Il6 Cas9-CKO Strategy

Designer: Jing Jin

Reviewer: Yang Zeng

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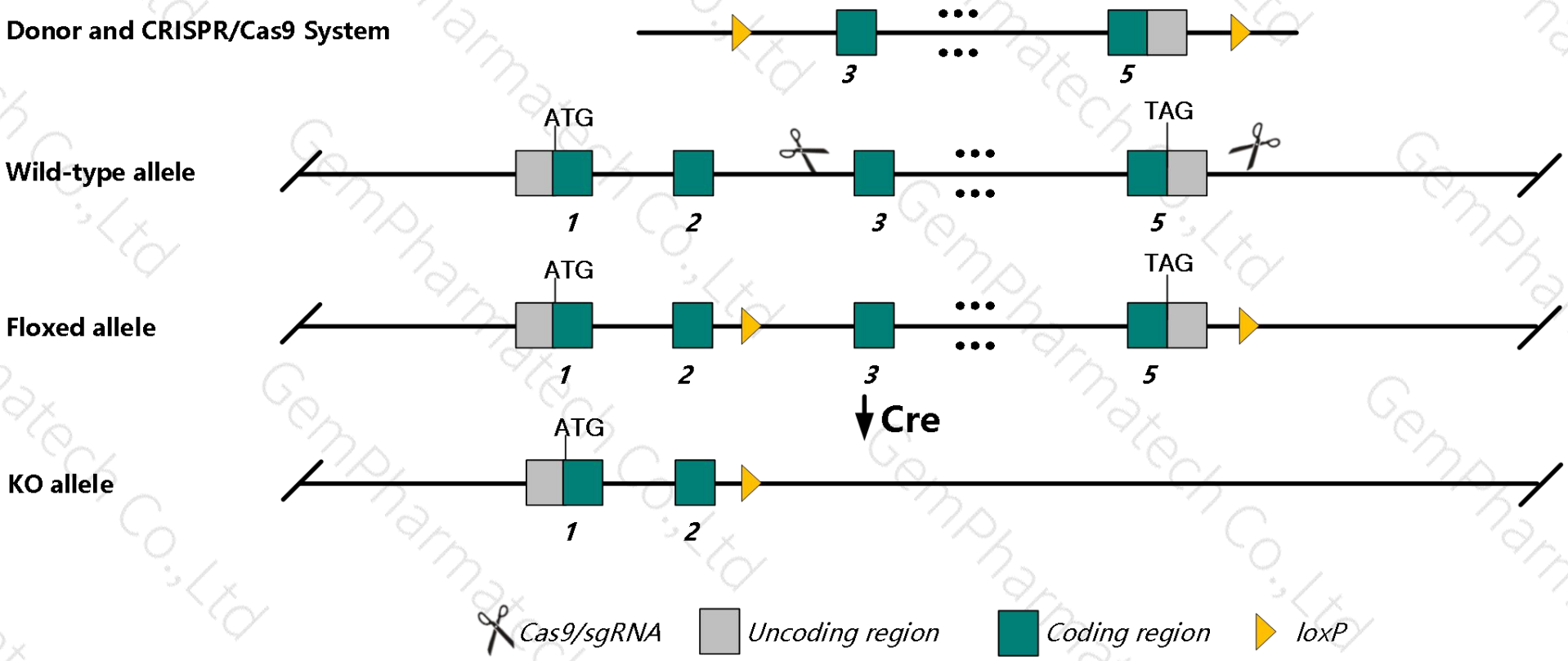
Project Overview

Project Name	***Il6***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Il6* gene. The schematic diagram is as follows:



Technical routes

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➢ The *Il6* gene has 4 transcripts. According to the structure of *Il6* gene, exon3-exon5 of *Il6-201* (ENSMUST00000026845.11) transcript is recommended as the knockout region.The region contains 432bp coding sequence and 3'UTR region. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Il6* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 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, Homozygous null mutants show impaired immune response to pathogens, decreased T cell numbers and resistance to plasma cell neoplasia. They are defective in wound healing and liver regeneration and show increased emotionality and high bone turnover rate.

