Eef2-T57M cas9-ki(PM) Mouse Model Strategy

-CRISPR/Cas9 technology

Designer: Xiaojing Li

Reviewer: JiaYu

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Project Overview

Project Name	**Eef2-T57M**
Project type	**cas9-ki(PM)**
Strain background	**C57BL/6JGpt**

A comparison of the aa homology of human and mouse *Eef2* gene

The 57th amino acid(T) of human *EEF2* gene corresponds to the 57th amino acid(T) of mouse *Eef2* gene after comparing homology of mouse *Eef2* gene and human *EEF2* gene.

Technical Description

- The mouse *Eef2* gene has 3 transcripts. The human *EEF2* gene has 7 transcripts.

- According to the structure of *Eef2* gene and requirements of customer, the 57th amino acid(T) of human *EEF2*(NM_001961.4) gene corresponds to the 57th amino acid(T) of mouse *Eef2* gene after comparing homology of mouse *Eef2* gene and human *EEF2* gene. This project produced *Eef2*-T57A point mutation on exon2 of the transcript of *Eef2*-201(ENSMUST00000047864.11, NM_007907.2). The 57th amino acids will be mutated from T to M, and the corresponding codon will be mutated to ATG by the ACT.

- The mouse *Eef2*-201 transcript contains 14 exons. The translation initiation site ATG is located at exon1, and the translation termination site TAG is located at exon14, encoding 858aa.

- In this project, *Eef2* gene will be modified by CRISPR/Cas9 technology. The brief process is as follows: In vitro, sgRNA and donor vectors were constructed. Cas9, sgRNA and donor were injected into the fertilized eggs of C57BL/6JGpt mice for homologous recombination, and obtained positive F0 mice identified by PCR and sequencing analysis. The stable inheritable positive F1 mice model was obtained by mating F0 mice with C57BL/6JGpt mice.

Mutation Site

Before mutation

After mutation

The yellow region is exon2 of *Eef2-201,* the red region represents the mutation site.
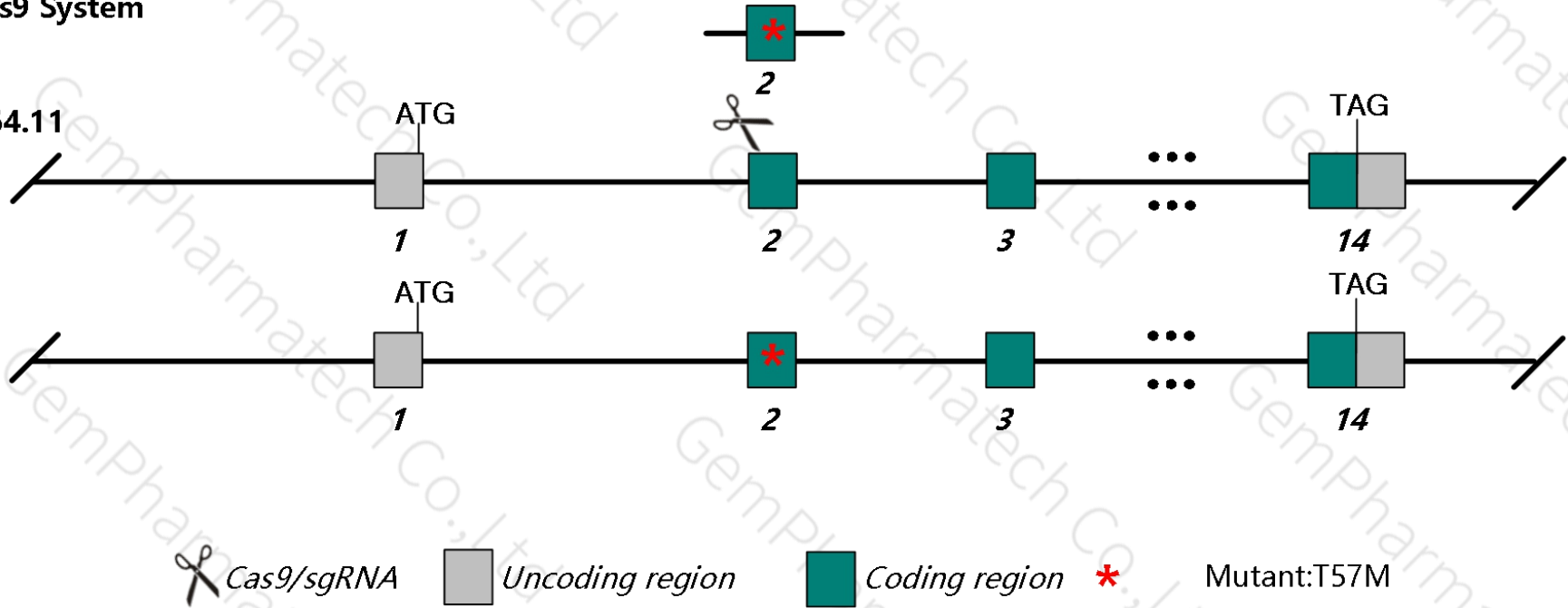
Strategy

