Pomc Cas9-CKO *Strategy*

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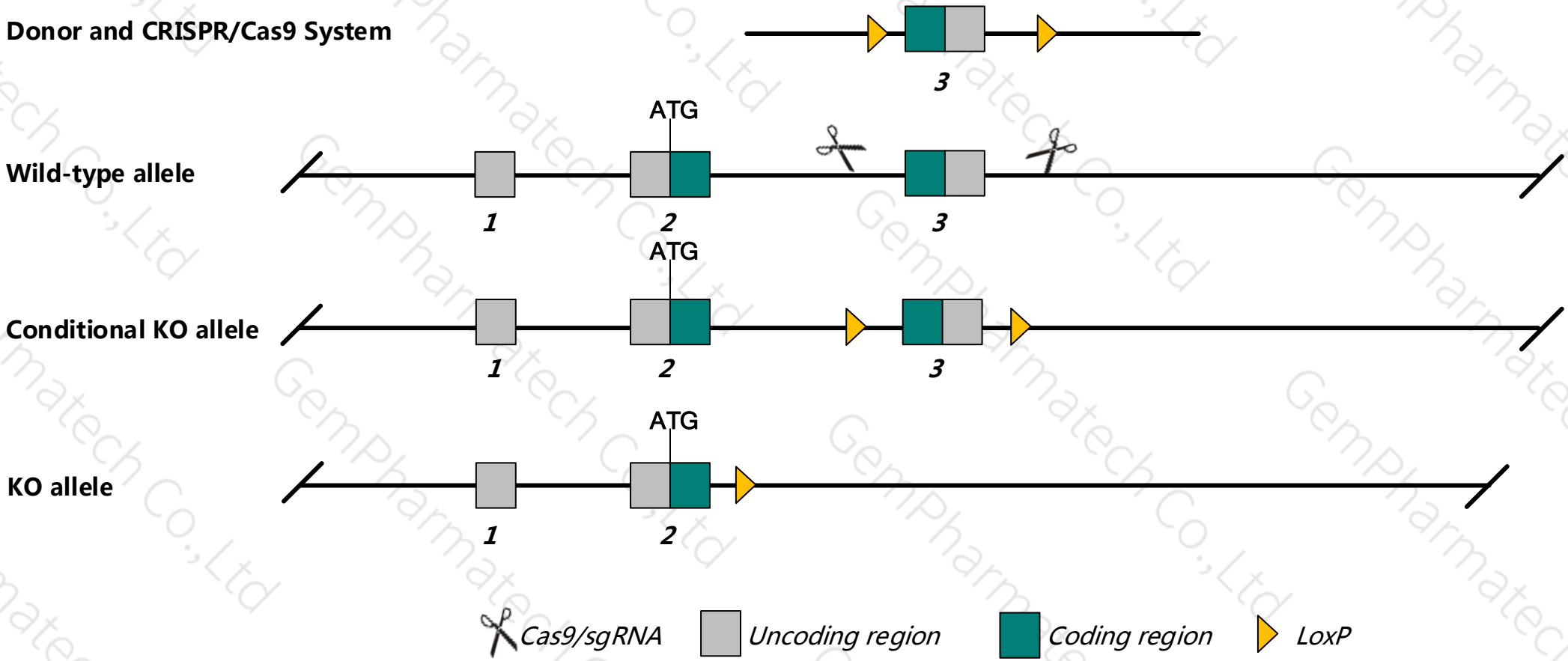
Project Overview

Project Name	*Pomc*
Project type	Cas9-CKO
Animal background	C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Pomc* gene has 5 transcripts, According to the structure of *Pomc* gene, exon3 of *Pomc-201* transcript is recommended as the knockout region.The region contains the most of coding seqence.Knock out the region,result in destruction of protein.

- This project uses CRISPR/Cas9 technology to modify *Pomc* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed, Cas9, sgRNA and donor 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.

- 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 and cell types.

Notice

- According to the existing MGI data，Homozygotes for a targeted null mutation are obese and exhibit abnormal hormone levels, abnormal pigmentation, increased food intake, and adiposity. Mice homozygous for another knock-out allele exhibit altered reward based behavior and immune response toLPS treatment.

- The *Pomc* 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）

集萃药康 GemPharmatech

Pomc pro-opiomelanocortin-alpha [*Mus musculus* (house mouse)]

Gene ID: 18976, updated on 23-Dec-2018

Summary

Official Symbol	Pomc provided by MGI
Official Full Name	pro-opiomelanocortin-alpha provided by MGI
Primary source	MGI:MGI:97742
See related	Ensembl:ENSMUSG00000020660
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BE; Npp; ACTH; Clip; Pomc1; Pomc-1; Beta-LPH; alphaMSH; beta-MSH; Gamma-LPH; alpha-MSH; gamma-MSH
Summary	This gene encodes a polypeptide hormone precursor that undergoes extensive, tissue-specific, post-translational processing. Processing yields several biologically active peptides, which are involved in diverse cellular functions, such as energy homeostasis, steroidogenesis, and increased melanin production in melanocytes. In mouse deficiency of this gene is associated with obesity, defects in adrenal development, and altered pigmentation. A pseudogene of this gene is located on chromosome 19. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Jun 2013]
Expression	Biased expression in testis adult (RPKM 4.3), CNS E14 (RPKM 2.4) and 11 other tissues See more
Orthologs	human all

