

Tut7 Cas9-CKO Strategy

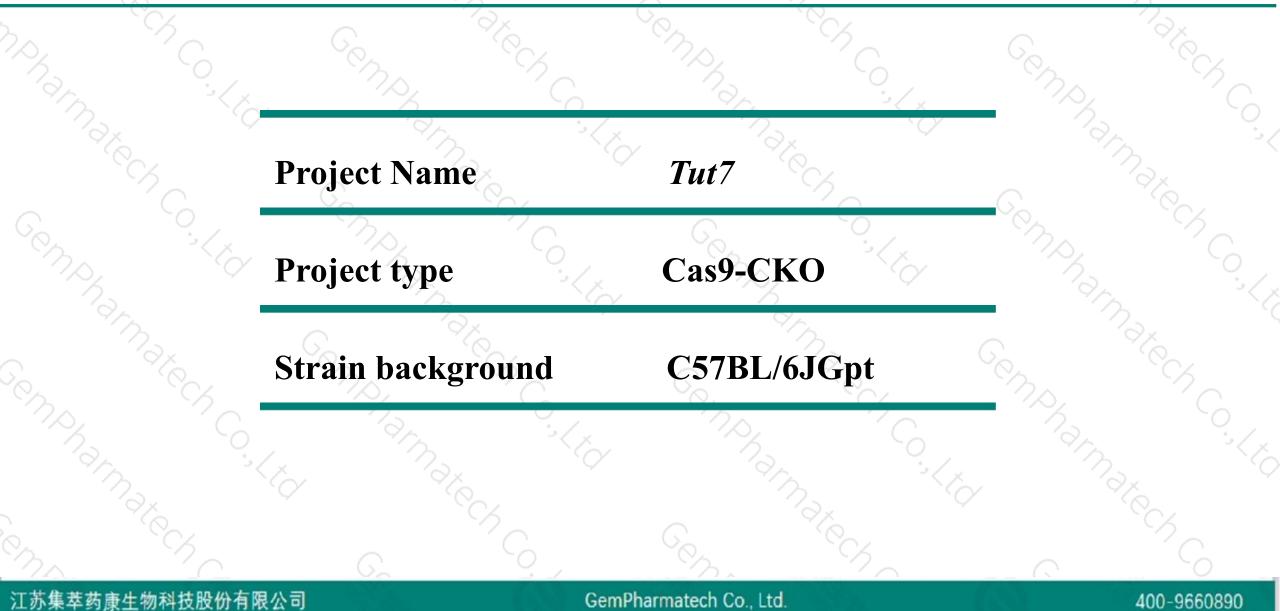
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Design Date: 2020-5-11

Project Overview

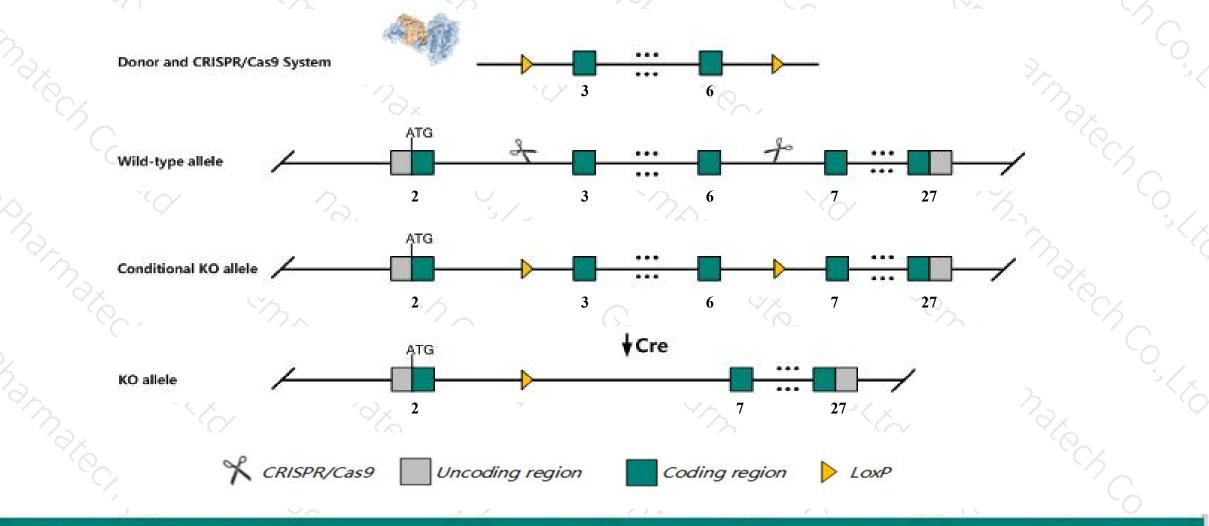




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Tut7* gene. The schematic diagram is as follows:



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The *Tut7* gene has 10 transcripts. According to the structure of *Tut7* gene, exon3-exon6 of *Tut7-201* (ENSMUST00000071703.5) transcript is recommended as the knockout region. The region contains 566bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Tut7* gene. The brief process is as follows:CRISPR/Cas9 system 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 will be 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.



- According to the existing MGI data, mice homozygous for conditional alleles of tut4 and tut7 activated in oocytes exhibit female infertility due to abnormal female meiosis i.
- The *Tut7* gene is located on the Chr13. 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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Tut7 terminal uridylyl transferase 7 [Mus musculus (house mouse)]

Gene ID: 214290, updated on 13-Mar-2020

Summary

Official Symbol	Tut7 provided by MGI
Official Full Name	terminal uridylyl transferase 7 provided by <u>MGI</u>
Primary source	MGI:MGI:2387179
See related	Ensembl:ENSMUSG0000035248
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	6030448M23Rik, AA420405, Tent3b, Zcchc6
Expression	Ubiquitous expression in liver E14 (RPKM 11.4), liver E14.5 (RPKM 9.7) and 28 other tissues See more
Orthologs	human all

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Transcript information (Ensembl)



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

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Name 🔺	Transcript ID	bp 🖕	Protein 🖕	Biotype 🍦	CCDS 🍦	UniProt 🝦	Flags
Tut7-201	ENSMUST0000071703.5	5869	<u>1474aa</u>	Protein coding	<u>CCDS36689</u> &	E9PUA2 &	TSL:1 GENCODE basic APPRIS P1
Tut7-202	ENSMUST00000224320.1	4337	No protein	Retained intron	15	(7)	
Tut7-203	ENSMUST00000224480.1	4186	<u>1089aa</u>	Protein coding	17	<u>A0A286YDD7</u> &	CDS 5' incomplete
Tut7-204	ENSMUST00000224845.1	2452	No protein	Retained intron	17	1.72	
Tut7-205	ENSMUST00000225179.1	619	<u>149aa</u>	Protein coding		<u>A0A286YDR9</u> 교	CDS 3' incomplete
Tut7-206	ENSMUST00000225241.1	1056	<u>148aa</u>	Nonsense mediated decay	5	A0A286YDH2&	CDS 5' incomplete
Tut7-207	ENSMUST00000225576.1	2499	<u>210aa</u>	Protein coding	5	A0A286YD79	GENCODE basic
Tut7-208	ENSMUST00000225607.1	757	No protein	Processed transcript	5	853	
Tut7-209	ENSMUST00000225944.1	2095	No protein	Retained intron	15	87.8	-
Tut7-210	ENSMUST00000225987.1	435	<u>90aa</u>	Protein coding		A0A286YCY9@	CDS 3' incomplete

The strategy is based on the design of *Tut7-201* transcript, The transcription is shown below

