Cd226 Cas9-KO Strategy

Designer:Xueting Zhang

Project Overview

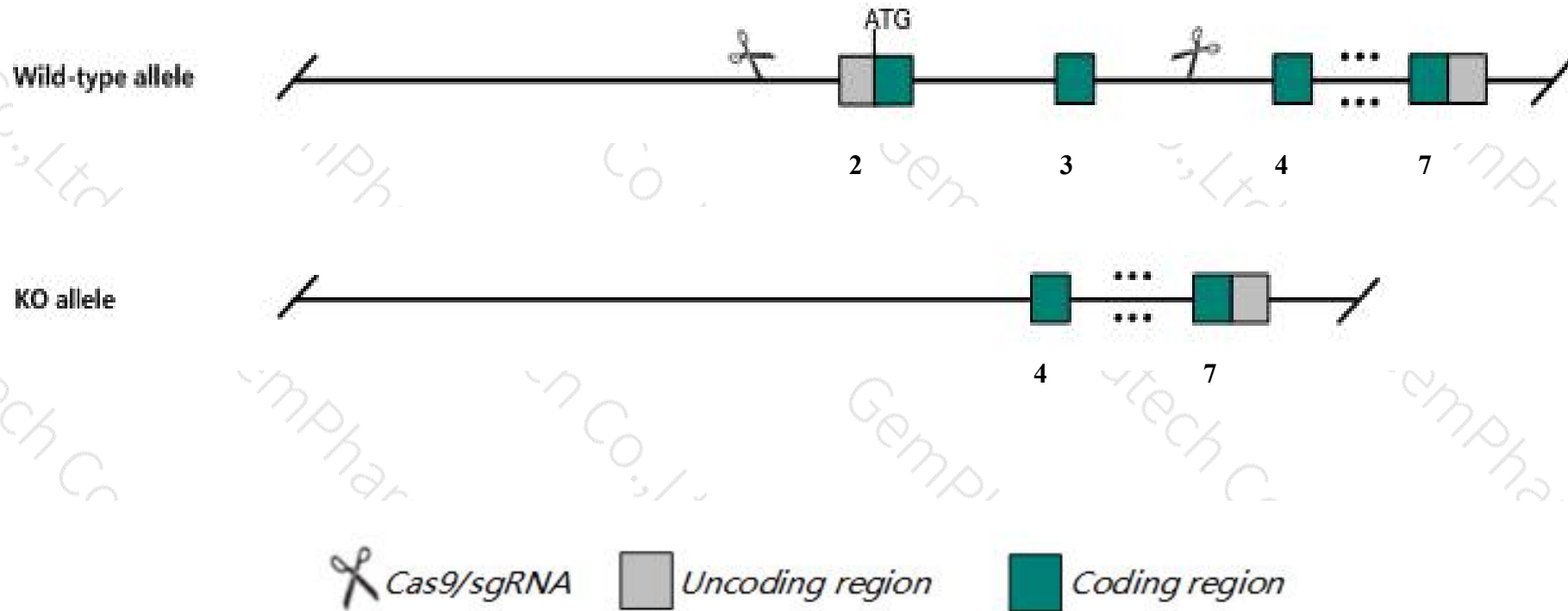|	
Project Name	*Cd226*
Project type	**Cas9-KO**
Strain background	**C57BL/6J**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cd226* gene. The schematic diagram is as follows:

Technical routes

- The *Cd226* gene has 10 transcripts. According to the structure of *Cd226* gene, exon3-exon4 of *Cd226-201* (ENSMUST00000037142.12) transcript is recommended as the knockout region.The region contains start codon ATG. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Cd226* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

Notice

➢ According to the existing MGI data,Mice homozygous for a knock-out allele exhibit impaired NK cell cytolysis and increased incidence of tumor formation and mortality.