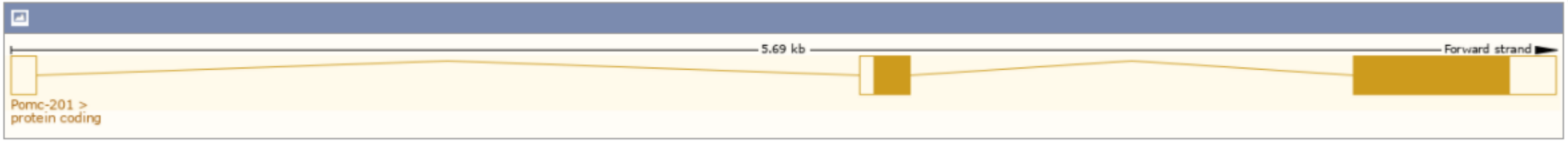
Transcript information （Ensembl）

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

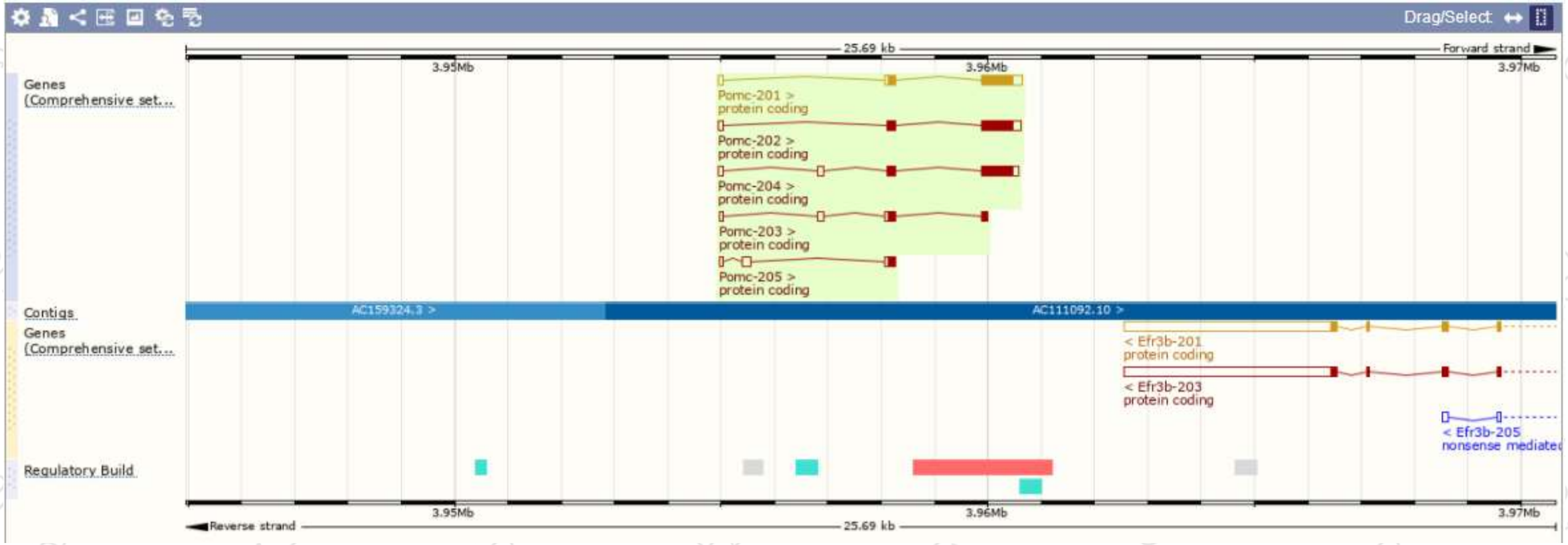
Show/hide columns (1 hidden) Filter

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags
Pomc-204	ENSMUST00000219543.1	1043	235aa	Protein coding	CCDS25785	P01193	NM_001278582 NM_001278583 NP_001265511 NP_001265512	TSL:1 GENCODE basic APPRIS P1
Pomc-201	ENSMUST00000020990.6	1031	235aa	Protein coding	CCDS25785	P01193	NM_001278581 NM_008895 NP_001265510 NP_032921	TSL:1 GENCODE basic APPRIS P1
Pomc-202	ENSMUST00000218089.1	977	235aa	Protein coding	CCDS25785	P01193	NM_001278584 NP_001265513	TSL:1 GENCODE basic APPRIS P1
Pomc-203	ENSMUST00000218169.1	480	79aa	Protein coding	-	A0A1W2P7R2	-	CDS 3' incomplete TSL:2
Pomc-205	ENSMUST00000220006.1	438	44aa	Protein coding	-	A0A1W2P724	-	CDS 3' incomplete TSL:3