This model uses CRISPR/Cas9 technology to edit the *Eef2* gene and the schematic diagram is as follow:

Donor and CRISPR/Cas9 System

ENSMUST00000047864.11

Wild-type allele

Targeted allele

Cas9/sgRNA   Uncoding region   Coding region   * Mutant:T57M

Notice

> According to the data of MGI, mice homozygous for a mutation removing the diphthamide modification display partial neonatal lethality, fetal growth retardation and abnormal cell physiology.Most of the homozygous mice with *Eef2*-G717R point mutation died shortly after birth.

> One or Two synonymous mutations of amino acids will be introduced on exon2 of *Eef2*.

> The mutation site is about 5.2kb away from the 5-terminal of *Dapk3* gene, which may affect its 5-terminal regulation.

> Mouse *Eef2* gene is located on Chr10. Please take the loci in consideration when breeding this mutation mice with other gene modified strains, if the other gene is also on Chr10, it may be extremely hard to get double gene positive homozygotes.

> The scheme is designed according to the genetic information in the existing database. Due to the complex process of gene transcription and translation, it cannot be predicted completely at the present technology level.
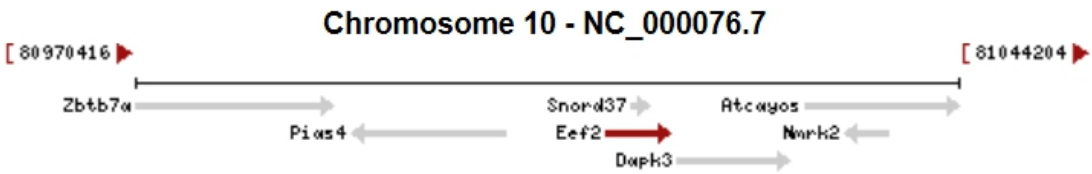
Gene name and location（NCBI）

Eef2 eukaryotic translation elongation factor 2 [*Mus musculus* (house mouse)]

⬇ **Download Datasets**

Gene ID: 13629, updated on 3-Jan-2021

▲ **Summary** ⏏ ?

Official Symbol	Eef2 provided by MGI
Official Full Name	eukaryotic translation elongation factor 2 provided by MGI
Primary source	MGI:MGI:95288
See related	Ensembl:ENSMUSG00000034994
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	Ef-2
Expression	Ubiquitous expression in ovary adult (RPKM 1248.0), colon adult (RPKM 810.0) and 28 other tissues See more
Orthologs	human all

