

# ***Lepr*** Cas9-CKO Strategy

**Designer:**

**Huan Fan**

**Reviewer:**

**Yun Li**

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# Project Overview

**Project Name**

*Lepr*

**Project type**

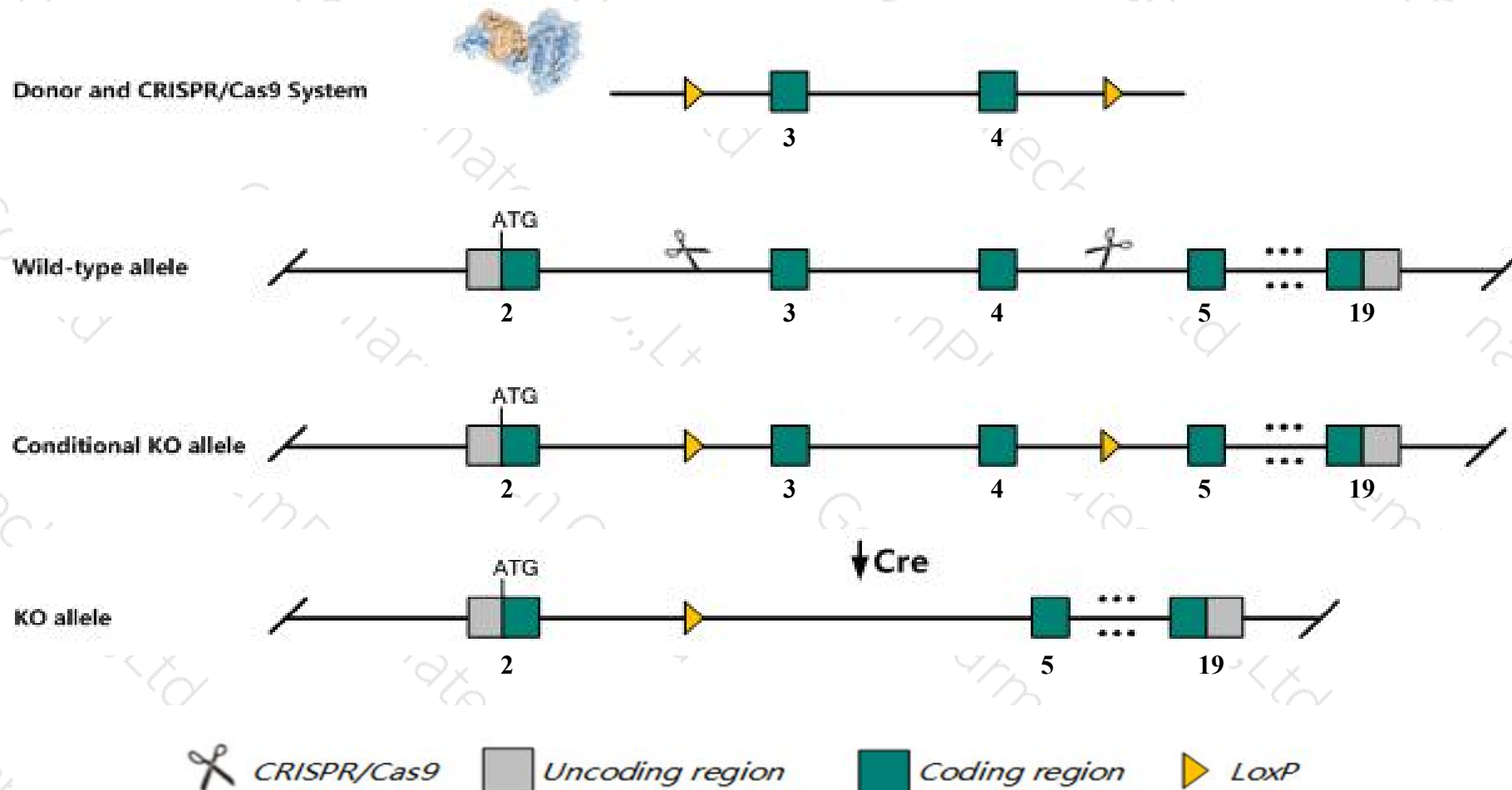
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

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



- The *Lepr* gene has 7 transcripts. According to the structure of *Lepr* gene, exon3-exon4 of *Lepr*-203 (ENSMUST00000106921.8) transcript is recommended as the knockout region. The region contains 454bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Lepr* 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 mutants are hyperphagic, low-activity, poorly cold-adapted, sterile and have enhanced fat conversion. They are obese, hyperinsulinemic and, on certain strains, severely hyperglycemic. Heterozygotes are normal but resistant to prolonged fasting.
- The *Lepr* gene is located on the Chr4. 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)

