

# Tsks Cas9-CKO Strategy

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Reviewer: Daohua Xu

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## **Project Overview**



Project Name Tsks

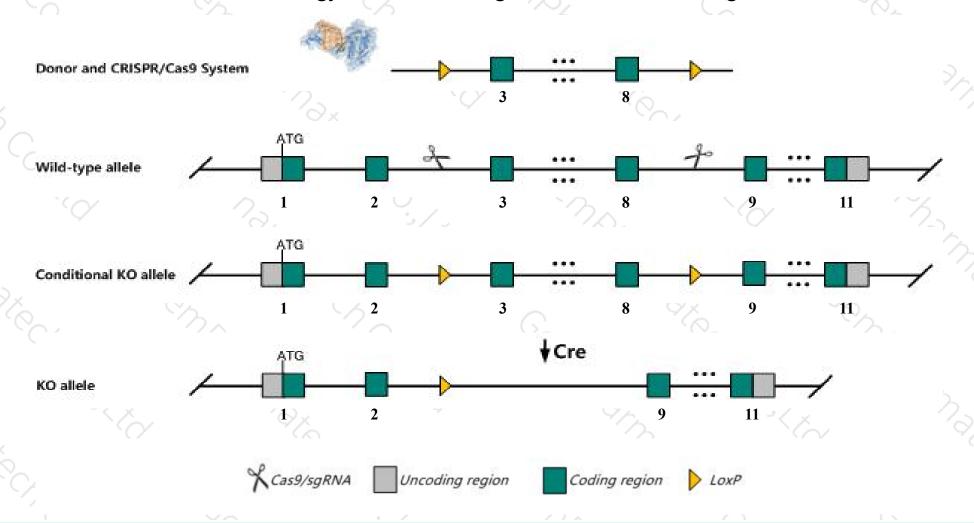
Project type Cas9-CKO

Strain background C57BL/6JGpt

## Conditional Knockout strategy



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



### Technical routes



- ➤ The *Tsks* gene has 4 transcripts. According to the structure of *Tsks* gene, exon3-exon8 of *Tsks*
  202(ENSMUST00000120929.8) transcript is recommended as the knockout region. The region contains 947bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Tsks* 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**



- > The *Tsks* gene is located on the Chr7. 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)



#### Tsks testis-specific serine kinase substrate [ Mus musculus (house mouse) ]

Gene ID: 22116, updated on 25-Sep-2020

#### Summary

↑ ?

Official Symbol Tsks provided by MGI

Official Full Name testis-specific serine kinase substrate provided by MGI

Primary source MGI:MGI:1347560

See related Ensembl: ENSMUSG00000059891

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 Ts; Tss; Stk22; Tssks1; Stk22s1

Summary This gene is a member of a small family of testis-specific serine/threonine kinases. This gene may play a role in testicular

physiology or spermiogenesis. Its expression is restricted to the testis during the last stages of spermatid maturation. Alternative

splicing results in multiple transcript variants encoding distinct proteins. [provided by RefSeq, Jul 2008]

Expression Restricted expression toward testis adult (RPKM 199.6) See more

Orthologs human all

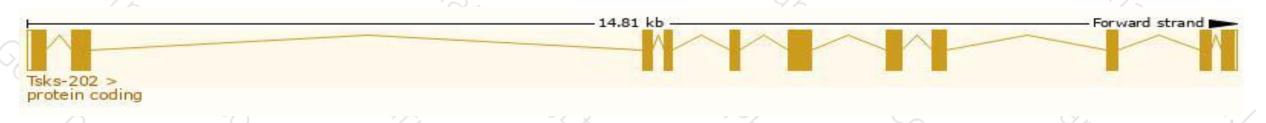
## Transcript information (Ensembl)