➢ The *Cd226* gene is located on the Chr18. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

Cd226 CD226 antigen [Mus musculus (house mouse)]
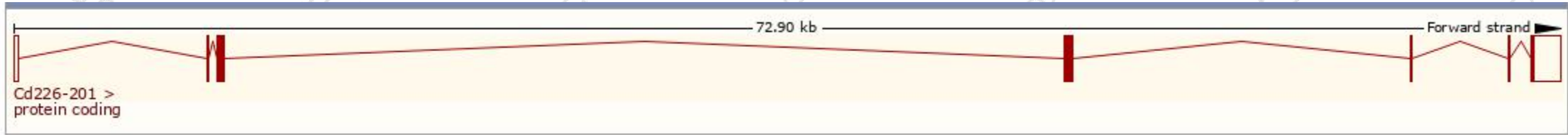
Gene ID: 225825, updated on 19-Mar-2019

▲Summary

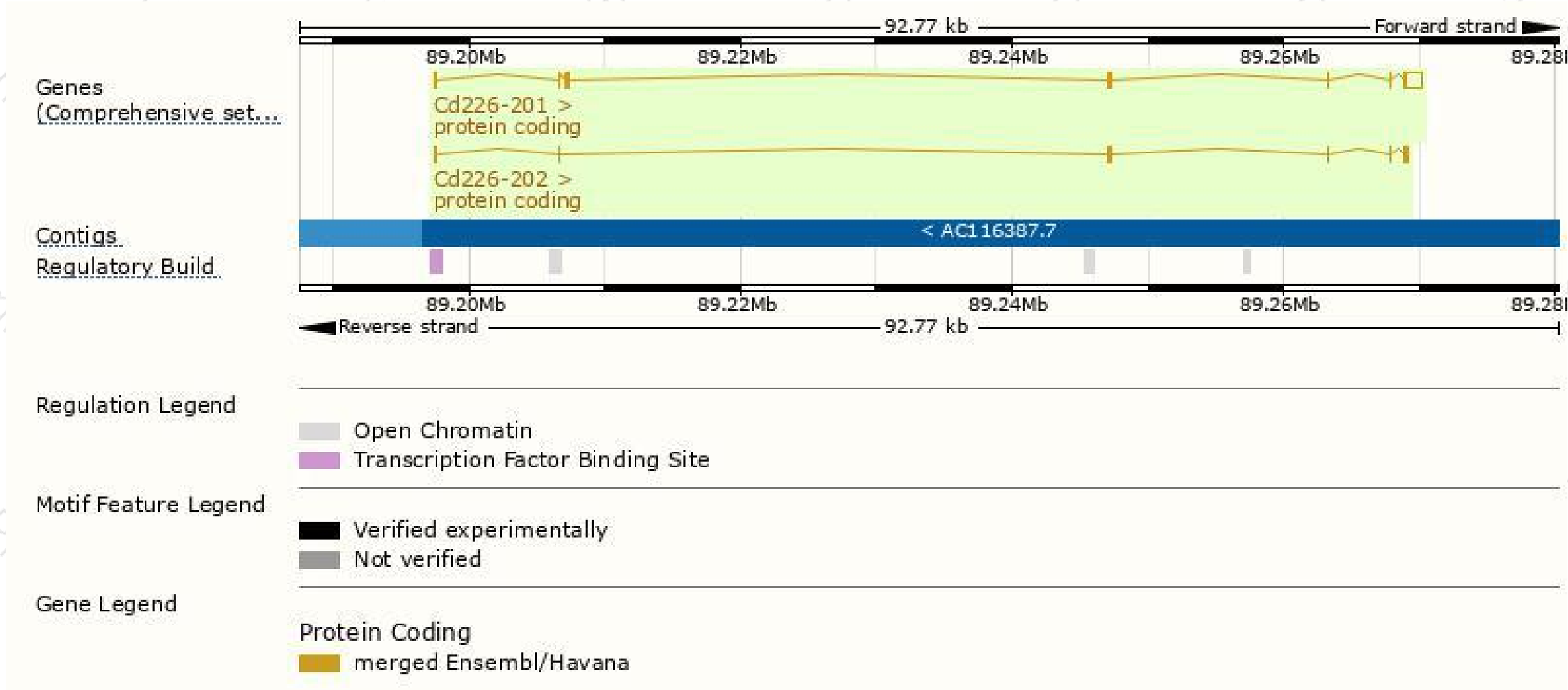|	
Official Symbol	Cd226 provided by MGI
Official Full Name	CD226 antigen provided by MGI
Primary source	MGI:MGI:3039602
See related	Ensembl:ENSMUSG00000034028
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BC051526, DNAM-1, DNAM1, Pta1, TLiSA1
Expression	Low expression observed in reference datasetSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 10 transcripts,all transcripts are shown below:

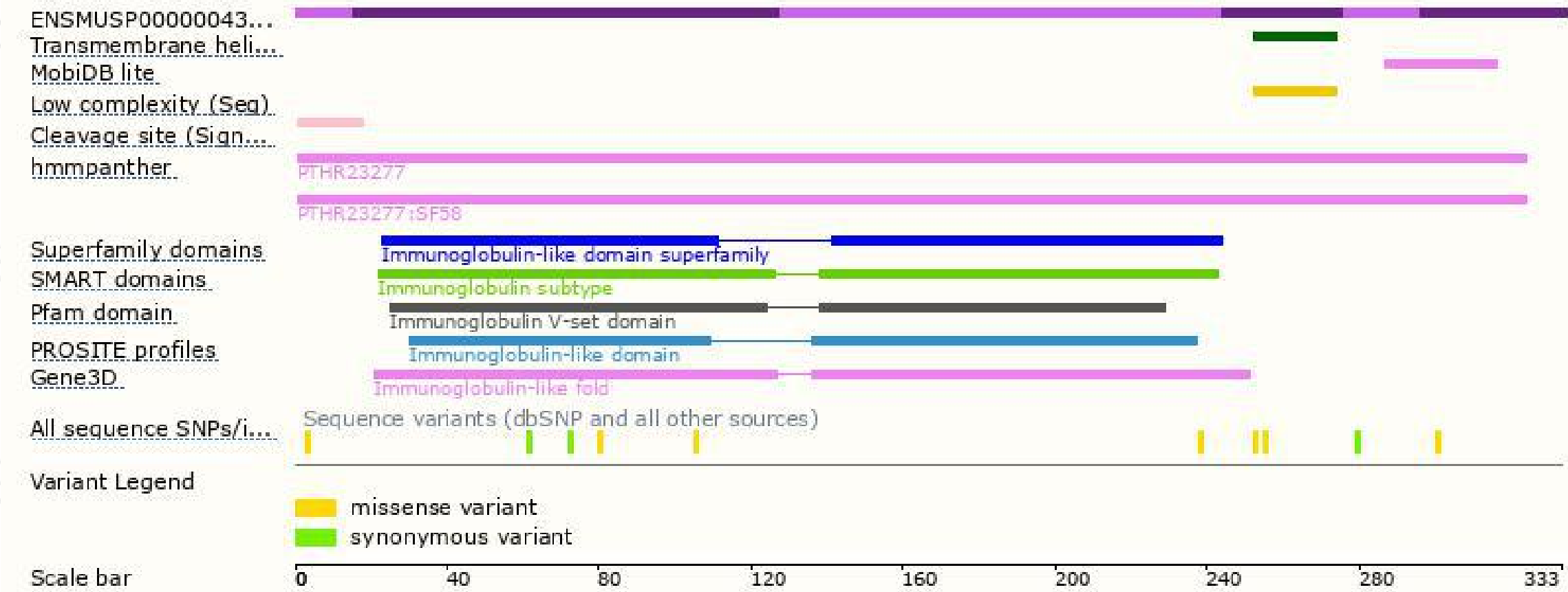| Name	Transcript ID ▲	bp	Protein	Biotype	CCDS	UniProt	Flags
Cd226-201	ENSMUST00000037142.12	2487	333aa	▌Protein coding	CCDS29392	Q5DW69 Q8K4F0	TSL:1 GENCODE basic APPRIS P1
Cd226-202	ENSMUST00000097496.3	1239	220aa	▌Protein coding	CCDS29391	Q8K4E3	TSL:1 GENCODE basic
Cd226-203	ENSMUST00000235651.1	2456	154aa	▌Nonsense mediated decay	-	-	-
Cd226-204	ENSMUST00000236450.1	592	No protein	▌Retained intron	-	-	-
Cd226-205	ENSMUST00000236452.1	742	129aa	▌Protein coding	-	Q8K4E2	GENCODE basic
Cd226-206	ENSMUST00000236644.1	446	38aa	▌Protein coding	-	-	CDS 3' incomplete
Cd226-207	ENSMUST00000236828.1	4494	333aa	▌Protein coding	CCDS29392	Q5DW69	GENCODE basic APPRIS P1
Cd226-208	ENSMUST00000236835.1	451	119aa	▌Protein coding	-	-	CDS 3' incomplete
Cd226-209	ENSMUST00000237110.1	408	24aa	▌Protein coding	-	-	CDS 3' incomplete
Cd226-210	ENSMUST00000237979.1	1364	187aa	▌Nonsense mediated decay	-	Q8K4E4	-