- The *Il6* is close to 5'UTR region of the *A230098N10Rik* gene.Knockout the region may affect the function of the *A230098N10Rik* gene.

- The *Il6* gene is located on the Chr5. 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）

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Il6 interleukin 6 [*Mus musculus* (house mouse)]

Gene ID: 16193, updated on 13-Aug-2019

Summary

Official Symbol	Il6 provided by MGI
Official Full Name	interleukin 6 provided by MGI
Primary source	MGI:MGI:96559
See related	Ensembl:ENSMUSG00000025746
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	Il-6
Summary	This gene encodes a member of the interleukin family of cytokines that have important functions in immune response, hematopoiesis, inflammation and the acute phase response. The ectopic overexpression of the encoded protein in mice results in excessive plasma cells in circulation, leading to death. Mice lacking the encoded protein exhibit abnormalities in hepatic acute phase response, some immune mechanisms, bone resorption in response to estrogen, liver regeneration and wound healing. Alternative splicing results in multiple transcript variants encoding different isoforms. [provided by RefSeq, Sep 2015]
Expression	Low expression observed in reference dataset See more
Orthologs	human all

Transcript information（Ensembl）

The gene has 4 transcripts,and all transcripts are shown below:

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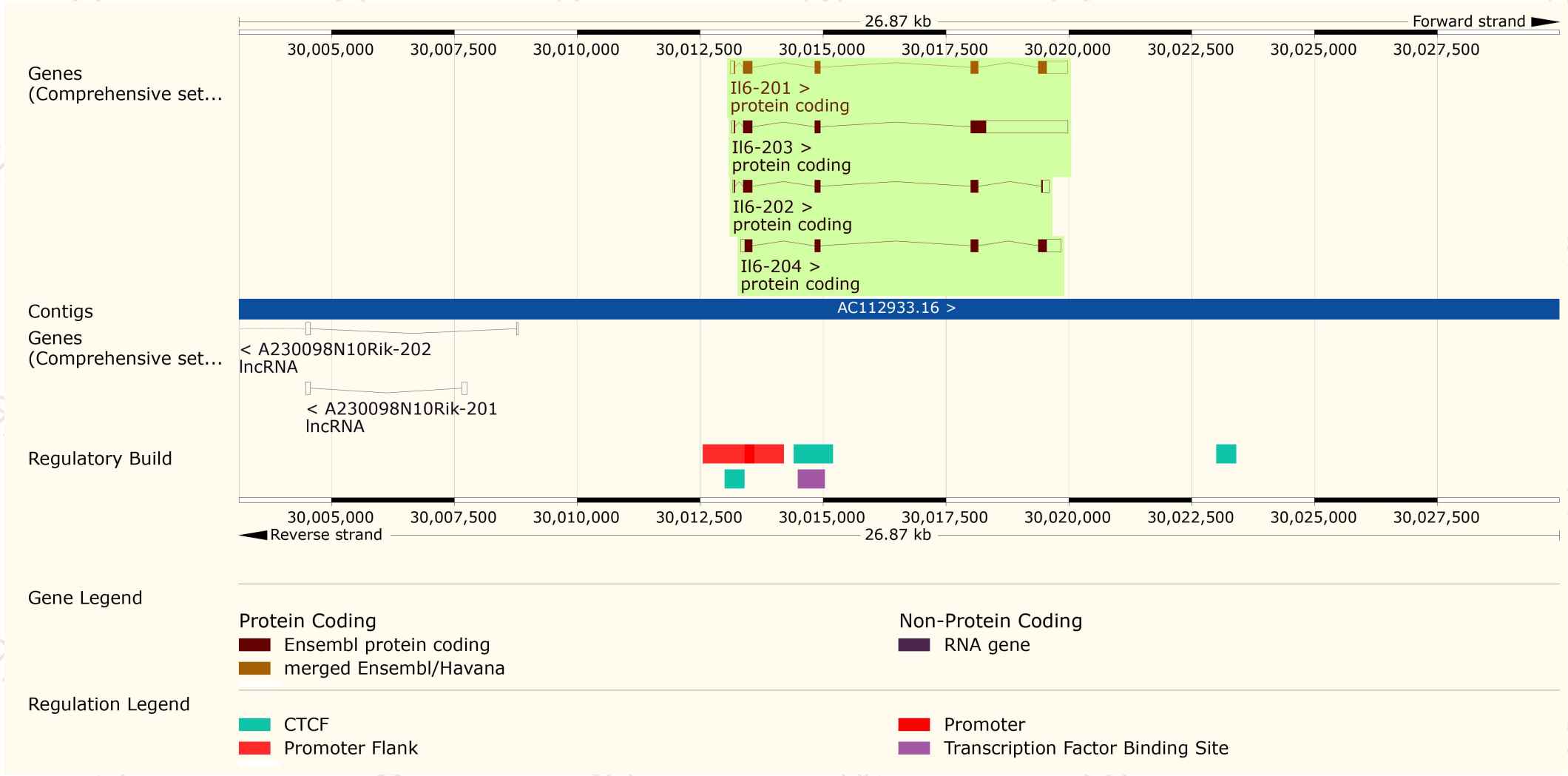
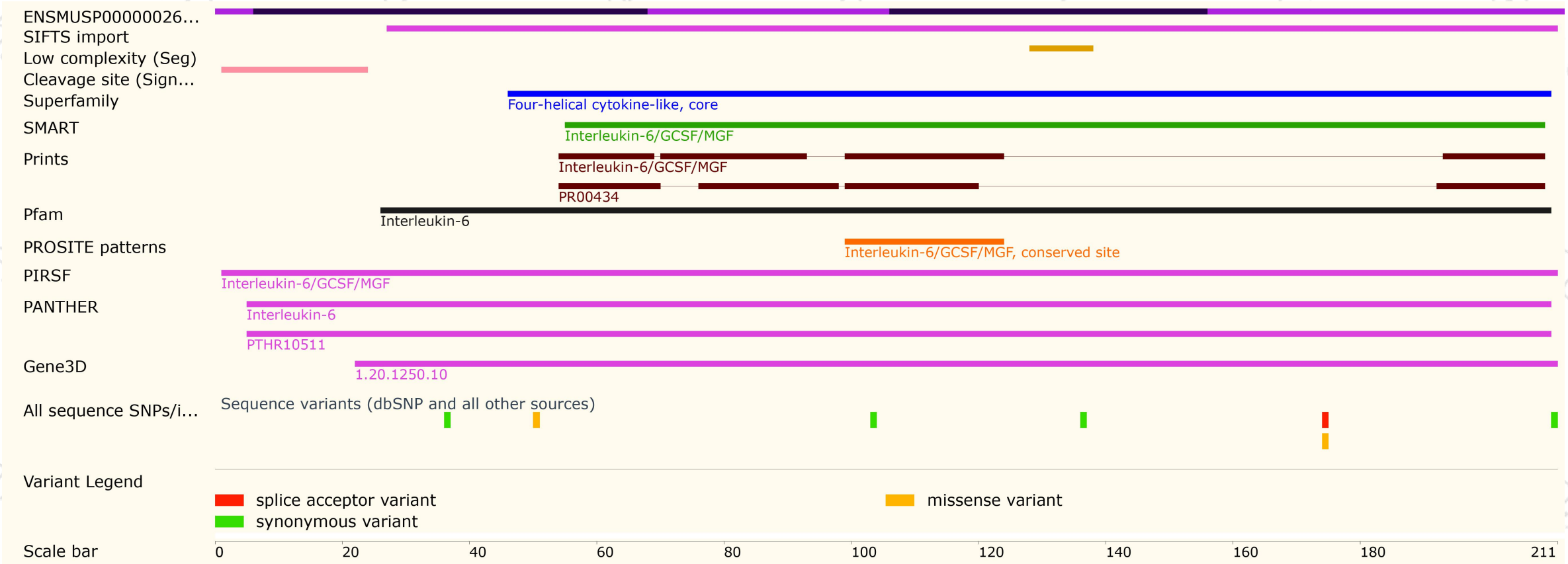
Name ▲	Transcript ID	bp	Protein	Translation ID	Biotype	CCDS	UniProt	Flags
Il6-201	ENSMUST00000026845.11	1141	211aa	ENSMUSP00000026845.7	Protein coding	CCDS19153	A2RTD1 P08505	TSL:1 GENCODE basic APPRIS P2
Il6-202	ENSMUST00000195978.4	651	165aa	ENSMUSP00000143544.1	Protein coding	CCDS84856	A0A0G2JGF4	TSL:1 GENCODE basic
Il6-203	ENSMUST00000199183.4	2343	208aa	ENSMUSP00000143293.1	Protein coding	-	A0A0G2JFT1	TSL:2 GENCODE basic
Il6-204	ENSMUST00000199765.1	963	194aa	ENSMUSP00000143157.1	Protein coding	-	A0A0G2JFG1	TSL:2 GENCODE basic APPRIS ALT2