## Lepr leptin receptor [Mus musculus (house mouse)]

Gene ID: 16847, updated on 9-Apr-2019

### Summary



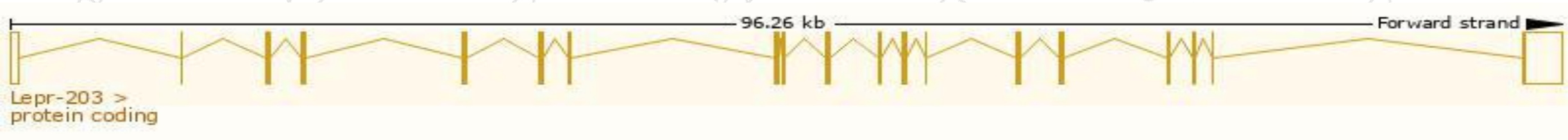
<b>Official Symbol</b>	Lepr provided by <a href="#">MGI</a>
<b>Official Full Name</b>	leptin receptor provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:104993</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000057722</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	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>	LEPROT, Leprb, Modb1, OB-RGRP, Obr, db, diabetes, obese-like, obl
<b>Expression</b>	Broad expression in bladder adult (RPKM 5.0), placenta adult (RPKM 3.3) and 18 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

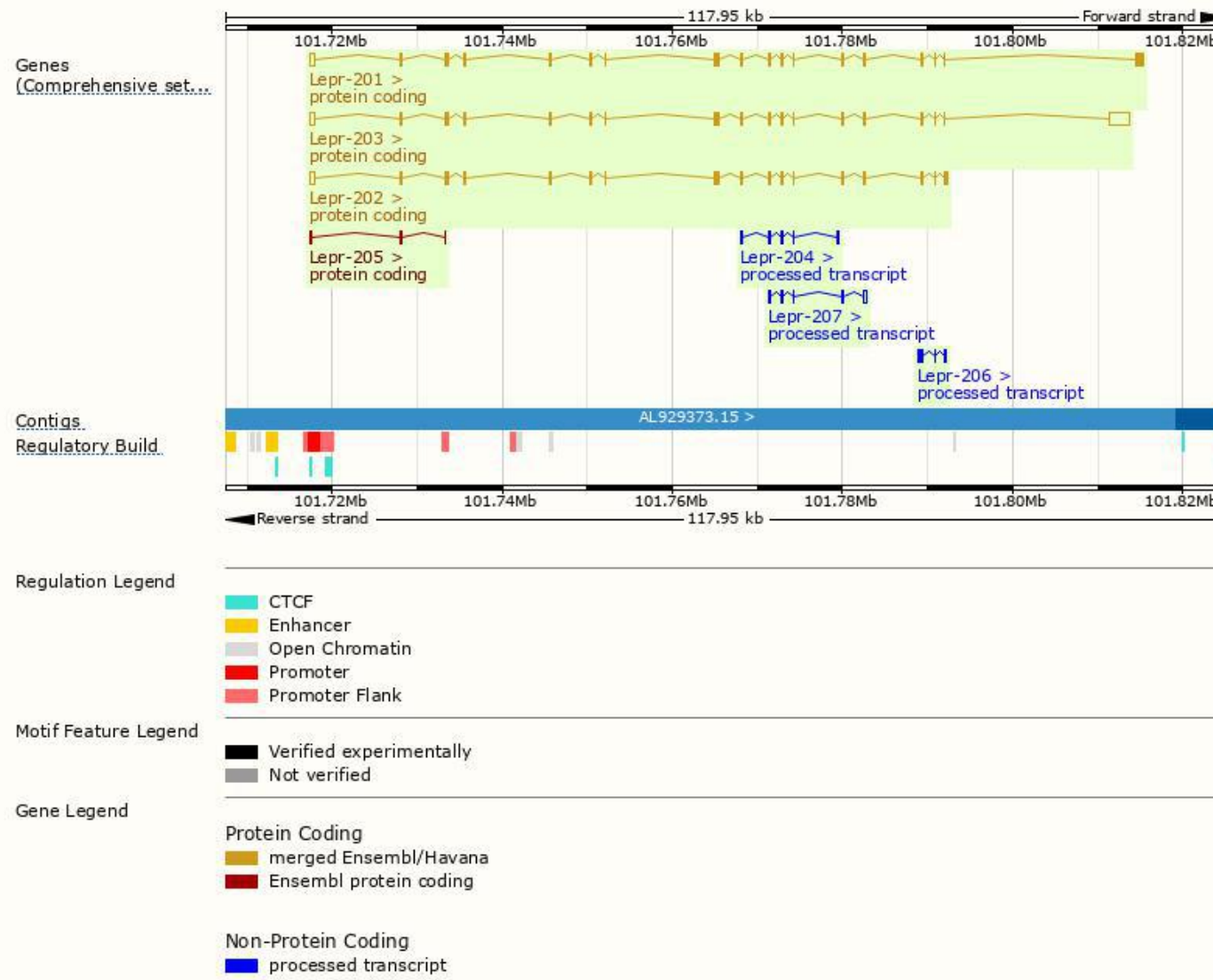
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Lepr-203	<a href="#">ENSMUST00000106921.8</a>	5542	<a href="#">894aa</a>	Protein coding	<a href="#">CCDS51239</a>	<a href="#">P48356 Q3US58</a>	TSL:1 GENCODE basic APPRIS ALT2
Lepr-201	<a href="#">ENSMUST00000037552.9</a>	4127	<a href="#">1162aa</a>	Protein coding	<a href="#">CCDS51240</a>	<a href="#">P48356</a>	TSL:1 GENCODE basic APPRIS ALT2
Lepr-202	<a href="#">ENSMUST00000102777.9</a>	3410	<a href="#">892aa</a>	Protein coding	<a href="#">CCDS18397</a>	<a href="#">P48356 Q3UNU8</a>	TSL:1 GENCODE basic APPRIS P3
Lepr-205	<a href="#">ENSMUST00000145024.1</a>	208	<a href="#">30aa</a>	Protein coding	-	<a href="#">A2AV66</a>	CDS 3' incomplete TSL:5
Lepr-207	<a href="#">ENSMUST00000156402.1</a>	889	No protein	Processed transcript	-	-	TSL:2
Lepr-204	<a href="#">ENSMUST00000128948.7</a>	787	No protein	Processed transcript	-	-	TSL:3
Lepr-206	<a href="#">ENSMUST00000151733.1</a>	415	No protein	Processed transcript	-	-	TSL:3

