

Tgds Cas9-CKO Strategy

Designer: Yun Li

Reviewer: Shuang Zhang

Design Date: 2021-3-30

Project Overview

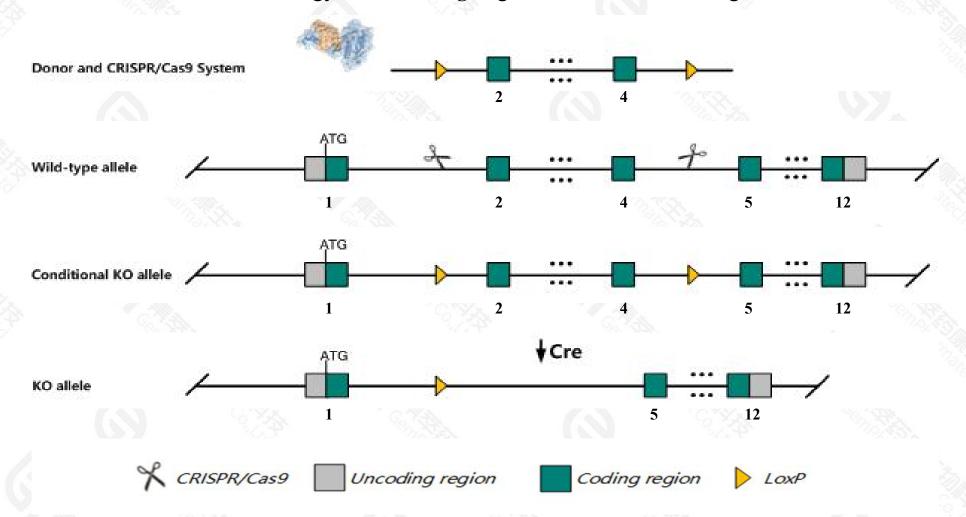


Project Name	Tgds
Project type	Cas9-CKO
Strain background	C57BL/6JGpt

Conditional Knockout strategy



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



Technical routes



- The *Tgds* gene has 4 transcripts. According to the structure of *Tgds* gene, exon2-exon4 of *Tgds*201(ENSMUST00000022727.10) transcript is recommended as the knockout region. The region contains 227bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Tgds* 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.

Notice



- > According to the existing MGI data, mice homozygous for an ENU-induced allele exhibit micrognathia and cleft palate.
- > The *Tgds* gene is located on the Chr14. 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)



Tgds TDP-glucose 4,6-dehydratase [Mus musculus (house mouse)]

Gene ID: 76355, updated on 25-Sep-2020

Summary

☆ ?

Official Symbol Tgds provided by MGI

Official Full Name TDP-glucose 4,6-dehydratase provided by MGI

Primary source MGI:MGI:1923605

See related Ensembl:ENSMUSG00000022130

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 2610017J16Rik, 2610025M23Rik, Al648925

Expression Ubiquitous expression in placenta adult (RPKM 12.4), limb E14.5 (RPKM 9.1) and 28 other tissuesSee more

Orthologs <u>human all</u>

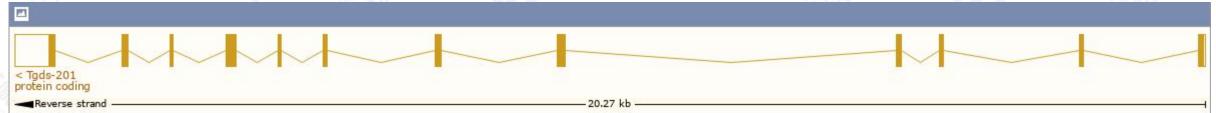
Transcript information (Ensembl)



