

***Tut4* Cas9-KO Strategy**

Designer: Huimin Su

Project Overview

Project Name

Tut4

Project type

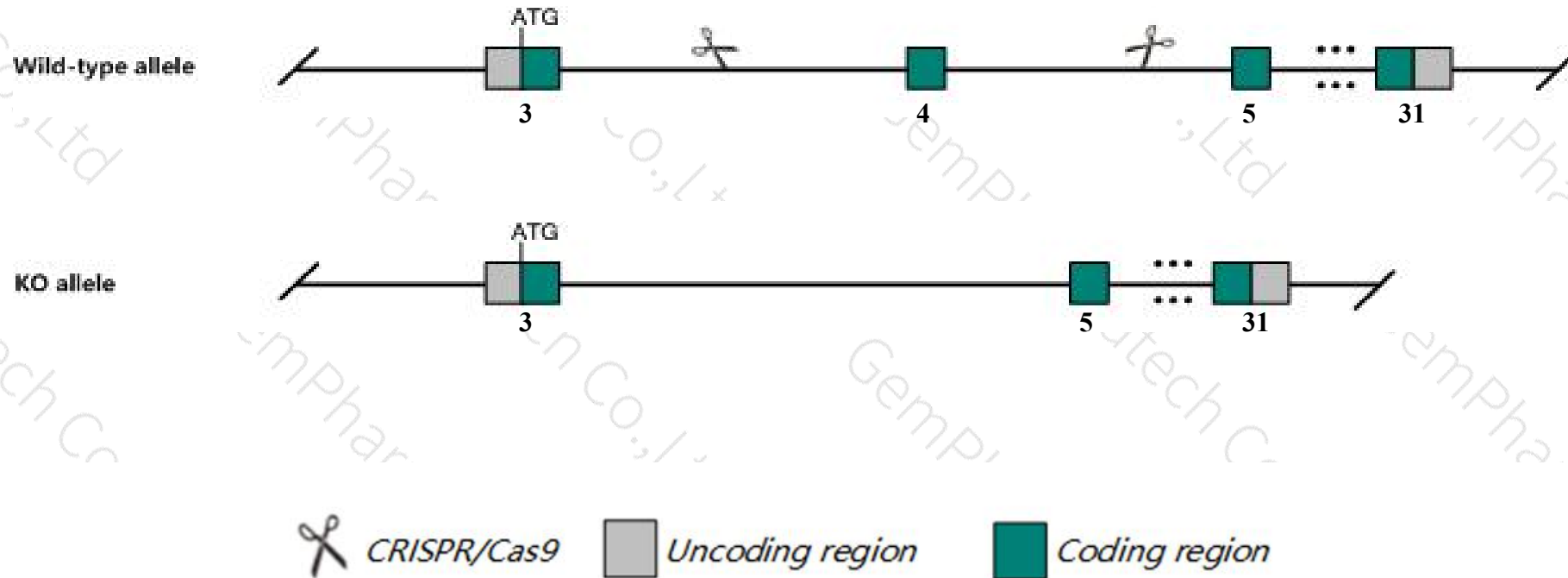
Cas9-KO

Strain background

C57BL/6JGpt

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Tut4* gene. The schematic diagram is as follows:



- The *Tut4* gene has 11 transcripts. According to the structure of *Tut4* gene, exon4 of *Tut4-201* (ENSMUST00000043368.11) transcript is recommended as the knockout region. The region contains 164bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Tut4* gene. The brief process is as follows: gRNA was transcribed in vitro. Cas9 and gRNA 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.

- According to the existing MGI data, Mice homozygous for a gene trap allele exhibit partial postnatal lethality associated with postnatal growth retardation and reduced circulating insulin-like growth factor I levels.
- The distance between exon4 of *Tut4-201* and *Gm12739* is about 2.0kb, and the regulation of *Gm12739* may be affected.
- The *Tut4* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Tut4 terminal uridylyl transferase 4 [Mus musculus (house mouse)]