Chromosome 10 - NC_000076.7

[80970416 ▶   [81044204 ▶

Zbtb7a   Snord37   Atcayos

Pias4   Eef2 ▬▶   Nmrk2

Dapk3

Transcript information （Ensembl）

The gene has 3 transcripts, and all transcripts are shown below:

| Name ▲	Transcript ID	bp	Protein	Biotype	CCDS	UniProt Match	Flags		
Eef2-201	ENSMUST00000047864.11	3089	858aa	Protein coding	CCDS35993 ⊡	P58252 ⊡	TSL:1	GENCODE basic	APPRIS P1
Eef2-202	ENSMUST00000219497.2	651	No protein	Retained intron	-	-		TSL:3	
Eef2-203	ENSMUST00000219943.2	519	No protein	Retained intron	-	-		TSL:1	

The strategy is based on the design of *Eef2*-201 transcript, the transcription is shown below:
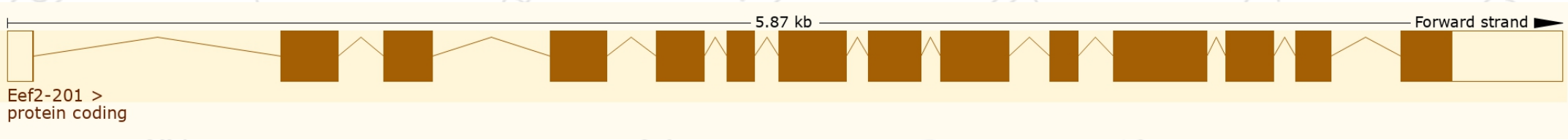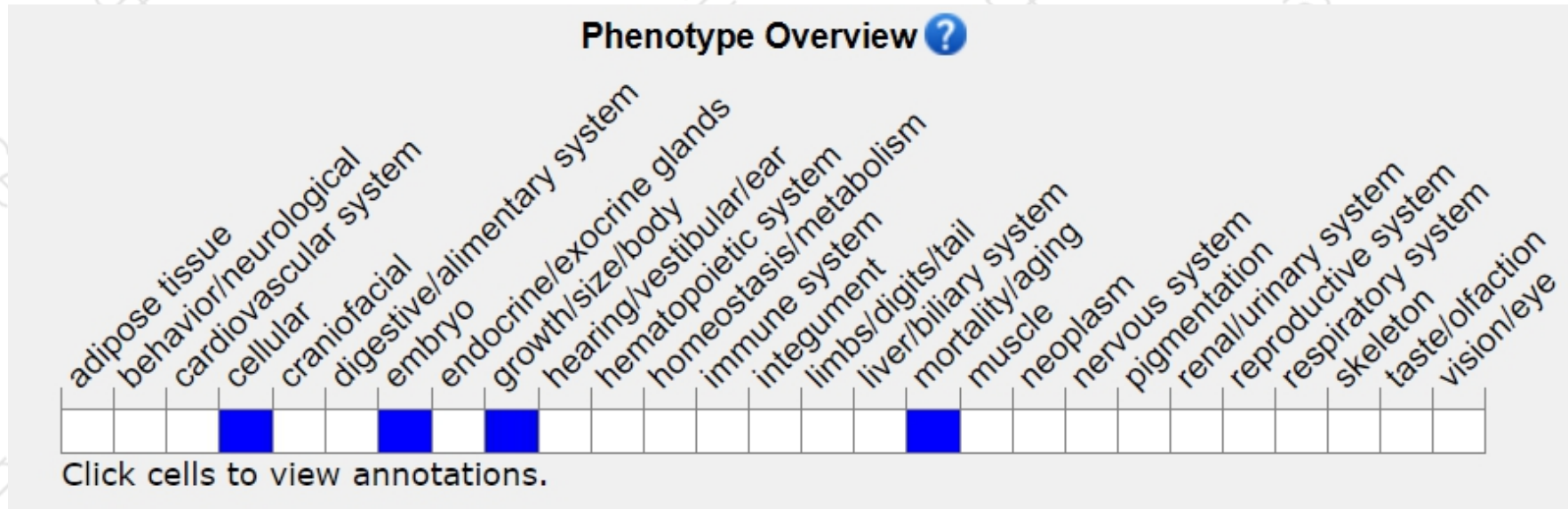
Genomic location distribution
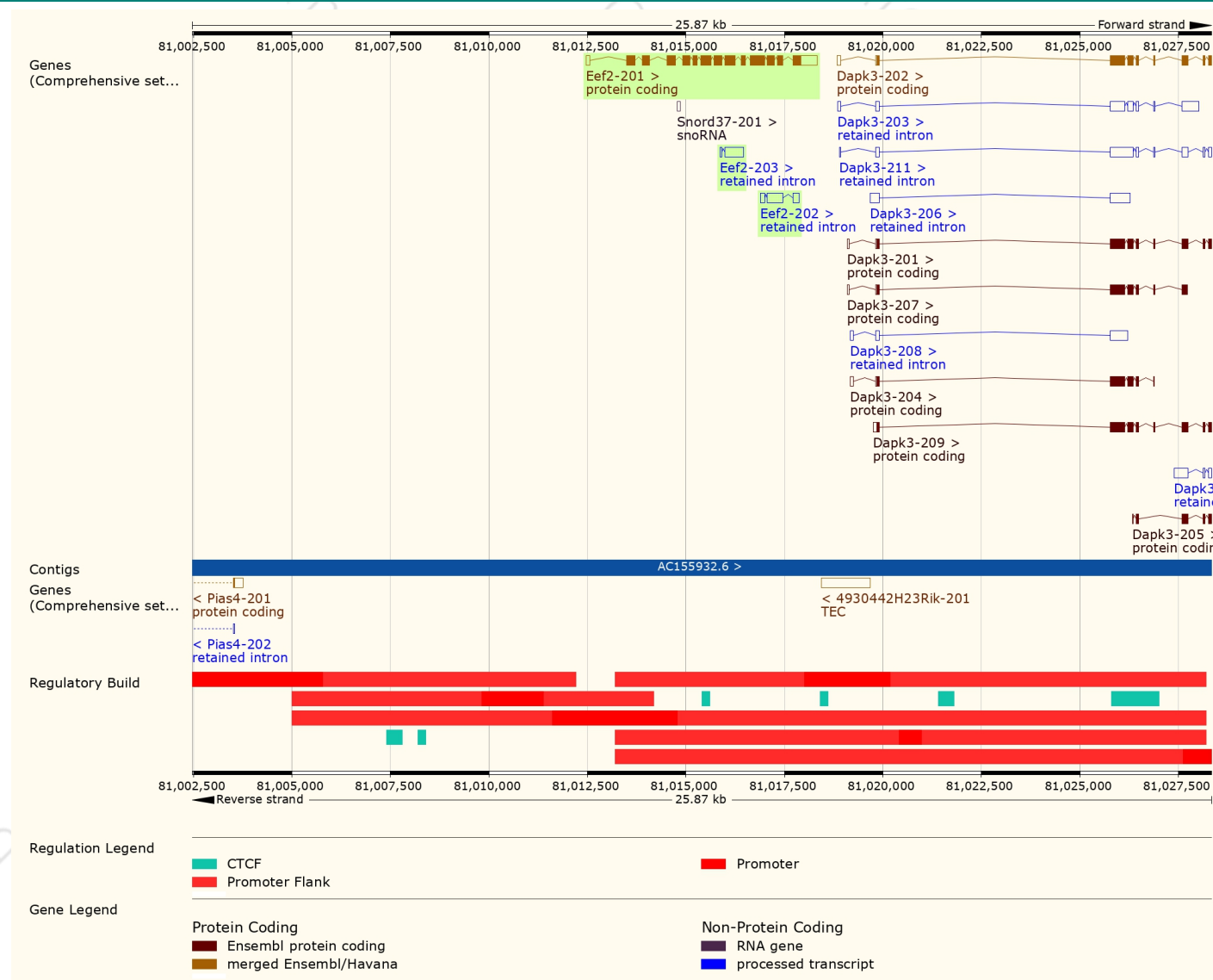
Protein domain