The strategy is based on the design of *Il6-201* transcript,The transcription is shown below

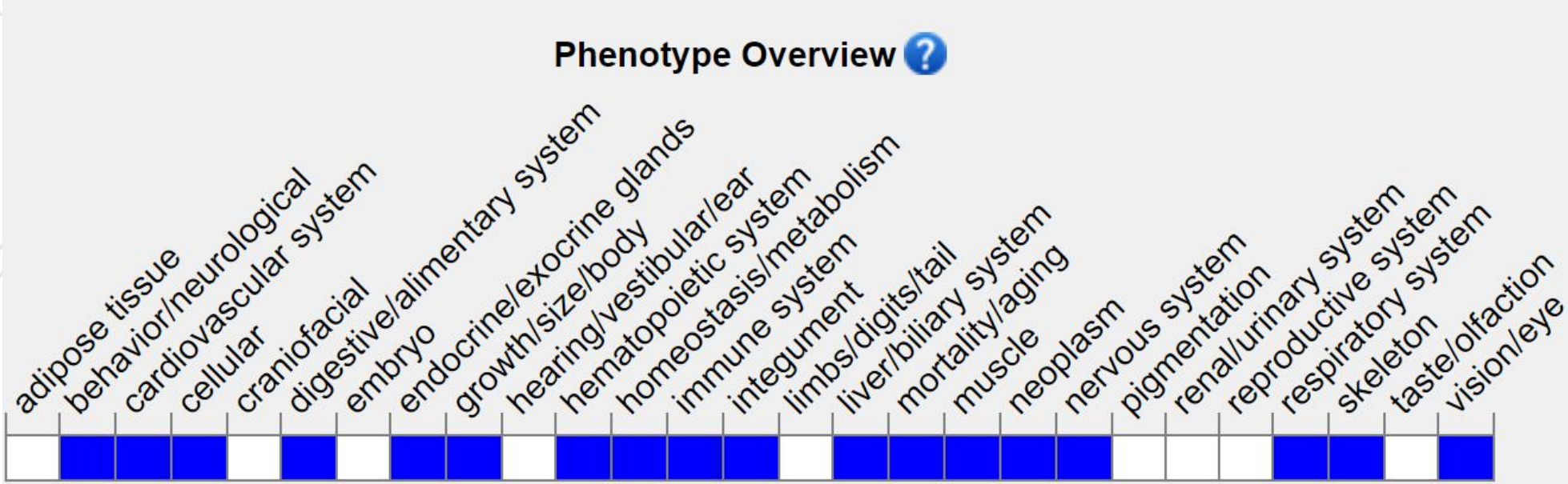
Genomic location distribution



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/).

According to the existing MGI data, Homozygous null mutants show impaired immune response to pathogens, decreased T cell numbers and resistance to plasma cell neoplasia. They are defective in wound healing and liver regeneration and show increased emotionality and high bone turnover rate.

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

Tel: 025-5864 1534

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GemPharmatech Co.,Ltd