Gene ID: 230594, updated on 31-Jan-2019

Summary



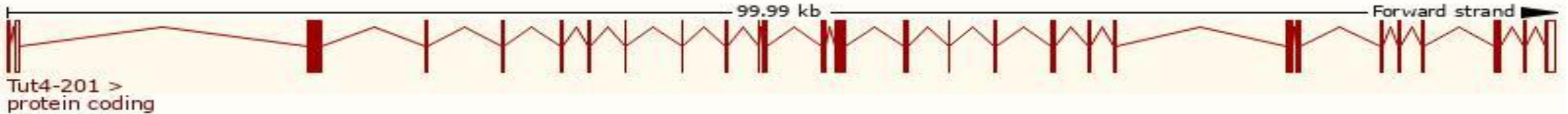
Official Symbol	Tut4 provided by MGI
Official Full Name	terminal uridylyl transferase 4 provided by MGI
Primary source	MGI:MGI:2445126
See related	Ensembl:ENSMUSG000000034610
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	6030404K05Rik, 9230115F04Rik, PPAPD3, Tent3a, Zcchc11, mKIAA0191
Expression	Ubiquitous expression in CNS E11.5 (RPKM 17.3), limb E14.5 (RPKM 15.9) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

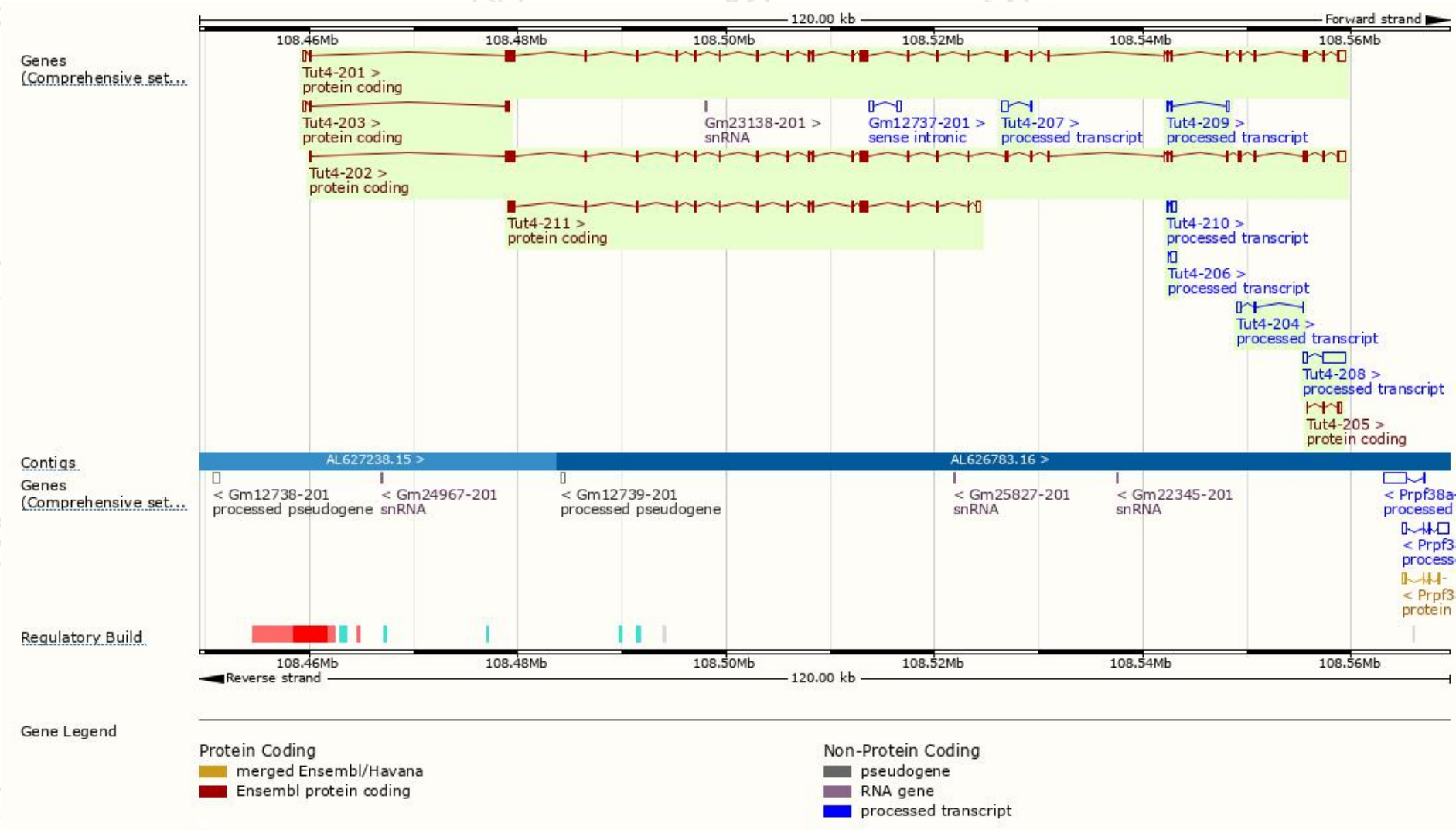
The gene has 11 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tut4-201	ENSMUST00000043368.11	6054	1644aa	Protein coding	CCDS18451	B2RX14	TSL:5 GENCODE basic APPRIS P2
Tut4-202	ENSMUST00000097925.8	5865	1648aa	Protein coding	-	A2A8R7	TSL:5 GENCODE basic APPRIS ALT2
Tut4-211	ENSMUST00000155068.1	3283	1006aa	Protein coding	-	A0A0A0MQJ6	CDS 5' incomplete TSL:1
Tut4-203	ENSMUST00000106673.7	679	63aa	Protein coding	-	A2A9H0	CDS 3' incomplete TSL:3
Tut4-205	ENSMUST00000128042.1	587	105aa	Protein coding	-	A2A8R1	CDS 5' incomplete TSL:2
Tut4-208	ENSMUST00000145590.1	2472	No protein	Processed transcript	-	-	TSL:1
Tut4-207	ENSMUST00000138809.1	636	No protein	Processed transcript	-	-	TSL:3
Tut4-206	ENSMUST00000138088.1	591	No protein	Processed transcript	-	-	TSL:5
Tut4-210	ENSMUST00000153916.1	567	No protein	Processed transcript	-	-	TSL:3
Tut4-209	ENSMUST00000151349.7	541	No protein	Processed transcript	-	-	TSL:2
Tut4-204	ENSMUST00000127602.1	484	No protein	Processed transcript	-	-	TSL:2