The strategy is based on the design of *Lepr-203* transcript,The transcription is shown below

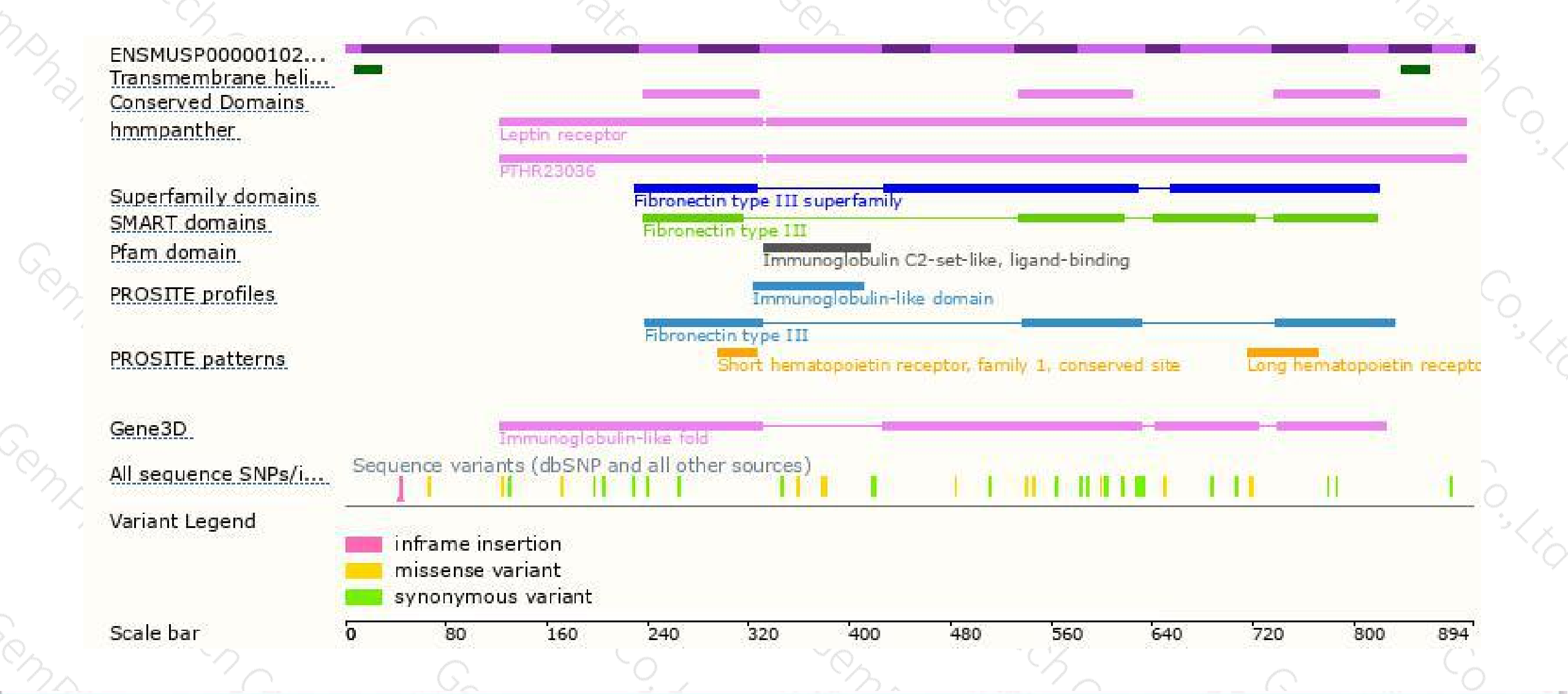


# Genomic location distribution

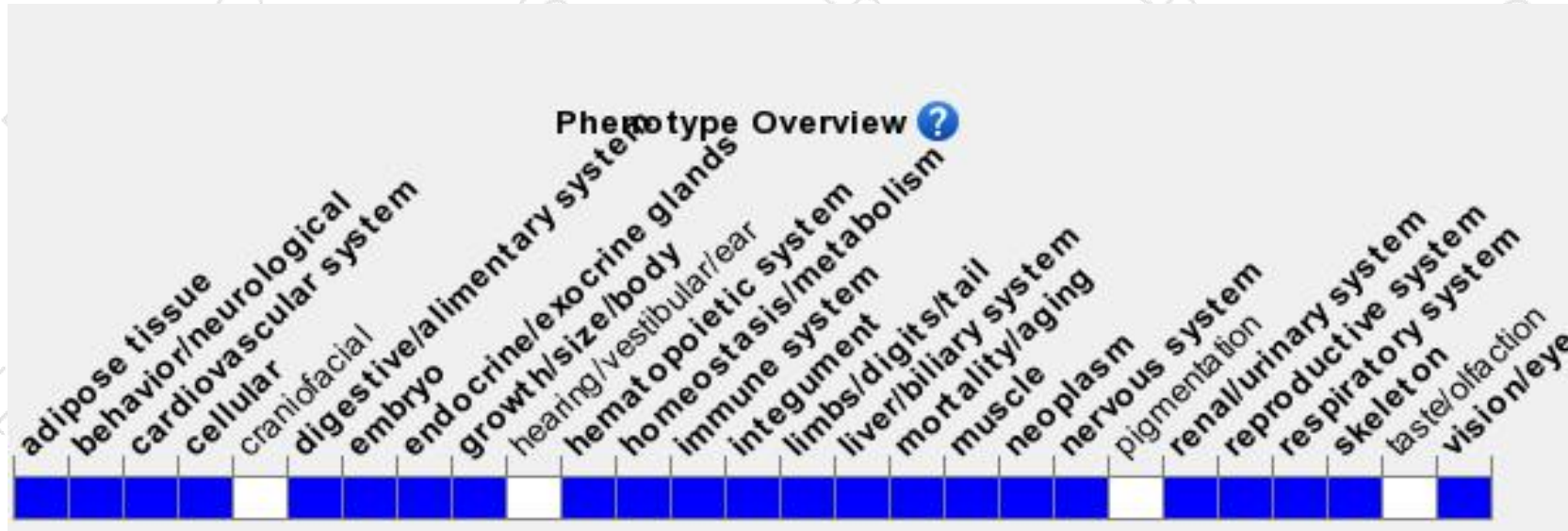




# Protein domain



# 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 mutants are hyperphagic, low-activity, poorly cold-adapted, sterile and have enhanced fat conversion. They are obese, hyperinsulinemic and, on certain strains, severely hyperglycemic.

Heterozygotes are normal but resistant to prolonged fasting.

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

Tel: 400-9660890