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

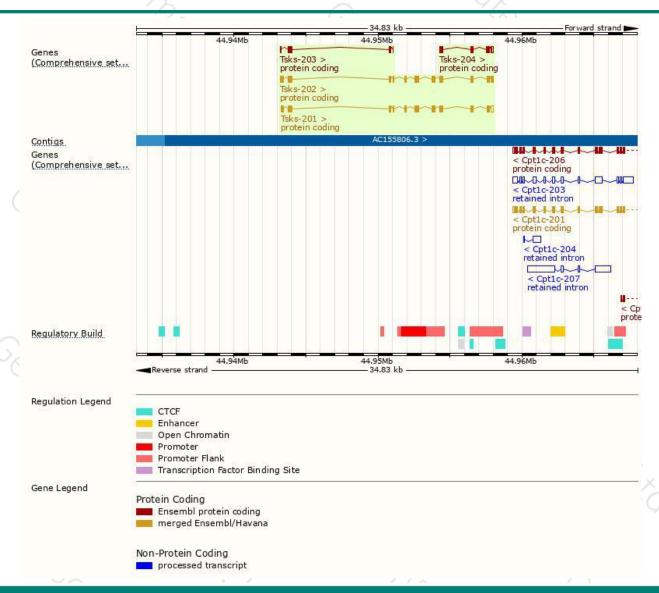
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tsks-202	ENSMUST00000120929.8	1843	585aa	Protein coding	CCDS52240	O54887	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT2
Tsks-201	ENSMUST00000080233.8	1806	<u>525aa</u>	Protein coding	CCDS39946	Q147V4	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Tsks-204	ENSMUST00000208475.1	651	<u>185aa</u>	Protein coding	-	A0A140LIA8	CDS 5' incomplete TSL:5
Tsks-203	ENSMUST00000207719.1	388	<u>109aa</u>	Protein coding	2	A0A140LJ33	CDS 3' incomplete TSL:5

The strategy is based on the design of *Tsks-202* transcript, the transcription is shown below:



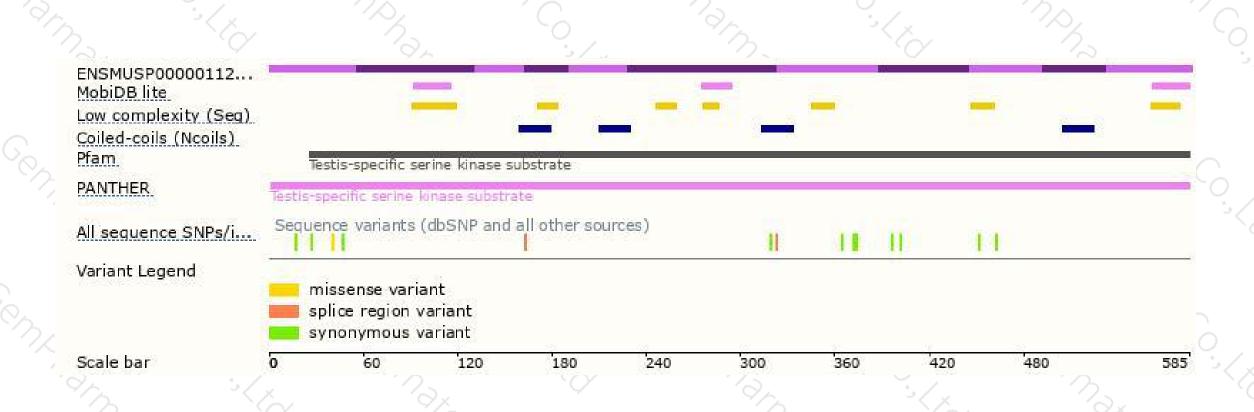
### Genomic location distribution





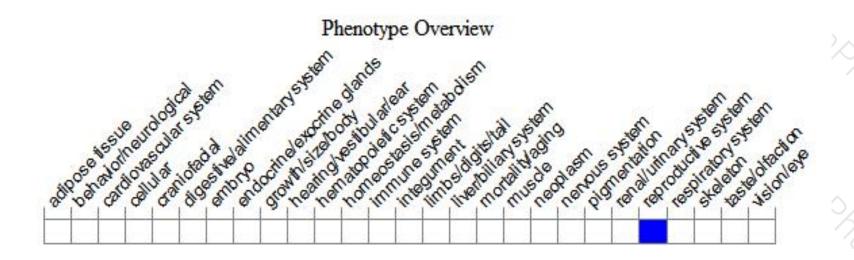
### Protein domain





## Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).



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

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