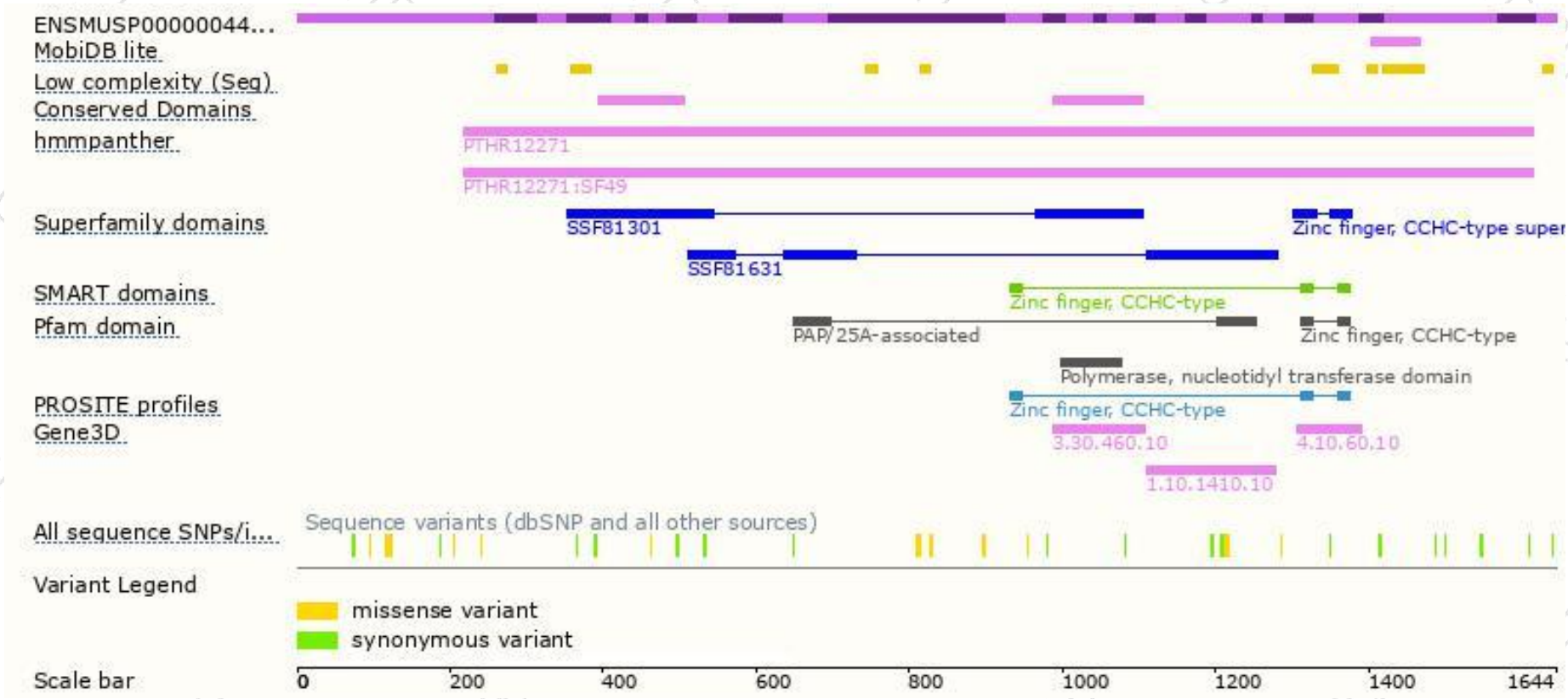
The strategy is based on the design of *Tut4-201* transcript,The transcription is shown below