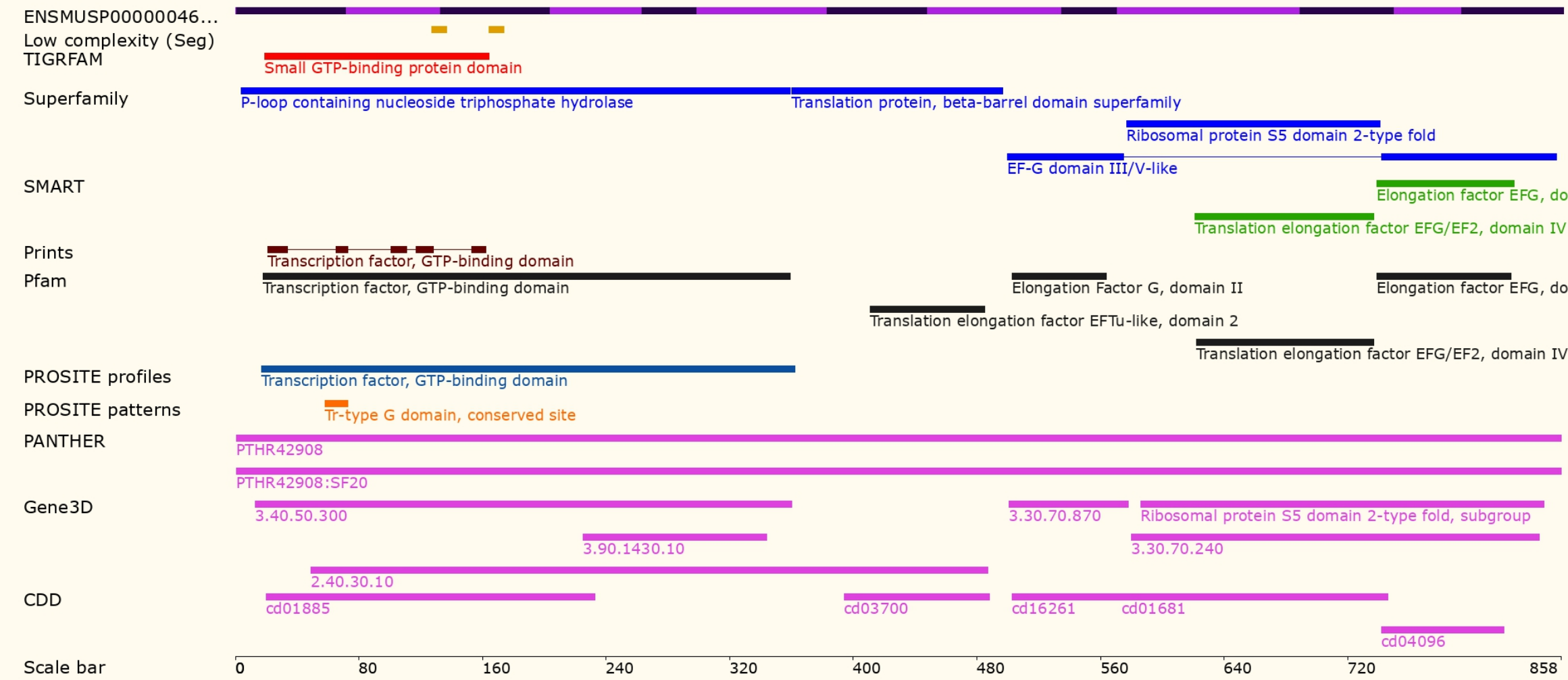
Mouse phenotype description(MGI)

URL link is as follows:

http://www.informatics.jax.org/marker/MGI:95288

Mice homozygous for a mutation removing the diphthamide modification display partial neonatal lethality, fetal growth retardation and abnormal cell physiology.

If you have any questions, please feel free to contact us.
Tel: 025-5864 1534

集萃药康生物科技
GemPharmatech Co.,Ltd