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

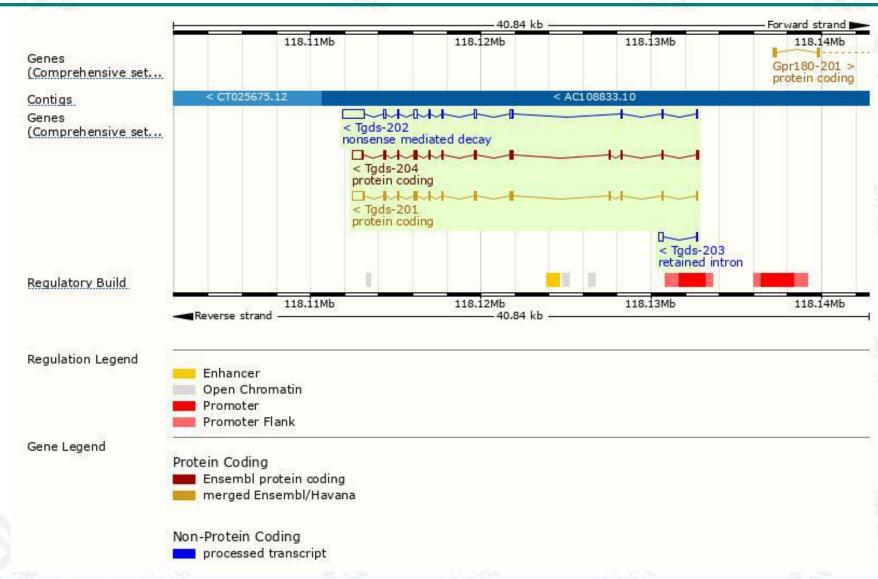
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tgds-201	ENSMUST00000022727.10	1682	<u>355aa</u>	Protein coding	CCDS27332		TSL:1 , GENCODE basic , APPRIS P1
Tgds-204	ENSMUST00000228543.2	1594	<u>343aa</u>	Protein coding	-3		GENCODE basic ,
Tgds-202	ENSMUST00000227350.2	2153	<u>95aa</u>	Nonsense mediated decay	5		
Tgds-203	ENSMUST00000228085.2	350	No protein	Retained intron	-1		

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



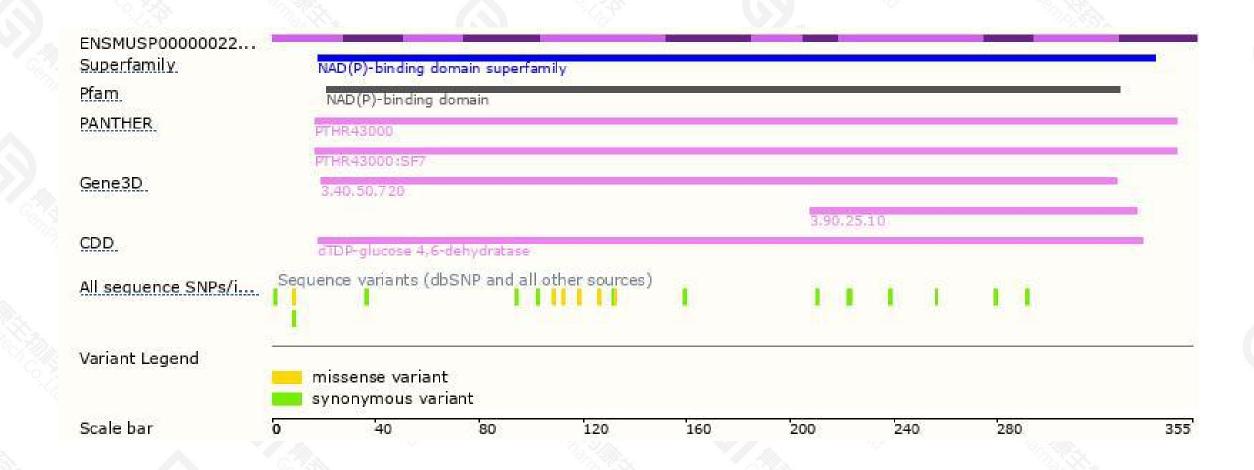
Genomic location distribution





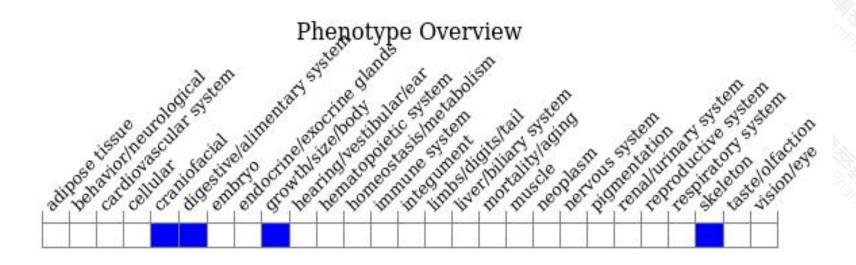
Protein domain





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 an ENU-induced allele exhibit micrognathia and cleft palate.



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

Tel: 400-9660890





