>	 Protein coding	-	<a href="#">E9Q4Q6</a>	-	CDS 3' incomplete TSL:5					
Pum2-208	<a href="#">ENSMUST00000166965.1</a>	472	<a href="#">109aa</a>	 Protein coding	-	<a href="#">E9Q765</a>	-	CDS 3' incomplete TSL:3					
Pum2-207	<a href="#">ENSMUST00000165293.7</a>	6218	<a href="#">35aa</a>	 Nonsense mediated decay	-	<a href="#">E9Q0P1</a>	<a href="#">NR_027670</a>	TSL:1					
Pum2-213	<a href="#">ENSMUST00000170037.1</a>	994	<a href="#">35aa</a>	 Nonsense mediated decay	-	<a href="#">E9Q0P1</a>	-	TSL:5					
Pum2-211	<a href="#">ENSMUST00000169177.7</a>	518	No protein	 Processed transcript	-	-	-	TSL:2					
Pum2-215	<a href="#">ENSMUST00000171608.1</a>	612	No protein	 Retained intron	-	-	-	TSL:2					

The strategy is based on the design of *Pum2-204* transcript,The transcription is shown below :

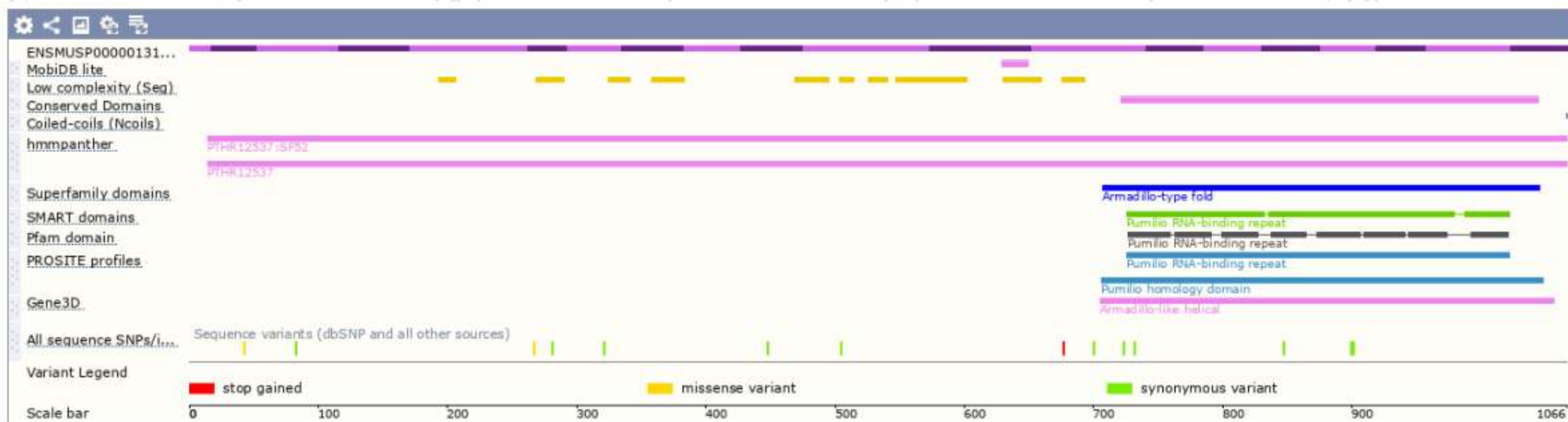


# Genomic location distribution

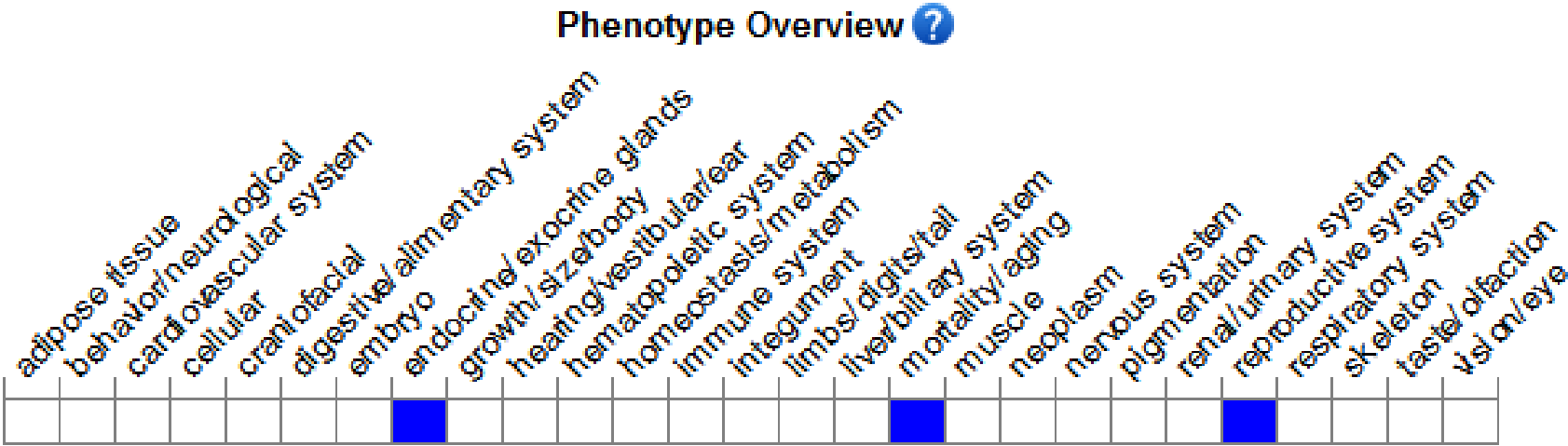




# Protein domain



# Mouse phenotype description(MGI )



Click cells to view annotations.

*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 gene trapped allele exhibit significantly smaller testes and seminiferous tubule degeneration but are otherwise viable and fertile.

If you have any questions, you are welcome to inquire.  
Tel: 025-5864 1534

