

# *Insr* Cas9-CKO Strategy

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# Project Overview

**Project Name**

*Insr*

**Project type**

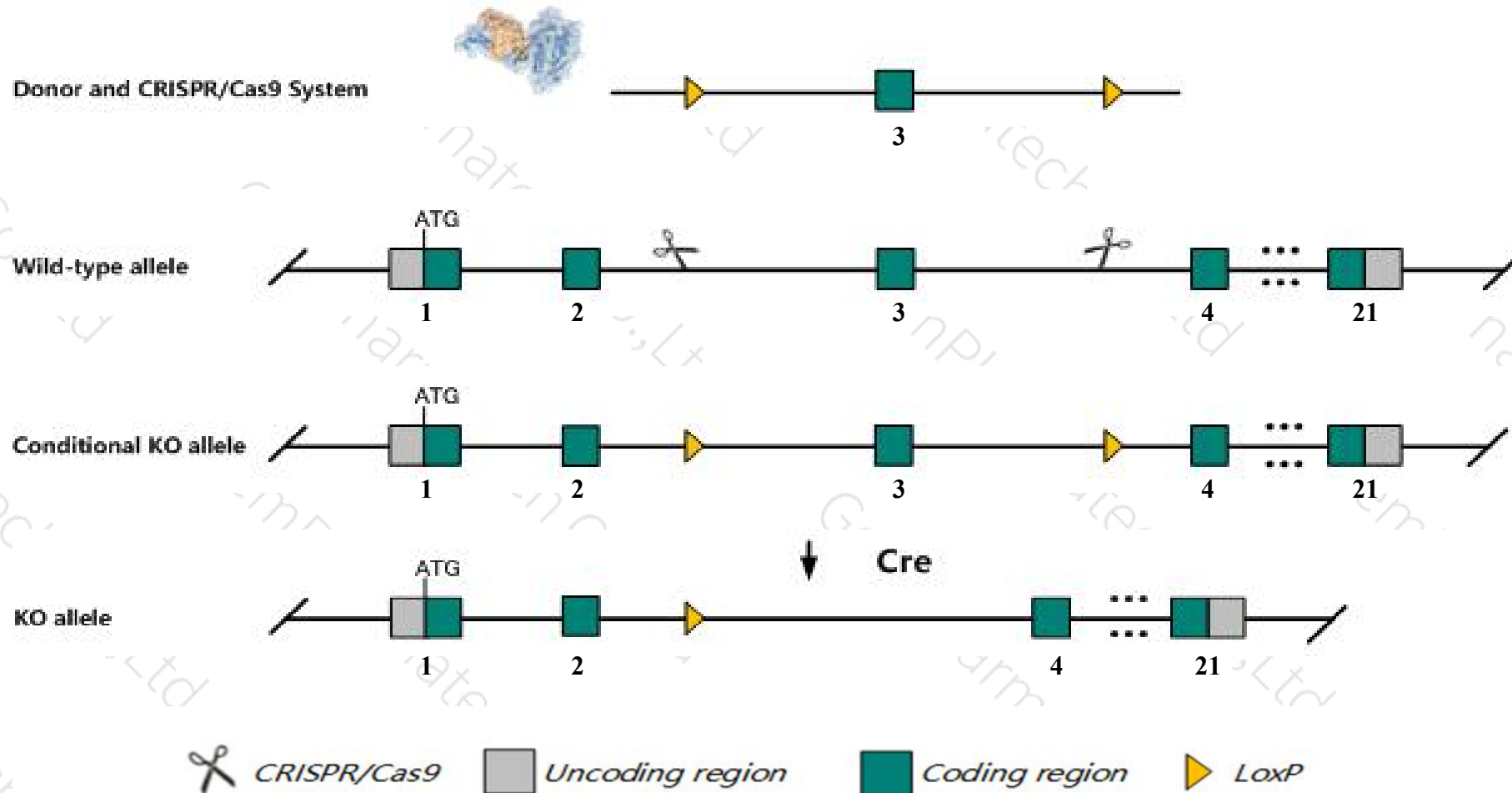
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

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



- The *Insr* gene has 5 transcripts. According to the structure of *Insr* gene, exon3 of *Insr-201* (ENSMUST00000091291.4) transcript is recommended as the knockout region. The region contains 322bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Insr* 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, Null mutants grow slowly and die by 7 days of age with ketoacidosis, high serum insulin and triglycerides, low glycogen stores and fatty livers. Tissue specific knockouts show milder lipid metabolism anomalies. Point mutation heterozygotes exhibit hyperglycemia, hyperinsulinemia and glucosuria.
- The *Insr* gene is located on the Chr8. 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)