< Tut7-201 protein coding

Reverse strand -

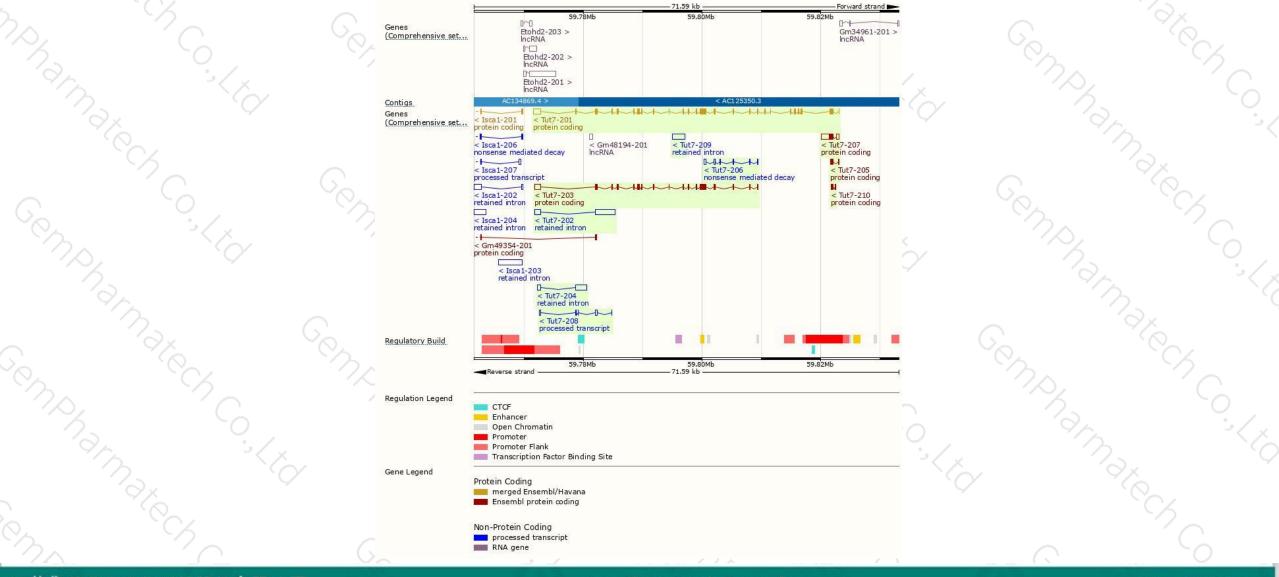
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Genomic location distribution



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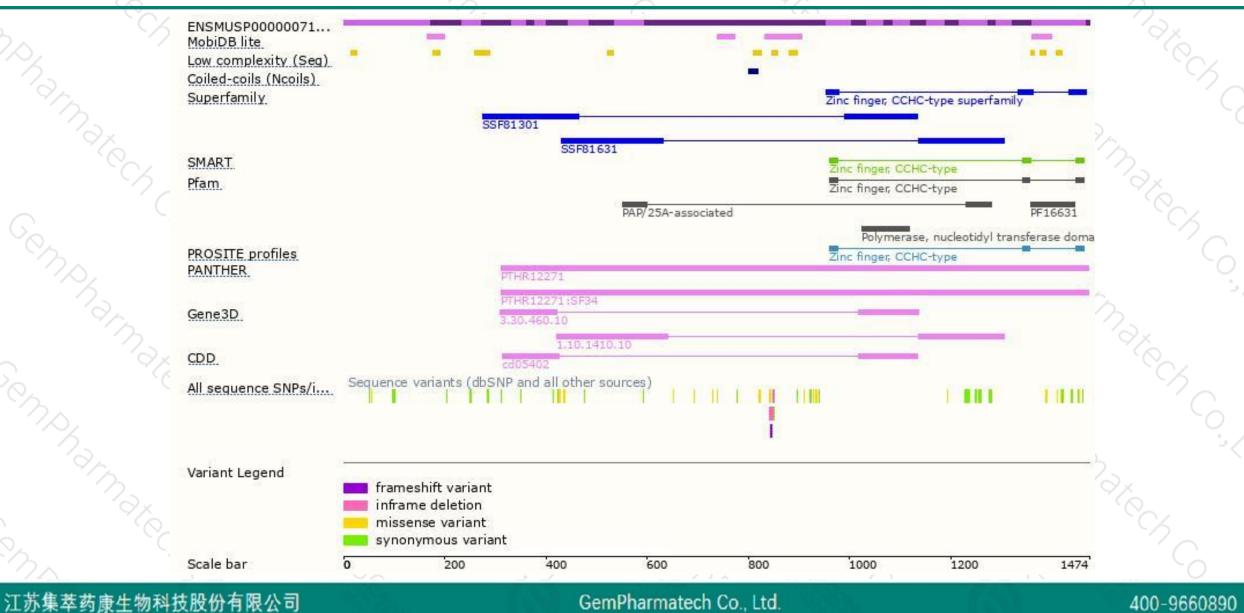


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Protein domain







If you have any questions, you are welcome to inquire. Tel: 400-9660890