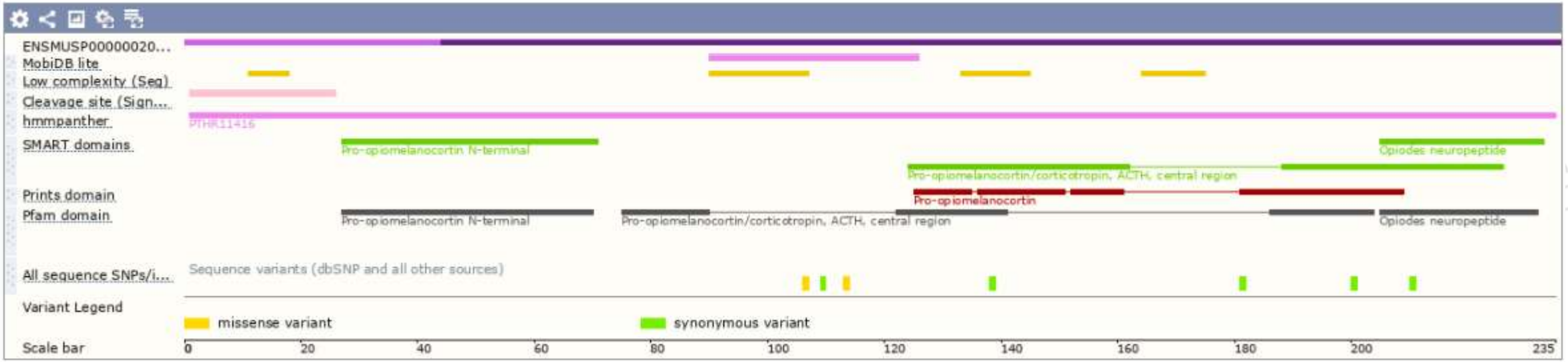
The strategy is based on the design of *Pomc-201* transcript,The transcription is shown below：



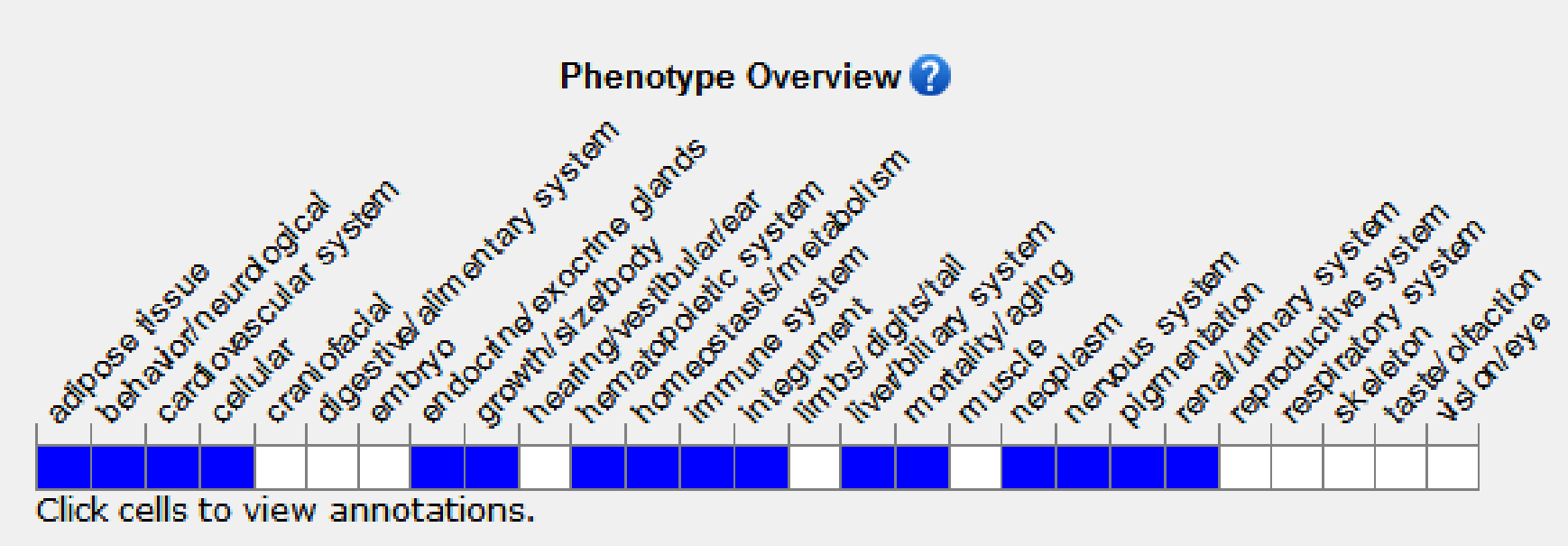
Genomic location distribution



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/) .

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