## Insr insulin receptor [ *Mus musculus* (house mouse) ]

Gene ID: 16337, updated on 27-Aug-2019

### Summary

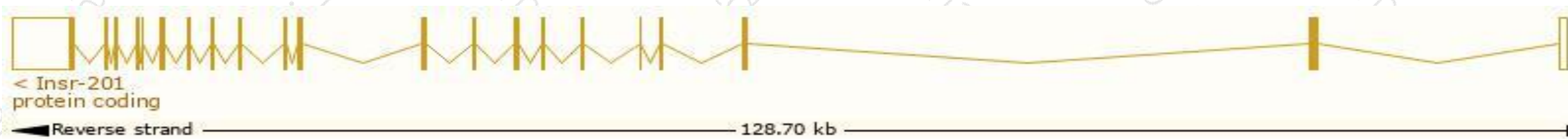
<b>Official Symbol</b>	Insr provided by <a href="#">MGI</a>
<b>Official Full Name</b>	insulin receptor provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:96575</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000005534</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	REVIEWED
<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>	IR; IR-A; IR-B; CD220; 4932439J01Rik; D630014A15Rik
<b>Summary</b>	This gene encodes a member of the receptor tyrosine kinase family of transmembrane signaling proteins that play important roles in cell differentiation, growth and metabolism. The encoded preproprotein undergoes proteolytic processing to generate alpha and beta chains that form a disulfide-linked heterodimer which, in turn homodimerizes to form a mature, functional receptor. Mice lacking the encoded protein develop severe hyperglycemia and hyperketonemia, and die within a couple of days after birth as a result of diabetic ketoacidosis. [provided by RefSeq, Aug 2016]
<b>Expression</b>	Ubiquitous expression in heart adult (RPKM 8.5), adrenal adult (RPKM 7.8) and 28 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

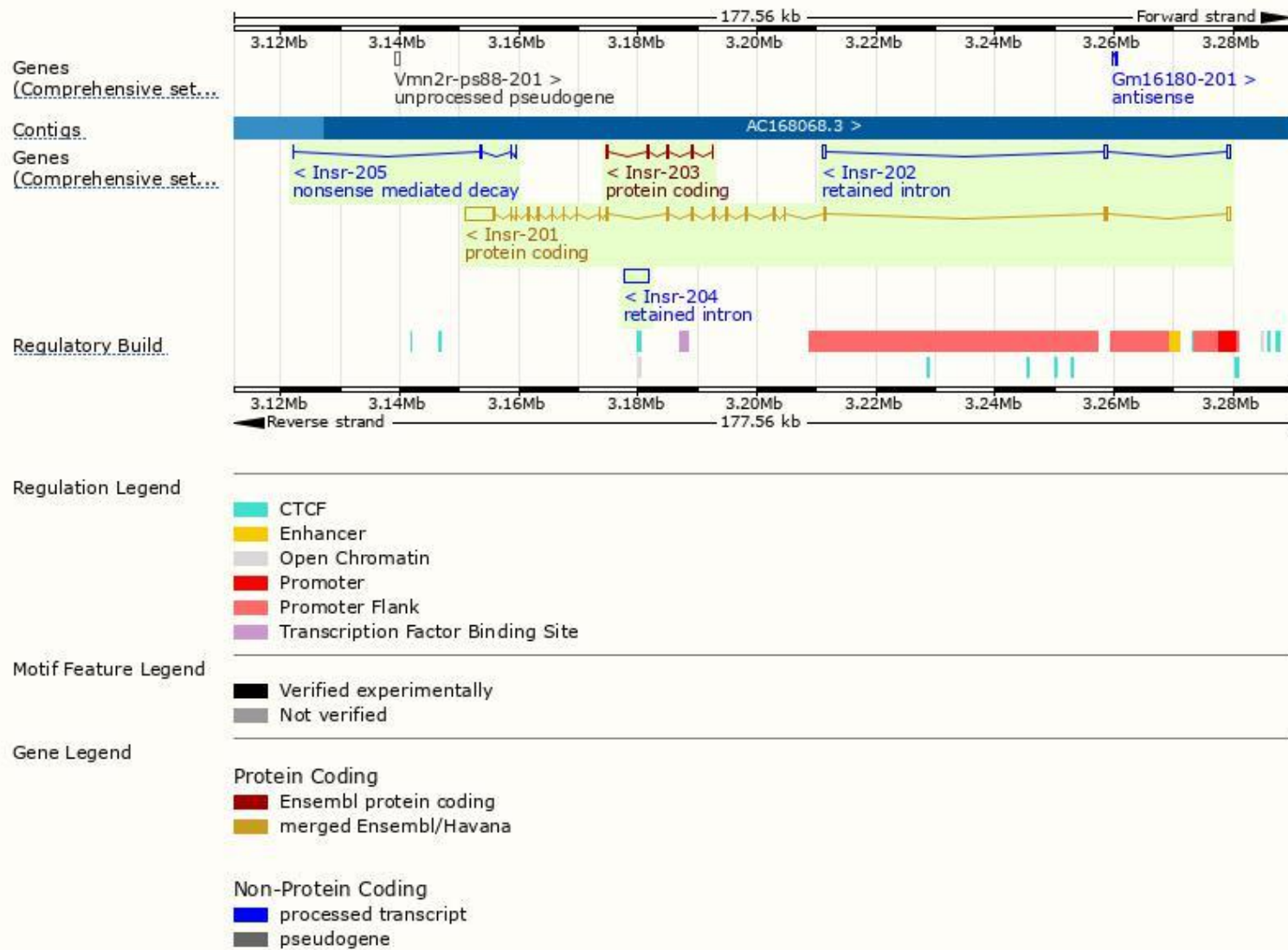
The gene has 5 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Insr-201	<a href="#">ENSMUST00000091291.4</a>	9355	<a href="#">1372aa</a>	Protein coding	<a href="#">CCDS22059</a>	<a href="#">P15208</a>	TSL:1 GENCODE basic APPRIS P1
Insr-203	<a href="#">ENSMUST00000207100.1</a>	672	<a href="#">224aa</a>	Protein coding	-	<a href="#">B8Q3N4</a>	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:1
Insr-205	<a href="#">ENSMUST00000208839.1</a>	551	<a href="#">90aa</a>	Nonsense mediated decay	-	<a href="#">A0A140LI30</a>	CDS 5' incomplete TSL:3
Insr-204	<a href="#">ENSMUST00000207295.1</a>	4019	No protein	Retained intron	-	-	TSL:NA
Insr-202	<a href="#">ENSMUST00000139504.1</a>	1675	No protein	Retained intron	-	-	TSL:1