The strategy is based on the design of *Cd226-201* transcript,The transcription is shown below
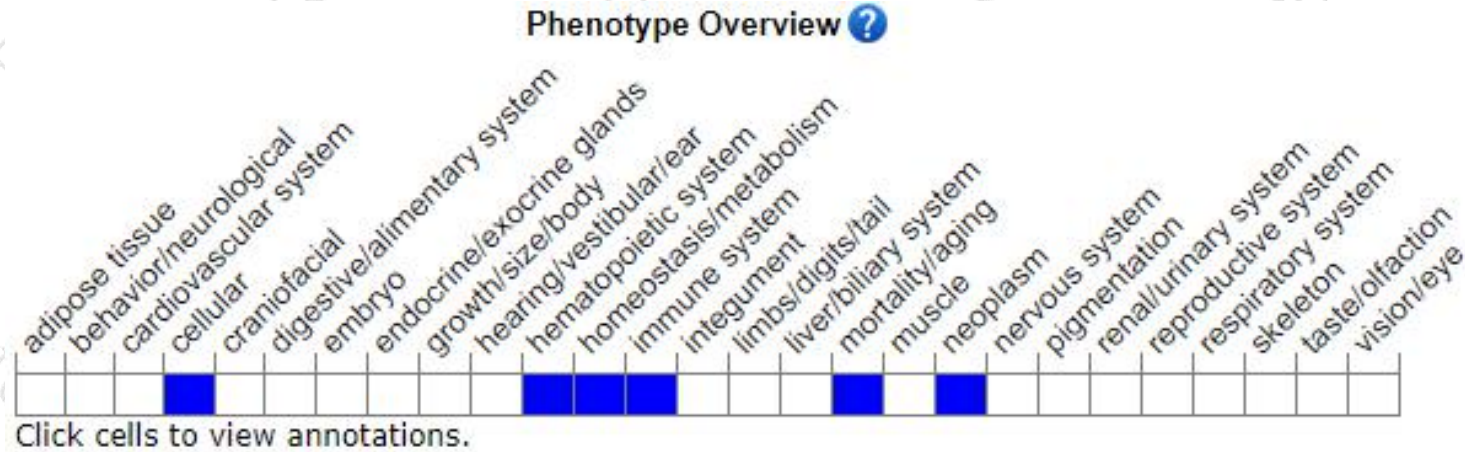
Genomic location distribution

Protein domain

ENSMUSP00000043...	
Transmembrane heli...	
MobiDB lite	
Low complexity (Seg)	
Cleavage site (Sign...	
hmmpanther	PTHR23277
	PTHR23277:SF58
Superfamily domains	Immunoglobulin-like domain superfamily
SMART domains	Immunoglobulin subtype
Pfam domain	Immunoglobulin V-set domain
PROSITE profiles	Immunoglobulin-like domain
Gene3D	Immunoglobulin-like fold
All sequence SNPs/i...	Sequence variants (dbSNP and all other sources)

Variant Legend

- missense variant
- synonymous variant

Scale bar: 0   40   80   120   160   200   240   280   333

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,Mice homozygous for a knock-out allele exhibit impaired NK cell cytolysis and increased incidence of tumor formation and mortality.

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

集萃药康生物科技
GemPharmatech Co.,Ltd