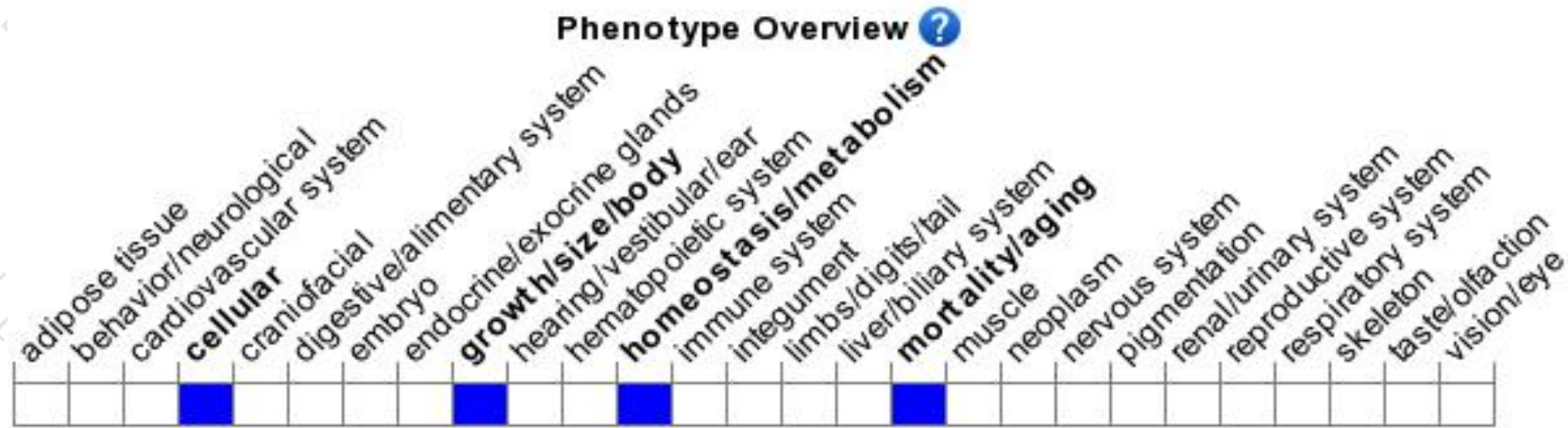
Genomic location distribution



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, Mice homozygous for a gene trap allele exhibit partial postnatal lethality associated with postnatal growth retardation and reduced circulating insulin-like growth factor I levels.

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

Tel: 400-9660890