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



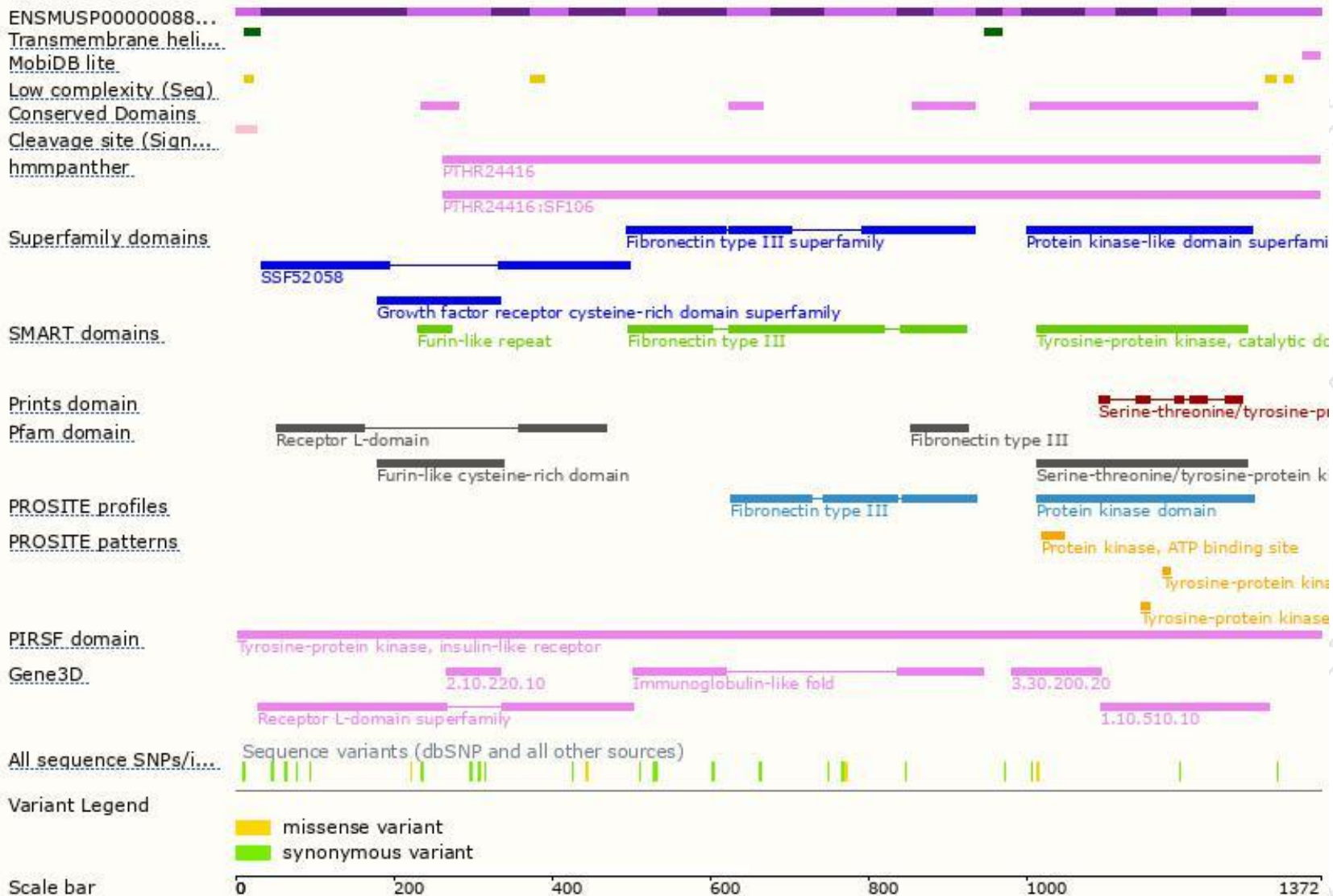
# Genomic location distribution



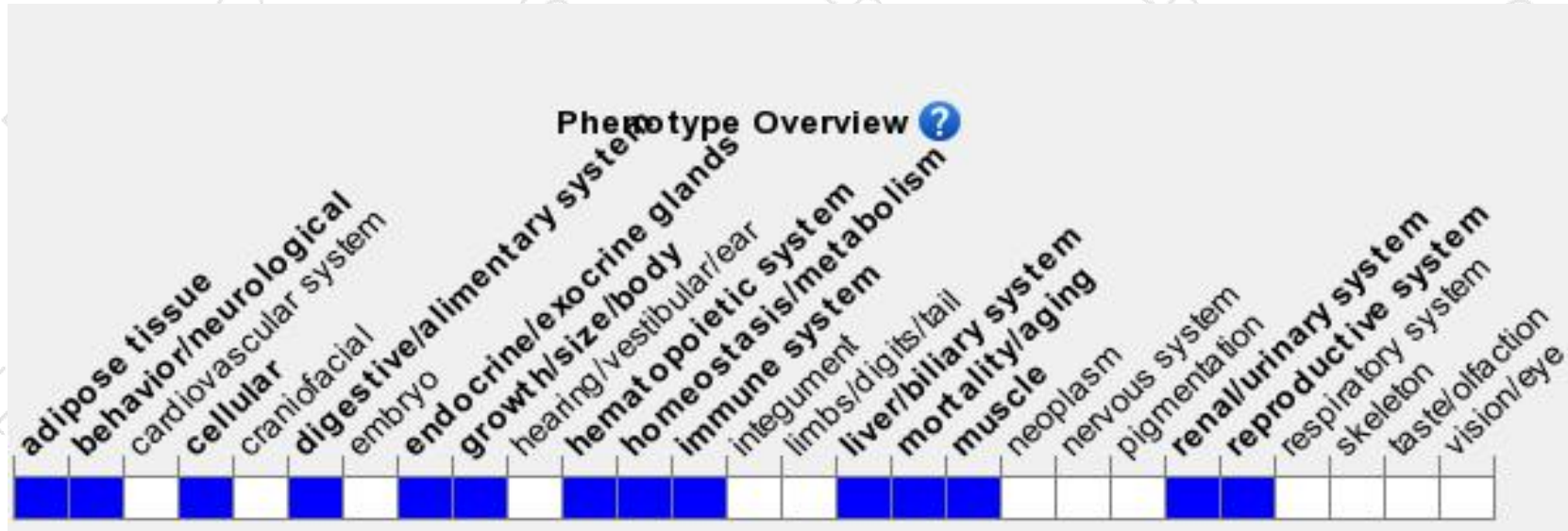
# Protein domain



集萃药康  
GemPharmatech



# 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, Null mutants grow slowly and die by 7 days of age with ketoacidosis, high serum insulin and triglycerides, low glycogen stores and fatty livers. Tissue specific knockouts show milder lipid metabolism anomalies. Point mutation heterozygotes exhibit hyperglycemia, hyperinsulinemia and glucosuria.

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

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